INHIBITORS OF SERINE PROTEASES FOR THE TREATMENT OF HCV INFECTIONS

CROSS-REFERENCE

[001] This application claim priority to U.S. Patent Application No. 11/711,845, filed on February 27, 2007, which is hereby incorporated in its entirety by reference.

FIELD OF THE INVENTION

[002] The present invention relates to compounds that inhibit serine protease activity, particularly the activity of hepatitis C virus NS3-NS4A protease. As such, they act by interfering with the life cycle of the hepatitis C virus and are also useful as antiviral agents. The invention further relates to compositions comprising these compounds either for ex vivo use or for administration to a patient suffering from HCV infection. The invention also relates to methods of treating an HCV infection in a patient by administering a composition comprising a compound of this invention.

BACKGROUND OF THE INVENTION

[003] Infection by hepatitis C virus ("HCV") is a compelling human medical problem. HCV is recognized as the causative agent for most cases of non-A, non-B hepatitis, with an estimated human sero-prevalence of 3% globally [A. Alberti et al., "Natural History of Hepatitis C," J. Hepatology, 31 (Suppl. 1), pp. 17-24 (1999)]. Nearly four million individuals may be infected in the United States alone [M.J. Alter et al., "The Epidemiology of Viral Hepatitis in the United States, Gastroenterol. Clin. North Am., 23, pp. 437-455 (1994); M. J. Alter "Hepatitis C Virus Infection in the United States," J. Hepatology, 31., (Suppl. 1), pp. 88-91 (1999)].

[004] Upon first exposure to HCV only about 20% of infected individuals develop acute clinical hepatitis while others appear to resolve the infection spontaneously. In almost 70% of instances, however; the virus establishes a chronic infection that persists for decades [S. Iwarson, "The Natural Course of Chronic Hepatitis," FEMS Microbiology Reviews, 14, pp. 201-204 (1994); D. Lavanchy, "Global Surveillance and Control of Hepatitis C," J. Viral Hepatitis, 6, pp. 35-47 (1999)]. This usually results in recurrent and progressively worsening liver inflammation, which often leads to more severe disease states such as cirrhosis and hepatocellular carcinoma [M.C. Kew, "Hepatitis C and Hepatocellular Carcinoma", FEMS Microbiology Reviews, 14, pp. 211-220 (1994); I. Saito et. al., "Hepatitis C Virus Infection is Associated with the Development of Hepatocellular Carcinoma," Proc. Natl. Acad. Sci. USA, 87, pp. 6547-6549 (1990)]. Unfortunately, there are no broadly effective treatments for the debilitating progression of chronic HCV.

[005] The HCV genome encodes a polyprotein of 3010-3033 amino acids [Q.L. Choo, et. al., "Genetic Organization and Diversity of the Hepatitis C Virus." Proc. Natl. Acad. Sci. USA, 88, pp. 2451-2455 (1991); N. Kato et al., "Molecular Cloning of the Human Hepatitis C Virus Genome From Japanese Patients with Non-A, Non-B Hepatitis," Proc. Natl. Acad. Sci. USA, 87, pp. 9524-9528 (1990); A. Takamizawa et. al., "Structure and Organization of the Hepatitis C Virus Genome Isolated From Human Carriers," J. Virol., 65, pp. 1105-1113 (1991)]. The HCV nonstructural (NS) proteins are presumed to provide the essential catalytic machinery for viral replication. The NS proteins are derived by proteolytic cleavage of the polyprotein [R. Bartenschlager et. al., "Nonstructural Protein 3 of the Hepatitis C Virus Encodes a Serine-Type Proteinase Required for Cleavage at the NS3/4 and NS4/5 Junctions," J. Virol., 67, pp. 3835-3844 (1993); A. Grakoui et. al., "Characterization of the Hepatitis C Virus-Encoded Serine Proteinase: Determination of Proteinase-Dependent Polyprotein Cleavage Sites," J. Virol., 67, pp. 2832-2843 (1993); A. Grakoui et. al., "Expression and Identification of Hepatitis C Virus Polyprotein Cleavage Products," J. Virol., 67, pp. 1385-1395 (1993); L. Tomei et. al., "NS3 is a serine protease required for processing of hepatitis C virus polyprotein", J. Virol., 67, pp. 4017-4026 (1993)].

[006] The HCV NS protein 3 (NS3) contains a serine protease activity that helps process the majority of the viral enzymes, and is thus considered essential for viral replication and infectivity. It is known that mutations in the yellow fever virus NS3 protease decrease viral infectivity [Chambers, T.J. et. al., "Evidence that the N-terminal Domain of Nonstructural Protein NS3 From Yellow Fever Virus is a Serine Protease Responsible for Site-Specific Cleavages in the Viral Polyprotein", Proc. Natl. Acad. Sci. USA, 87, pp. 8898-8902 (1990)]. The first 181 amino acids of NS3 (residues 1027-1207 of the viral polyprotein) have been shown to contain the serine protease domain of NS3 that processes all four downstream sites of the HCV polyprotein [C. Lin et al., "Hepatitis C Virus NS3 Serine Proteinase: Trans-Cleavage Requirements and Processing Kinetics", J. Virol., 68, pp. 8147-8157 (1994)].

[007] The HCV NS3 serine protease and its associated cofactor, NS4A, helps process all of the viral enzymes, and is thus considered essential for viral replication. This processing appears to be analogous to that carried out by the human immunodeficiency virus aspartyl protease, which is also involved in viral enzyme processing. HIV protease inhibitors, which inhibit viral protein processing, are potent antiviral agents in man indicating that interrupting this stage of the viral life cycle results in therapeutically active agents. Consequently HCV NS3 serine protease is also an attractive target for drug discovery.

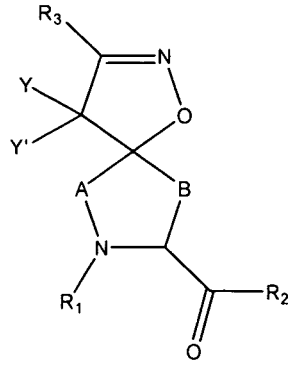
[008] There are not currently any satisfactory anti-HCV agents or treatments. Until

recently, the only established therapy for HCV disease was interferon treatment. However, interferons have significant side effects [M. A. Walker et al., "Hepatitis C Virus: An Overview of Current Approaches and Progress," DDT, 4, pp. 518-29 (1999); D. Moradpour et al., "Current and Evolving Therapies for Hepatitis C," Eur. J. Gastroenterol. Hepatol., 11, pp. 1199-1202 (1999); H. L. A. Janssen et al. "Suicide Associated with Alfa-Interferon Therapy for Chronic Viral Hepatitis," J. Hepatol., 21, pp. 241-243 (1994); P.F. Renault et al., "Side Effects of Alpha Interferon," Seminars in Liver Disease, 9, pp. 273-277. (1989)] and induce long term remission in only a fraction (≈ 25%) of cases [O. Weiland, "Interferon Therapy in Chronic Hepatitis C Virus Infection", FEMS Microbiol. Rev., 14, pp. 279-288 (1994)]. Recent introductions of the pegylated forms of interferon (PEG-INTRON® and PEGASYS®) and the combination therapy of ribavirin and pegylated interferon (REBETROL®) have resulted in only modest improvements in remission rates and only partial reductions in side effects. Moreover, the prospects for effective anti-HCV vaccines remain uncertain.

[009] Thus, there is a need for more effective anti-HCV therapies. Such inhibitors would have therapeutic potential as protease inhibitors, particularly as serine protease inhibitors, and more particularly as HCV NS3 protease inhibitors. Specifically, such compounds may be useful as antiviral agents, particularly as anti-HCV agents.

SUMMARY OF THE INVENTION

[010] This invention relates to compounds of formula (I)



(I)

or pharmaceutically acceptable salts thereof wherein,

each A is $-(CX_1X_2)_a-$;

each B is $-(CX_1X_2)_b-$;

each X_1 is independently hydrogen, halo, amino, sulfanyl, optionally substituted aliphatic, optionally substituted aryl, or $-O-X_{1A}$;

each X_2 is independently hydrogen, halo, amino, sulfanyl, optionally substituted aliphatic, optionally substituted aryl, or -O-X_{1B};

X_{1A} and X_{1B} are each independently an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or

X_1 and X_2 together form an oxo group;

each R_1 is -Z^AR_4, wherein each Z^A is independently a bond or an optionally substituted aliphatic wherein up to three carbon units of Z^A are optionally and independently replaced by -C(O)-, -C(S)-, -C(O)NR^A-, -C(O)NR^ANR^A-, -C(O)O-, -NR^AC(O)O-, -O-, -NR^AC(O)NR^A-, -NR^ANR^A-, -S-, -SO-, -SO_2-, -NR^A-, -SO_2NR^A-, or -$NR^ASO_2NR^A$- provided that -NR^ANR^A-, -NR^AC(O)NR^A-, or -$NR^ASO_2NR^A$- is not directly bound to the nitrogen ring atom of formula (I);

each R_4 is independently R^A, halo, -OH, -CN, -NO_2, -NH_2, or -OCF_3;

each R^A is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_2 is -Z^BR_5, wherein each Z^B is independently a bond or an optionally substituted aliphatic wherein up to three carbon units of Z^B are optionally and independently replaced by -C(O)-, -C(S)-, -C(O)NR^B-, -C(O)NR^BNR^B-, -C(O)O-, -NR^BC(O)O-, -NR^BC(O)NR^B-, -NR^BNR^B-, -S-, -SO-, -SO_2-, -NR^B-, -SO_2NR^B-, or -$NR^BSO_2NR^B$-, provided that -SO-, -SO_2-, or -SO_2NR^B- is not directly bound to the carbonyl of formula (I);

each R_5 is independently R^B, halo, -OH, -CN, -NO_2, -NH_2, alkoxy, or haloalkoxy;

Each R^B is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or,

R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted heterocycloaliphatic ring;

each R_3 is an optionally substituted aliphatic, amino, sulfonyl, sulfanyl, sulfinyl, sulfonamide, sulfamide, sulfo, -O-R_{3A}, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_{3A} is independently an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

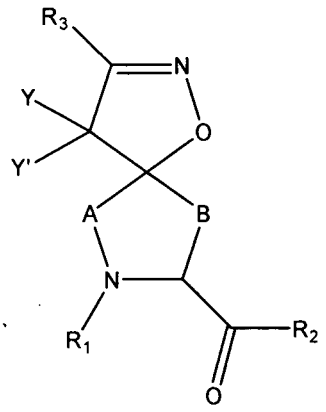
each of Y and Y' is independently $-Z^DR_7$, wherein each Z^D is independently a bond or an optionally substituted straight or branched C_{1-6} aliphatic chain wherein up to two carbon units of Z^D are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^D-$, $-C(O)NR^DNR^D-$, -C(O)O-, $-NR^DC(O)O-$, -O-, $-NR^DC(O)NR^D-$, $-NR^DNR^D-$, -S-, -SO-, $-SO_2-$, $-NR^D-$, $-SO_2NR^D-$, $-NR^DSO_2-$, or $-NR^DSO_2NR^D-$, or Y and Y' together form =O or =S;

each R_7 is independently R^D, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$;

each R^D is independently hydrogen, or optionally substituted aryl; and

each of **a** and **b** is independently 0, 1, 2, or 3, provided that the sum of **a** and **b** is 2 or 3.

[011] The invention also relates to compounds of formula (I) below.



(I)

In this formula,

each A is $-CH_2-$;

each B is $-CH_2-$;

each R_1 is $-Z^AR_4$, wherein each Z^A is -C(O)-, -C(S)-, $-C(O)NR^A-$, $-C(O)NR^ANR^A-$, -C(O)O-, $-NR^AC(O)O-$, -O-, -S-, -SO-, $-SO_2-$, $-NR^A-$, or $-SO_2NR^A-$;

each R_4 is independently R^A, halo, -OH, -CN, $-NO_2$, $-NH_2$, alkoxy, or haloalkoxy;

each R^A is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_2 is $-Z^BR_5$, wherein each Z^B is independently a bond or an optionally substituted aliphatic wherein up to four carbon units of Z^B are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^B-$, $-C(O)NR^BNR^B-$, -C(O)O-, $-C(O)C(O)-NR^B-$, $-NR^BC(O)O-$, $-NR^BC(O)NR^B-$, $-NR^BNR^B-$, -S-, -SO-, $-SO_2-$, $-NR^B-$, $-SO_2NR^B-$, or $-NR^BSO_2NR^B-$, provided that -SO-, $-SO_2-$, or $-SO_2NR^B-$ is not directly bound to the carbonyl in formula (I);

each R_5 is independently R^B, halo, -OH, -CN, $-NO_2$, $-NH_2$, alkoxy, or haloalkoxy;

each R^B is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or,

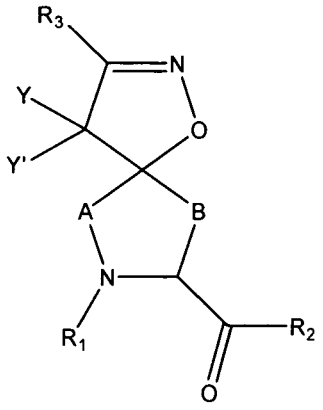
R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted heterocycloaliphatic ring;

each R_3 is an optionally substituted aliphatic, amino, sulfonyl, sulfanyl, sulfinyl, sulfonamide, sulfamide, sulfo, $-O-R_{3A}$, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_{3A} is independently an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; and

each of Y and Y' is H.

[012] Still, the invention also provides compounds of Formula (I)



(I)

or a pharmaceutically acceptable salt thereof. In formula (I),

each A is $-CH_2-$;

each B is $-CH_2-$;

each R_1 is $-Z^AR_4$, wherein Z^A is -C(O)- and R_4 is a substituted alkyl;

each R_2 is $-Z^BR_5$, wherein each Z^B is independently a bond or an optionally substituted aliphatic wherein up to four carbon units of Z^B are optionally and independently replaced by -C(O)-, $-C(O)NR^B-$, $-C(O)C(O)-NR^B-$, or $-NR^B-$;

each R_5 is independently R^B, halo, -OH, -CN, $-NO_2$, $-NH_2$, alkoxy, or haloalkoxy;

each R^B is independently hydrogen, an optionally substituted aliphatic, or an optionally substituted cycloaliphatic; or

R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted 6-membered heterocycloaliphatic ring; each R_3 is an optionally substituted amino, -O-R_{3A}, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; each R_{3A} is independently an optionally substituted aryl or an optionally substituted heteroaryl; and each of Y and Y' is H.

In some aspects the invention features compound numbers 595 through 1044 described below in Table 1.

[013] In some aspects, the invention features a pharmaceutical composition comprising a compound of formula (I) or a pharmaceutically acceptable salt thereof in an amount effective to inhibit a serine protease; and an acceptable carrier, adjuvant or vehicle. The composition may include an additional agent selected from an immunomodulatory agent; an antiviral agent; a second inhibitor of HCV protease; an inhibitor of another target in the HCV life cycle; and a cytochrome P-450 inhibitor; or combinations thereof. The immunomodulatory agent is α–, β–, or γ–interferon or thymosin; said antiviral agent is ribavirin, amantadine, or telbivudine; or said inhibitor of another target in the HCV life cycle is an inhibitor of HCV helicase, polymerase, or metalloprotease. Cytochrome P-450 inhibitor may be ritonavir.

[014] In other aspects, a method of inhibiting the activity of a serine protease comprising the step of contacting said serine protease with a compound of formula (I). The serine protease may be an HCV NS3 protease. The methods also include treating an HCV infection in a patient by administering a compound of formula (I). The method may also include administering to said patient an additional agent selected from an immunomodulatory agent; an antiviral agent; a second inhibitor of HCV protease; an inhibitor of another target in the HCV life cycle; or combinations thereof; wherein said additional agent is administered to said patient in the same dosage form as the serine protease inhibitor or as a separate dosage form. The immunomodulatory agent is α–, β–, or γ–interferon or thymosin; said antiviral agent is ribavarin or amantadine; or said inhibitor of another target in the HCV life cycle is an inhibitor of HCV helicase, polymerase, or metalloprotease.

[015] In still other aspects, a method of eliminating or reducing HCV contamination of a biological sample or medical or laboratory equipment includes the step of contacting said biological sample or medical or laboratory equipment with a compound of formula (I). The sample or equipment may be selected from blood, other body fluids, biological tissue, a surgical instrument, a surgical garment, a laboratory instrument, a laboratory garment, a blood or other body fluid collection apparatus; a blood or other body fluid storage material.

[016] The compounds of the invention, as described herein, also exhibit advantageous PK

properties and/or increased potency.

[017] The invention also relates to compositions that comprise the above compounds and the use thereof; methods of preparing compounds of formula (I), and methods of assaying compounds for serine protease activity. Such compositions may be used to pre-treat devices that are to be inserted into a patient, to treat biological samples, and for direct administration to a patient. In each case, the composition will be used to lessen the risk of or the severity of the HCV infection.

DEFINITIONS

[018] For purposes of this invention, the chemical elements are identified in accordance with the Periodic Table of the Elements, CAS version, Handbook of Chemistry and Physics, 75th Ed. Additionally, general principles of organic chemistry are described in "Organic Chemistry", Thomas Sorrell, University Science Books, Sausalito: 1999, and "Advanced Organic Chemistry", 5th Ed., Ed.: Smith, M.B. and March, J., John Wiley & Sons, New York: 2001, the entire contents of which are hereby incorporated by reference.

[019] As described herein, compounds of the invention may optionally be substituted with one or more substituents, such as are illustrated generally above, or as exemplified by particular classes, subclasses, and species of the invention.

[020] As used herein the term "aliphatic" encompasses the terms alkyl, alkenyl, alkynyl, each of which being optionally substituted as set forth below.

[021] As used herein, an "alkyl" group refers to a saturated aliphatic hydrocarbon group containing 1 to 12 (e.g., 1 to 10, 1 to 8, 1 to 6, or 1 to 4) carbon atoms. An alkyl group can be straight or branched. Examples of alkyl groups include, but are not limited to, methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, n-pentyl, n-heptyl, or 2-ethylhexyl. An alkyl group can be substituted (i.e., optionally substituted) with one or more substituents such as halo, phospho, cycloaliphatic (e.g., cycloalkyl or cycloalkenyl), heterocycloaliphatic (e.g., heterocycloalkyl or heterocycloalkenyl), aryl, heteroaryl, alkoxy, aroyl, heteroaroyl, acyl (e.g., (aliphatic)carbonyl, (cycloaliphatic)carbonyl, or (heterocycloaliphatic)carbonyl), nitro, cyano, amido (e.g., (cycloalkylalkyl)carbonylamino, arylcarbonylamino, aralkylcarbonylamino, (heterocycloalkyl)carbonylamino, (heterocycloalkylalkyl)carbonylamino, heteroarylcarbonylamino, heteroaralkylcarbonylamino alkylaminocarbonyl, cycloalkylaminocarbonyl, heterocycloalkylaminocarbonyl, arylaminocarbonyl, or heteroarylaminocarbonyl), amino (e.g., aliphaticamino, cycloaliphaticamino, or heterocycloaliphaticamino), sulfonyl (e.g., aliphatic-SO_2-), sulfinyl, sulfanyl, sulfoxy, urea, thiourea, sulfamoyl, sulfamide, oxo,

carboxy, carbamoyl, cycloaliphaticoxy, heterocycloaliphaticoxy, aryloxy, heteroaryloxy, aralkyloxy, heteroarylalkoxy, alkoxycarbonyl, alkylcarbonyloxy, or hydroxy. Without limitation, some examples of substituted alkyls include carboxyalkyl (such as HOOC-alkyl, alkoxycarbonylalkyl, and alkylcarbonyloxyalkyl), cyanoalkyl, hydroxyalkyl, alkoxyalkyl, acylalkyl, aralkyl, (alkoxyaryl)alkyl, (sulfonylamino)alkyl (such as (alkyl-SO_2-amino)alkyl), aminoalkyl, amidoalkyl, (cycloaliphatic)alkyl, or haloalkyl.

[022] As used herein, an "alkenyl" group refers to an aliphatic carbon group that contains 2 to 12 (e.g., 2 to 8, 2 to 6, or 2 to 4) carbon atoms and at least one double bond. Like an alkyl group, an alkenyl group can be straight or branched. Examples of an alkenyl group include, but are not limited to, allyl, isoprenyl, 2-butenyl, and 2-hexenyl. An alkenyl group can be optionally substituted with one or more substituents such as halo, phospho, cycloaliphatic (e.g., cycloalkyl or cycloalkenyl), heterocycloaliphatic (e.g., heterocycloalkyl or heterocycloalkenyl), aryl, heteroaryl, alkoxy, aroyl, heteroaroyl, acyl (e.g., (aliphatic)carbonyl, (cycloaliphatic)carbonyl, or (heterocycloaliphatic)carbonyl), nitro, cyano, amido (e.g., (cycloalkylalkyl)carbonylamino, arylcarbonylamino, aralkylcarbonylamino, (heterocycloalkyl)carbonylamino, (heterocycloalkylalkyl)carbonylamino, heteroarylcarbonylamino, heteroaralkylcarbonylamino alkylaminocarbonyl, cycloalkylaminocarbonyl, heterocycloalkylaminocarbonyl, arylaminocarbonyl, or heteroarylaminocarbonyl), amino (e.g., aliphaticamino, cycloaliphaticamino, heterocycloaliphaticamino, or aliphaticsulfonylamino), sulfonyl (e.g., alkyl-SO_2-, cycloaliphatic-SO_2-, or aryl-SO_2-), sulfinyl, sulfanyl, sulfoxy, urea, thiourea, sulfamoyl, sulfamide, oxo, carboxy, carbamoyl, cycloaliphaticoxy, heterocycloaliphaticoxy, aryloxy, heteroaryloxy, aralkyloxy, heteroaralkoxy, alkoxycarbonyl, alkylcarbonyloxy, or hydroxy. Without limitation, some examples of substituted alkenyls include cyanoalkenyl, alkoxyalkenyl, acylalkenyl, hydroxyalkenyl, aralkenyl, (alkoxyaryl)alkenyl, (sulfonylamino)alkenyl (such as (alkyl-SO_2-amino)alkenyl), aminoalkenyl, amidoalkenyl, (cycloaliphatic)alkenyl, or haloalkenyl.

[023] As used herein, an "alkynyl" group refers to an aliphatic carbon group that contains 2 to 12 (e.g., 2 to 8, 2 to 6, or 2 to 4) carbon atoms and has at least one triple bond. An alkynyl group can be straight or branched. Examples of an alkynyl group include, but are not limited to, propargyl and butynyl. An alkynyl group can be optionally substituted with one or more substituents such as aroyl, heteroaroyl, alkoxy, cycloalkyloxy, heterocycloalkyloxy, aryloxy, heteroaryloxy, aralkyloxy, nitro, carboxy, cyano, halo, hydroxy, sulfo, mercapto, sulfanyl (e.g., aliphaticsulfanyl or cycloaliphaticsulfanyl), sulfinyl (e.g., aliphaticsulfinyl or

cycloaliphaticsulfinyl), sulfonyl (e.g., aliphatic-SO_2-, aliphaticamino-SO_2-, or cycloaliphatic-SO_2-), amido (e.g., aminocarbonyl, alkylaminocarbonyl, alkylcarbonylamino, cycloalkylaminocarbonyl, heterocycloalkylaminocarbonyl, cycloalkylcarbonylamino, arylaminocarbonyl, arylcarbonylamino, aralkylcarbonylamino, (heterocycloalkyl)carbonylamino, (cycloalkylalkyl)carbonylamino, heteroaralkylcarbonylamino, heteroarylcarbonylamino or heteroarylaminocarbonyl), urea, thiourea, sulfamoyl, sulfamide, alkoxycarbonyl, alkylcarbonyloxy, cycloaliphatic, heterocycloaliphatic, aryl, heteroaryl, acyl (e.g., (cycloaliphatic)carbonyl or (heterocycloaliphatic)carbonyl), amino (e.g., aliphaticamino), sulfoxy, oxo, carboxy, carbamoyl, (cycloaliphatic)oxy, (heterocycloaliphatic)oxy, or (heteroaryl)alkoxy.

[024] As used herein, an "amido" encompasses both "aminocarbonyl" and "carbonylamino". These terms when used alone or in connection with another group refers to an amido group such as $-N(R^X)-C(O)-R^Y$ or $-C(O)-N(R^X)_2$, when used terminally, and $-C(O)-N(R^X)-$ or $-N(R^X)-C(O)-$ when used internally, wherein R^X and R^Y are defined below. Examples of amido groups include alkylamido (such as alkylcarbonylamino or alkylaminocarbonyl), (heterocycloaliphatic)amido, (heteroaralkyl)amido, (heteroaryl)amido, (heterocycloalkyl)alkylamido, arylamido, aralkylamido, (cycloalkyl)alkylamido, or cycloalkylamido.

[025] As used herein, an "amino" group refers to $-NR^XR^Y$ wherein each of R^X and R^Y is independently hydrogen, aliphatic, cycloaliphatic, (cycloaliphatic)aliphatic, aryl, araliphatic, heterocycloaliphatic, (heterocycloaliphatic)aliphatic, heteroaryl, carboxy, sulfanyl, sulfinyl, sulfonyl, (aliphatic)carbonyl, (cycloaliphatic)carbonyl, ((cycloaliphatic)aliphatic)carbonyl, arylcarbonyl, (araliphatic)carbonyl, (heterocycloaliphatic)carbonyl, ((heterocycloaliphatic)aliphatic)carbonyl, (heteroaryl)carbonyl, or (heteroaraliphatic)carbonyl, each of which being defined herein and being optionally substituted. Examples of amino groups include alkylamino, dialkylamino, or arylamino. When the term "amino" is not the terminal group (e.g., alkylcarbonylamino), it is represented by $-NR^X-$. R^X has the same meaning as defined above.

[026] As used herein, an "aryl" group used alone or as part of a larger moiety as in "aralkyl", "aralkoxy", or "aryloxyalkyl" refers to monocyclic (e.g., phenyl); bicyclic (e.g., indenyl, naphthalenyl, tetrahydronaphthyl, tetrahydroindenyl); and tricyclic (e.g., fluorenyl tetrahydrofluorenyl, or tetrahydroanthracenyl, anthracenyl) ring systems in which the monocyclic ring system is aromatic or at least one of the rings in a bicyclic or tricyclic ring system is aromatic. The bicyclic and tricyclic groups include benzofused 2-3 membered

carbocyclic rings. For example, a benzofused group includes phenyl fused with two or more C_{4-8} carbocyclic moieties. An aryl is optionally substituted with one or more substituents including aliphatic [e.g., alkyl, alkenyl, or alkynyl]; cycloaliphatic; (cycloaliphatic)aliphatic; heterocycloaliphatic; (heterocycloaliphatic)aliphatic; aryl; heteroaryl; alkoxy; (cycloaliphatic)oxy; (heterocycloaliphatic)oxy; aryloxy; heteroaryloxy; (araliphatic)oxy; (heteroaraliphatic)oxy; aroyl; heteroaroyl; amino; oxo (on a non-aromatic carbocyclic ring of a benzofused bicyclic or tricyclic aryl); nitro; carboxy; amido; acyl (e.g., (aliphatic)carbonyl, (cycloaliphatic)carbonyl, ((cycloaliphatic)aliphatic)carbonyl, (araliphatic)carbonyl, (heterocycloaliphatic)carbonyl, ((heterocycloaliphatic)aliphatic)carbonyl, or (heteroaraliphatic)carbonyl); sulfonyl (e.g., aliphatic-SO_2- or amino-SO_2-); sulfinyl (e.g., aliphatic-S(O)- or cycloaliphatic-S(O)-); sulfanyl (e.g., aliphatic-S-); cyano; halo; hydroxy; mercapto; sulfoxy; urea; thiourea; sulfamoyl; sulfamide; or carbamoyl. Alternatively, an aryl can be unsubstituted.

[027] Non-limiting examples of substituted aryls include haloaryl (e.g., mono-, di-(such as *p,m*-dihaloaryl), and tri-(such as halo) aryl); (carboxy)aryl (e.g., (alkoxycarbonyl)aryl, ((aralkyl)carbonyloxy)aryl, and (alkoxycarbonyl)aryl); (amido)aryl (e.g., (aminocarbonyl)aryl, (((alkylamino)alkyl)aminocarbonyl)aryl, (alkylcarbonyl)aminoaryl, (arylaminocarbonyl)aryl, and (((heteroaryl)amino)carbonyl)aryl); aminoaryl (e.g., ((alkylsulfonyl)amino)aryl or ((dialkyl)amino)aryl); (cyanoalkyl)aryl; (alkoxy)aryl; (sulfamoyl)aryl [e.g., (aminosulfonyl)aryl]; (alkylsulfonyl)aryl; (cyano)aryl; (hydroxyalkyl)aryl; ((alkoxy)alkyl)aryl; (hydroxy)aryl, ((carboxy)alkyl)aryl; (((dialkyl)amino)alkyl)aryl; (nitroalkyl)aryl; (((alkylsulfonyl)amino)alkyl)aryl; ((heterocycloaliphatic)carbonyl)aryl; ((alkylsulfonyl)alkyl)aryl; (cyanoalkyl)aryl; (hydroxyalkyl)aryl; (alkylcarbonyl)aryl; alkylaryl; (trihaloalkyl)aryl; *p*-amino-*m*-alkoxycarbonylaryl; *p*-amino-*m*-cyanoaryl; *p*-halo-*m*-aminoaryl; or (*m*-(heterocycloaliphatic)-*o*-(alkyl))aryl.

[028] As used herein, an "araliphatic" such as an "aralkyl" group refers to an aliphatic group (e.g., a C_{1-4} alkyl group) that is substituted with an aryl group. "Aliphatic," "alkyl," and "aryl" are defined herein. An example of an araliphatic such as an aralkyl group is benzyl.

[029] As used herein, an "aralkyl" group refers to an alkyl group (e.g., a C_{1-4} alkyl group) that is substituted with an aryl group. Both "alkyl" and "aryl" have been defined above. An example of an aralkyl group is benzyl. An aralkyl is optionally substituted with one or more substituents such as aliphatic (e.g., alkyl, alkenyl, or alkynyl, including carboxyalkyl,

hydroxyalkyl, or haloalkyl such as trifluoromethyl); cycloaliphatic (e.g., cycloalkyl or cycloalkenyl); (cycloalkyl)alkyl; heterocycloalkyl; (heterocycloalkyl)alkyl; aryl; heteroaryl; alkoxy; cycloalkyloxy; heterocycloalkyloxy; aryloxy; heteroaryloxy; aralkyloxy; heteroaralkyloxy; aroyl; heteroaroyl; nitro; carboxy; alkoxycarbonyl; alkylcarbonyloxy; amido (e.g., aminocarbonyl, alkylcarbonylamino, cycloalkylcarbonylamino, (cycloalkylalkyl)carbonylamino, arylcarbonylamino, aralkylcarbonylamino, (heterocycloalkyl)carbonylamino, (heterocycloalkylalkyl)carbonylamino, heteroarylcarbonylamino, or heteroaralkylcarbonylamino); cyano; halo; hydroxy; acyl; mercapto; alkylsulfanyl; sulfoxy; urea; thiourea; sulfamoyl; sulfamide; oxo; or carbamoyl.

[030] As used herein, a "bicyclic ring system" includes 8- to 12- (e.g., 9, 10, or 11) membered structures that form two rings, wherein the two rings have at least one atom in common (e.g., 1 atom or 2 atoms in common). Bicyclic ring systems include bicycloaliphatics (e.g., bicycloalkyl or bicycloalkenyl), bicycloheteroaliphatics, bicyclic aryls, and bicyclic heteroaryls.

[031] As used herein, a "cycloaliphatic" group encompasses a "cycloalkyl" group and a "cycloalkenyl" group, each of which being optionally substituted as set forth below.

[032] As used herein, a "cycloalkyl" group refers to a saturated carbocyclic mono- or bicyclic (fused or bridged) ring of 3-12 (e.g., 5-12) carbon atoms. Examples of cycloalkyl groups include cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, adamantyl, norbornyl, cubyl, octahydro-indenyl, decahydro-naphthyl, spiro[5.5]undecanyl, spiro[2.5]octanyl, bicyclo[3.2.1]octyl, bicyclo[2.2.2]octyl, bicyclo[3.3.1]nonyl, bicyclo[3.3.2.]decyl, bicyclo[2.2.2]octyl, adamantyl, or ((aminocarbonyl)cycloalkyl)cycloalkyl.

[033] A "cycloalkenyl" group, as used herein, refers to a non-aromatic carbocyclic mono- or bicyclic ring of 3 to 12 (e.g., 4 to 8) carbon atoms having one or more double bonds. Examples of cycloalkenyl groups include cyclopentenyl, 1,4-cyclohexa-di-enyl, cycloheptenyl, cyclooctenyl, hexahydro-indenyl, octahydro-naphthyl, cyclohexenyl, cyclopentenyl, spiro[5.5]]undec-3-enyl, spiro[2.5]oct-5-enyl, bicyclo[2.2.2]octenyl, or bicyclo[3.3.1]nonenyl.

[034] A cycloalkyl or cycloalkenyl group can be optionally substituted with one or more substituents such as phosphor; aliphatic (e.g., alkyl, alkenyl, or alkynyl); cycloaliphatic; (cycloaliphatic) aliphatic; heterocycloaliphatic; (heterocycloaliphatic) aliphatic; aryl; heteroaryl; alkoxy; (cycloaliphatic)oxy; (heterocycloaliphatic)oxy; aryloxy; heteroaryloxy; (araliphatic)oxy; (heteroaraliphatic)oxy; aroyl; heteroaroyl; amino; amido (e.g.,

(aliphatic)carbonylamino, (cycloaliphatic)carbonylamino, ((cycloaliphatic)aliphatic)carbonylamino, (aryl)carbonylamino, (araliphatic)carbonylamino, (heterocycloaliphatic)carbonylamino, ((heterocycloaliphatic)aliphatic)carbonylamino, (heteroaryl)carbonylamino, or (heteroaraliphatic)carbonylamino); nitro; carboxy (e.g., HOOC-, alkoxycarbonyl, or alkylcarbonyloxy); acyl (e.g., (cycloaliphatic)carbonyl, ((cycloaliphatic) aliphatic)carbonyl, (araliphatic)carbonyl, (heterocycloaliphatic)carbonyl, ((heterocycloaliphatic)aliphatic)carbonyl, or (heteroaraliphatic)carbonyl); cyano; halo; hydroxy; mercapto; sulfonyl (e.g., alkyl-SO_2- and aryl-SO_2-); sulfinyl (e.g., alkyl-S(O)-); sulfanyl (e.g., alkyl-S-); sulfoxy; urea; thiourea; sulfamoyl; sulfamide; oxo; or carbamoyl.

[035] As used herein, the term "heterocycloaliphatic" encompasses a heterocycloalkyl group and a heterocycloalkenyl group, each of which being optionally substituted as set forth below.

[036] As used herein, a "heterocycloalkyl" group refers to a 3-12 membered mono- or bicylic (fused or bridged) (e.g., 5- to 12-membered mono- or bicyclic) saturated ring structure, in which one or more of the ring atoms is a heteroatom (e.g., N, O, S, or combinations thereof). Examples of a heterocycloalkyl group include piperidyl, piperazyl, tetrahydropyranyl, tetrahydrofuryl, 1,4-dioxolanyl, 1,4-dithianyl, 1,3-dioxolanyl, oxazolidyl, isoxazolidyl, morpholinyl, thiomorpholyl, octahydrobenzofuryl, octahydrochromenyl, octahydrothiochromenyl, octahydroindolyl, octahydropyrindinyl, decahydroquinolinyl, octahydrobenzo[*b*]thiopheneyl, 2-oxa-bicyclo[2.2.2]octyl, 1-aza-bicyclo[2.2.2]octyl, 3-aza-bicyclo[3.2.1]octyl, 3-oxaspiro[5.5]]undec-3-enyl, 6-oxaspiro[2.5]oct-5-enyl,and 2,6-dioxa-tricyclo[3.3.1.$0^{3,7}$]nonyl.

[037] A "heterocycloalkenyl" group, as used herein, refers to a mono- or bicylic (e.g., 5- to 10-membered mono- or bicyclic) non-aromatic ring structure having one or more double bonds, and wherein one or more of the ring atoms is a heteroatom (e.g., N, O, or S).

[038] Monocyclic and bicycloheteroaliphatics are numbered according to standard chemical nomenclature. A heterocycloalkyl or heterocycloalkenyl group can be optionally substituted with one or more substituents such as phosphor; aliphatic [e.g., alkyl, alkenyl, or alkynyl]; cycloaliphatic; (cycloaliphatic)aliphatic; heterocycloaliphatic; (heterocycloaliphatic)aliphatic; aryl; heteroaryl; alkoxy; (cycloaliphatic)oxy; (heterocycloaliphatic)oxy; aryloxy; heteroaryloxy; (araliphatic)oxy; (heteroaraliphatic)oxy; aroyl; heteroaroyl; amino; amido (e.g., (aliphatic)carbonylamino, (cycloaliphatic)carbonylamino, ((cycloaliphatic) aliphatic)carbonylamino, (aryl)carbonylamino, (araliphatic)carbonylamino, (heterocycloaliphatic)carbonylamino,

((heterocycloaliphatic) aliphatic)carbonylamino, (heteroaryl)carbonylamino, or (heteroaraliphatic)carbonylamino); nitro; carboxy (e.g., HOOC-, alkoxycarbonyl, or alkylcarbonyloxy); acyl (e.g., (cycloaliphatic)carbonyl, ((cycloaliphatic) aliphatic)carbonyl, (araliphatic)carbonyl, (heterocycloaliphatic)carbonyl, ((heterocycloaliphatic)aliphatic)carbonyl, or (heteroaraliphatic)carbonyl); nitro; cyano; halo; hydroxy; mercapto; sulfonyl (e.g., alkylsulfonyl or arylsulfonyl); sulfinyl (e.g., alkylsulfinyl); sulfanyl (e.g., alkylsulfanyl); sulfoxy; urea; thiourea; sulfamoyl; sulfamide; oxo; or carbamoyl.

[039] A "heteroaryl" group, as used herein, refers to a monocyclic, bicyclic, or tricyclic ring system having 4 to 15 ring atoms wherein one or more of the ring atoms is a heteroatom (e.g., N, O, S, or combinations thereof) and in which the monocyclic ring system is aromatic or at least one of the rings in the bicyclic or tricyclic ring systems is aromatic. A heteroaryl group includes a benzofused ring system having 2 to 3 rings. For example, a benzofused group includes benzo fused with one or two 4 to 8 membered heterocycloaliphatic moieties (e.g., indolizyl, indolyl, isoindolyl, 3H-indolyl, indolinyl, benzo[*b*]furyl, benzo[*b*]thiophenyl, quinolinyl, or isoquinolinyl). Some examples of heteroaryl are azetidinyl, pyridyl, 1H-indazolyl, furyl, pyrrolyl, thienyl, thiazolyl, oxazolyl, imidazolyl, tetrazolyl, benzofuryl, isoquinolinyl, benzthiazolyl, xanthene, thioxanthene, phenothiazine, dihydroindole, 1,2,3,4-tetrahydroisoquinoline, isoindoline, benzo[1,3]dioxole, benzo[b]furyl, benzo[b]thiophenyl, indazolyl, benzimidazolyl, benzthiazolyl, puryl, cinnolyl, quinolyl, quinazolyl,cinnolyl, phthalazyl, quinazolyl, quinoxalyl, isoquinolyl, 4H-quinolizyl, benzo-1,2,5-thiadiazolyl, or 1,8-naphthyridyl.

[040] Without limitation, monocyclic heteroaryls include furyl, thiophenyl, 2H-pyrrolyl, pyrrolyl, oxazolyl, thazolyl, imidazolyl, pyrazolyl, isoxazolyl, isothiazolyl, 1,3,4-thiadiazolyl, 2H-pyranyl, 4-H-pranyl, pyridyl, pyridazyl, pyrimidyl, pyrazolyl, pyrazyl, or 1,3,5-triazyl. Monocyclic heteroaryls are numbered according to standard chemical nomenclature.

[041] Without limitation, bicyclic heteroaryls include indolizyl, indolyl, isoindolyl, 3H-indolyl, indolinyl, benzo[*b*]furyl, benzo[*b*]thiophenyl, quinolinyl, isoquinolinyl, indolizyl, isoindolyl, indolyl, benzo[*b*]furyl, bexo[*b*]thiophenyl, indazolyl, benzimidazyl, benzthiazolyl, purinyl, 4H-quinolizyl, quinolyl, isoquinolyl, cinnolyl, phthalazyl, quinazolyl, quinoxalyl, 1,8-naphthyridyl, or pteridyl. Bicyclic heteroaryls are numbered according to standard chemical nomenclature.

[042] A heteroaryl is optionally substituted with one or more substituents such as aliphatic (e.g., alkyl, alkenyl, or alkynyl); cycloaliphatic; (cycloaliphatic)aliphatic;

heterocycloaliphatic; (heterocycloaliphatic)aliphatic; aryl; heteroaryl; alkoxy; (cycloaliphatic)oxy; (heterocycloaliphatic)oxy; aryloxy; heteroaryloxy; (araliphatic)oxy; (heteroaraliphatic)oxy; aroyl; heteroaroyl; amino; oxo (on a non-aromatic carbocyclic or heterocyclic ring of a bicyclic or tricyclic heteroaryl); carboxy; amido; acyl (e.g., aliphaticcarbonyl; (cycloaliphatic)carbonyl; ((cycloaliphatic)aliphatic)carbonyl; (araliphatic)carbonyl; (heterocycloaliphatic)carbonyl; ((heterocycloaliphatic)aliphatic)carbonyl; or (heteroaraliphatic)carbonyl); sulfonyl (e.g., aliphaticsulfonyl or aminosulfonyl); sulfinyl (e.g., aliphaticsulfinyl); sulfanyl (e.g., aliphaticsulfanyl)); nitro; cyano; halo; hydroxy; mercapto; sulfoxy; urea; thiourea; sulfamoyl; sulfamide; or carbamoyl. Alternatively, a heteroaryl can be unsubstituted.

[043] Non-limiting examples of substituted heteroaryls include (halo)heteroaryl (e.g., mono- and di-(halo)heteroaryl); (carboxy)heteroaryl (e.g., (alkoxycarbonyl)heteroaryl); cyanoheteroaryl; aminoheteroaryl (e.g., ((alkylsulfonyl)amino)heteroaryl and((dialkyl)amino)heteroaryl); (amido)heteroaryl (e.g., aminocarbonylheteroaryl, ((alkylcarbonyl)amino)heteroaryl, ((((alkyl)amino)alkyl)aminocarbonyl)heteroaryl, (((heteroaryl)amino)carbonyl)heteroaryl, ((heterocycloaliphatic)carbonyl)heteroaryl, and ((alkylcarbonyl)amino)heteroaryl); (cyanoalkyl)heteroaryl; (alkoxy)heteroaryl; (sulfamoyl)heteroaryl [e.g., (aminosulfonyl)heteroaryl]; (sulfonyl)heteroaryl [e.g., (alkylsulfonyl)heteroaryl]; (hydroxyalkyl)heteroaryl; (alkoxyalkyl)heteroaryl; (hydroxy)heteroaryl; ((carboxy)alkyl)heteroaryl; (((dialkyl)amino)alkyl]heteroaryl; (heterocycloaliphatic)heteroaryl; (cycloaliphatic)heteroaryl; (nitroalkyl)heteroaryl; (((alkylsulfonyl)amino)alkyl)heteroaryl; ((alkylsulfonyl)alkyl)heteroaryl; (cyanoalkyl)heteroaryl; (acyl)heteroaryl [e.g., (alkylcarbonyl)heteroaryl]; (alkyl)heteroaryl, and (haloalkyl)heteroaryl (e.g., trihaloalkylheteroaryl).

[044] A "heteroaraliphatic" (such as a heteroaralkyl group) as used herein, refers to an aliphatic group (e.g., a C_{1-4} alkyl group) that is substituted with a heteroaryl group. "Aliphatic," "alkyl," and "heteroaryl" have been defined above.

[045] A "heteroaralkyl" group, as used herein, refers to an alkyl group (e.g., a C_{1-4} alkyl group) that is substituted with a heteroaryl group. Both "alkyl" and "heteroaryl" have been defined above. A heteroaralkyl is optionally substituted with one or more substituents such as alkyl (including carboxyalkyl, hydroxyalkyl, and haloalkyl such as trifluoromethyl), alkenyl, alkynyl, cycloalkyl, (cycloalkyl)alkyl, heterocycloalkyl, (heterocycloalkyl)alkyl, aryl, heteroaryl, alkoxy, cycloalkyloxy, heterocycloalkyloxy, aryloxy, heteroaryloxy, aralkyloxy, heteroaralkyloxy, aroyl, heteroaroyl, nitro, carboxy, alkoxycarbonyl,

alkylcarbonyloxy, aminocarbonyl, alkylcarbonylamino, cycloalkylcarbonylamino, (cycloalkylalkyl)carbonylamino, arylcarbonylamino, aralkylcarbonylamino, (heterocycloalkyl)carbonylamino, (heterocycloalkylalkyl)carbonylamino, heteroarylcarbonylamino, heteroaralkylcarbonylamino, cyano, halo, hydroxy, acyl, mercapto, alkylsulfanyl, sulfoxy, urea, thiourea, sulfamoyl, sulfamide, oxo, or carbamoyl.

[046] As used herein, an "acyl" group refers to a formyl group or R^X-C(O)- (such as alkyl-C(O)-, also referred to as "alkylcarbonyl") where R^X and "alkyl" have been defined previously. Acetyl and pivaloyl are examples of acyl groups.

[047] As used herein, an "aroyl" or "heteroaroyl" refers to an aryl-C(O)- or a heteroaryl-C(O)-. The aryl and heteroaryl portion of the aroyl or heteroaroyl is optionally substituted as previously defined.

[048] As used herein, an "alkoxy" group refers to an alkyl-O- group where "alkyl" has been defined previously.

[049] As used herein, a "carbamoyl" group refers to a group having the structure -O-CO-NR^XR^Y or -NR^X-CO-O-R^Z wherein R^X and R^Y have been defined above and R^Z can be aliphatic, aryl, araliphatic, heterocycloaliphatic, heteroaryl, or heteroaraliphatic.

[050] As used herein, a "carboxy" group refers to -COOH, -$COOR^X$, -OC(O)H, -$OC(O)R^X$ when used as a terminal group; or -OC(O)- or -C(O)O- when used as an internal group.

[051] As used herein, a "haloaliphatic" group refers to an aliphatic group substituted with 1-3 halogen. For instance, the term haloalkyl includes the group -CF_3.

[052] As used herein, a "mercapto" group refers to -SH.

[053] As used herein, a "sulfo" group refers to -SO_3H or -SO_3R^X when used terminally or -$S(O)_3$- when used internally.

[054] As used herein, a "sulfamide" group refers to the structure -NR^X-$S(O)_2$-NR^YR^Z when used terminally and -NR^X-$S(O)_2$-NR^Y- when used internally, wherein R^X, R^Y, and R^Z have been defined above.

[055] As used herein, a "sulfonamide" group refers to the structure -$S(O)_2$-NR^XR^Y or -NR^X-$S(O)_2$-R^Z when used terminally; or -$S(O)_2$-NR^X- or -NR^X -$S(O)_2$- when used internally, wherein R^X, R^Y, and R^Z are defined above.

[056] As used herein a "sulfanyl" group refers to -S-R^X when used terminally and -S- when used internally, wherein R^X has been defined above. Examples of sulfanyls include aliphatic-S-, cycloaliphatic-S-, aryl-S-, or the like.

[057] As used herein a "sulfinyl" group refers to -S(O)-R^X when used terminally and -S(O)- when used internally, wherein R^X has been defined above. Exemplary sulfinyl groups

include aliphatic-S(O)-, aryl-S(O)-, (cycloaliphatic(aliphatic)) -S(O)-, cycloalkyl-S(O)-, heterocycloaliphatic-S(O)-, heteroaryl-S(O)-, or the like.

[058] As used herein, a "sulfonyl" group refers to-$S(O)_2$-R^X when used terminally and -$S(O)_2$- when used internally, wherein R^X has been defined above. Exemplary sulfonyl groups include aliphatic-$S(O)_2$-, aryl-$S(O)_2$-, (cycloaliphatic(aliphatic))-$S(O)_2$-, cycloaliphatic-$S(O)_2$-, heterocycloaliphatic-$S(O)_2$-, heteroaryl-$S(O)_2$-, (cycloaliphatic(amido(aliphatic)))-$S(O)_2$-or the like.

[059] As used herein, a "sulfoxy" group refers to -O-SO-R^X or -SO-O-R^X when used terminally and -O-S(O)- or -S(O)-O- when used internally, where R^X has been defined above.

[060] As used herein, a "halogen" or "halo" group refers to fluorine, chlorine, bromine or iodine.

[061] As used herein, an "alkoxycarbonyl," which is encompassed by the term carboxy, used alone or in connection with another group refers to a group such as alkyl-O-C(O)-.

[062] As used herein, an "alkoxyalkyl" refers to an alkyl group such as alkyl-O-alkyl-, wherein alkyl has been defined above.

[063] As used herein, a "carbonyl" refer to -C(O)-.

[064] As used herein, an "oxo" refers to =O.

[065] As used herein, the term "phospho" refers to phosphinates and phosphonates. Examples of phosphinates and phosphonates include -$P(O)(R^P)_2$, wherein R^P is aliphatic, alkoxy, aryloxy, heteroaryloxy, (cycloaliphatic)oxy, (heterocycloaliphatic)oxy aryl, heteroaryl, cycloaliphatic or amino.

[066] As used herein, an "aminoalkyl" refers to the structure $(R^X)_2N$-alkyl-.

[067] As used herein, a "cyanoalkyl" refers to the structure (NC)-alkyl-.

[068] As used herein, a "urea" group refers to the structure -NR^X-CO-NR^YR^Z and a "thiourea" group refers to the structure -NR^X-CS-NR^YR^Z when used terminally and -NR^X-CO-NR^Y- or -NR^X-CS-NR^Y- when used internally, wherein R^X, R^Y, and R^Z have been defined above.

[069] As used herein, a "guanidine" group refers to the structure -N=C(N(R^XR^Y))N(R^XR^Y) or -NR^X-C(=NR^X)NR^XR^Y wherein R^X and R^Y have been defined above.

[070] As used herein, the term "amidino" group refers to the structure -C=(NR^X)N(R^XR^Y) wherein R^X and R^Y have been defined above.

[071] In general, the term "vicinal" refers to the placement of substituents on a group that includes two or more carbon atoms, wherein the substituents are attached to adjacent carbon atoms.

[072] In general, the term "geminal" refers to the placement of substituents on a group that includes two or more carbon atoms, wherein the substituents are attached to the same carbon atom.

[073] The terms "terminally" and "internally" refer to the location of a group within a substituent. A group is terminal when the group is present at the end of the substituent not further bonded to the rest of the chemical structure. Carboxyalkyl, i.e., $R^XO(O)C$-alkyl is an example of a carboxy group used terminally. A group is internal when the group is present in the middle of a substituent of the chemical structure. Alkylcarboxy (e.g., alkyl-C(O)O- or alkyl-OC(O)-) and alkylcarboxyaryl (e.g., alkyl-C(O)O-aryl- or alkyl-O(CO)-aryl-) are examples of carboxy groups used internally.

[074] As used herein, "cyclic group" or "cyclic moiety" includes mono-, bi-, and tri-cyclic ring systems including cycloaliphatic, heterocycloaliphatic, aryl, or heteroaryl, each of which has been previously defined.

[075] As used herein, a "bridged bicyclic ring system" refers to a bicyclic heterocycloalipahtic ring system or bicyclic cycloaliphatic ring system in which the rings are bridged. Examples of bridged bicyclic ring systems include, but are not limited to, adamantanyl, norbornanyl, bicyclo[3.2.1]octyl, bicyclo[2.2.2]octyl, bicyclo[3.3.1]nonyl, bicyclo[3.2.3]nonyl, 2-oxabicyclo[2.2.2]octyl, 1-azabicyclo[2.2.2]octyl, 3-azabicyclo[3.2.1]octyl, and 2,6-dioxa-tricyclo[$3.3.1.0^{3,7}$]nonyl. A bridged bicyclic ring system can be optionally substituted with one or more substituents such as alkyl (including carboxyalkyl, hydroxyalkyl, and haloalkyl such as trifluoromethyl), alkenyl, alkynyl, cycloalkyl, (cycloalkyl)alkyl, heterocycloalkyl, (heterocycloalkyl)alkyl, aryl, heteroaryl, alkoxy, cycloalkyloxy, heterocycloalkyloxy, aryloxy, heteroaryloxy, aralkyloxy, heteroaralkyloxy, aroyl, heteroaroyl, nitro, carboxy, alkoxycarbonyl, alkylcarbonyloxy, aminocarbonyl, alkylcarbonylamino, cycloalkylcarbonylamino, (cycloalkylalkyl)carbonylamino, arylcarbonylamino, aralkylcarbonylamino, (heterocycloalkyl)carbonylamino, (heterocycloalkylalkyl)carbonylamino, heteroarylcarbonylamino, heteroaralkylcarbonylamino, cyano, halo, hydroxy, acyl, mercapto, alkylsulfanyl, sulfoxy, urea, thiourea, sulfamoyl, sulfamide, oxo, or carbamoyl.

[076] As used herein, an "aliphatic chain" refers to a branched or straight aliphatic group (e.g., alkyl groups, alkenyl groups, or alkynyl groups). A straight aliphatic chain has the structure $-(CH_2)_v-$, where v is 1-6. A branched aliphatic chain is a straight aliphatic chain that is substituted with one or more aliphatic groups. A branched aliphatic chain has the structure $-(CHQ)_v-$ where Q is hydrogen or an aliphatic group; however, Q shall be an aliphatic group

in at least one instance. The term aliphatic chain includes alkyl chains, alkenyl chains, and alkynyl chains, where alkyl, alkenyl, and alkynyl are defined above.

[077] The phrase "optionally substituted" is used interchangeably with the phrase "substituted or unsubstituted." As described herein, compounds of the invention can optionally be substituted with one or more substituents, such as are illustrated generally above, or as exemplified by particular classes, subclasses, and species of the invention. As described herein, the variables A, B, R_1, R_2, R_3, Y and Y', and other variables contained in formulae described herein encompass specific groups, such as alkyl and aryl. Unless otherwise noted, each of the specific groups for the variables A, B, R_1, R_2, R_3, Y and Y', and other variables contained therein can be optionally substituted with one or more substituents described herein. Each substituent of a specific group is further optionally substituted with one to three of halo, cyano, oxo, alkoxy, hydroxy, amino, nitro, aryl, cycloaliphatic, heterocycloaliphatic, heteroaryl, haloalkyl, and alkyl. For instance, an alkyl group can be substituted with alkylsulfanyl and the alkylsulfanyl can be optionally substituted with one to three of halo, cyano, oxo, alkoxy, hydroxy, amino, nitro, aryl, haloalkyl, and alkyl. As an additional example, the cycloalkyl portion of a (cycloalkyl)carbonylamino can be optionally substituted with one to three of halo, cyano, alkoxy, hydroxy, nitro, haloalkyl, and alkyl. When two alkoxy groups are bound to the same atom or adjacent atoms, the two alkoxy groups can form a ring together with the atom(s) to which they are bound.

[078] In general, the term "substituted," whether preceded by the term "optionally" or not, refers to the replacement of hydrogen radicals in a given structure with the radical of a specified substituent. Specific substituents are described above in the definitions and below in the description of compounds and examples thereof. Unless otherwise indicated, an optionally substituted group can have a substituent at each substitutable position of the group, and when more than one position in any given structure can be substituted with more than one substituent selected from a specified group, the substituent can be either the same or different at every position. A ring substituent, such as a heterocycloalkyl, can be bound to another ring, such as a cycloalkyl, to form a spiro-bicyclic ring system, e.g., both rings share one common atom. As one of ordinary skill in the art will recognize, combinations of substituents envisioned by this invention are those combinations that result in the formation of stable or chemically feasible compounds.

[079] The phrase "stable or chemically feasible," as used herein, refers to compounds that are not substantially altered when subjected to conditions to allow for their production, detection, and preferably their recovery, purification, and use for one or more of the purposes

disclosed herein. In some embodiments, a stable compound or chemically feasible compound is one that is not substantially altered when kept at a temperature of 40 °C or less, in the absence of moisture or other chemically reactive conditions, for at least a week.

[080] As used herein, an effective amount is defined as the amount required to confer a therapeutic effect on the treated patient, and is typically determined based on age, surface area, weight, and condition of the patient. The interrelationship of dosages for animals and humans (based on milligrams per meter squared of body surface) is described by Freireich et al., Cancer Chemother. Rep., 50: 219 (1966). Body surface area may be approximately determined from height and weight of the patient. See, e.g., Scientific Tables, Geigy Pharmaceuticals, Ardsley, New York, 537 (1970). As used herein, "patient" refers to a mammal, including a human.

[081] Unless otherwise stated, structures depicted herein are also meant to include all isomeric (e.g., enantiomeric, diastereomeric, and geometric (or conformational)) forms of the structure; for example, the R and S configurations for each asymmetric center, (Z) and (E) double bond isomers, and (Z) and (E) conformational isomers. Therefore, single stereochemical isomers as well as enantiomeric, diastereomeric, and geometric (or conformational) mixtures of the present compounds are within the scope of the invention.

[082] Unless otherwise stated, all tautomeric forms of the compounds of the invention are within the scope of the invention. Additionally, unless otherwise stated, structures depicted herein are also meant to include compounds that differ only in the presence of one or more isotopically enriched atoms. Such compounds are useful, for example, as analytical tools or probes in biological assays, or as therapeutic agents.

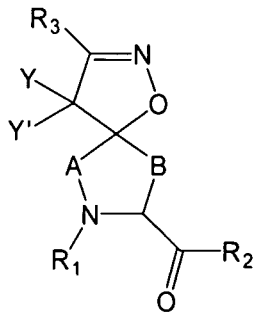
[083] In other aspects, the invention features certain compounds as described generically and specifically below. Such specific descriptions are illustrative only and are not meant to limit scope of the compounds or uses thereof.

DETAILED DESCRIPTION

I. COMPOUNDS

A. Generic Compounds

[084] In some aspects, the invention provides compounds of formula (I) useful for inhibiting serine protease activity and methods of inhibiting serine protease activity. Compounds of formula (I) include:



(I)

or a pharmaceutically acceptable salt thereof, wherein,

each A is $-(CX_1X_2)_a-$;

each B is $-(CX_1X_2)_b-$;

each X_1 is independently hydrogen, halo, amino, sulfanyl, optionally substituted C_{1-4} aliphatic, optionally substituted aryl, or $-O-X_{1A}$;

each X_2 is independently hydrogen, halo, amino, sulfanyl, optionally substituted C_{1-4} aliphatic, optionally substituted aryl, or $-O-X_{1B}$;

X_{1A} and X_{1B} are each independently an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or,

X_1 and X_2 together form an oxo group;

each R_1 is $-Z^AR_4$, wherein each Z^A is independently a bond or an optionally substituted branched or straight C_{1-12} aliphatic chain wherein up to three carbon units of Z^A are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^A-$, $-C(O)NR^ANR^A-$, -C(O)O-, $-NR^AC(O)O-$, -O-, $-NR^AC(O)NR^A-$, $-NR^ANR^A-$, -S-, -SO-, $-SO_2-$, $-NR^A-$, $-SO_2NR^A-$, or $-NR^ASO_2NR^A-$ provided that $-NR^ANR^A-$, $-NR^AC(O)NR^A-$, or $-NR^ASO_2NR^A-$ is not directly bound to the nitrogen ring atom of formula (I);

each R_4 is independently R^A, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$;

each R^A is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_2 is $-Z^BR_5$, wherein each Z^B is independently a bond or an optionally substituted branched or straight C_{1-12} aliphatic chain wherein up to three carbon units of Z^B are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^B-$, $-C(O)NR^BNR^B-$, -C(O)O-, $-NR^BC(O)O-$, $-NR^BC(O)NR^B-$, $-NR^BNR^B-$, -S-, -SO-, $-SO_2-$, $-NR^B-$, $-SO_2NR^B-$, or $-NR^BSO_2NR^B-$, provided that SO, SO_2, or $-SO_2NR^B-$ is not directly bound to the carbonyl of formula (I);

each R_5 is independently R^B, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$;

each R^B is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or,

R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted heterocycloaliphatic ring;

each R_3 is an optionally substituted aliphatic, amino, sulfonyl, sulfanyl, sulfinyl, sulfonamide, sulfamide, sulfo, $-O-R_{3A}$, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_{3A} is independently an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

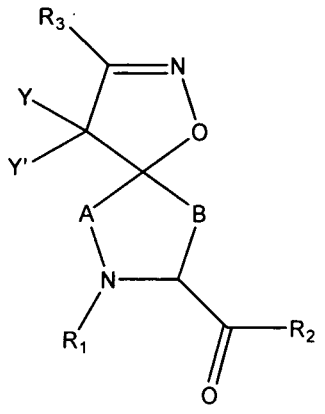
each OF Y and Y' is independently $-Z^DR_7$, wherein each Z^D is independently a bond or an optionally substituted straight or branched C_{1-6} aliphatic chain wherein up to two carbon units of Z^D are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^D-$, $-C(O)NR^DNR^D-$, -C(O)O-, $-NR^DC(O)O-$, -O-, $-NR^DC(O)NR^D-$, $-NR^DNR^D-$, -S-, -SO-, $-SO_2-$, $-NR^D-$, $-SO_2NR^D-$, $-NR^DSO_2-$, or $-NR^DSO_2NR^D-$, or Y and Y' together form =O or =S;

each R_7 is independently R^D, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$;

each R^D is independently hydrogen, or optionally substituted aryl; and

each of **a** and **b** is independently 0, 1, 2, or 3; provided that the sum of **a** and **b** is 2 or 3.

[085] In another aspect, the invention provides compounds of formula (I) below



(I)

or a pharmaceutically acceptable salt thereof. In this formula,

each A is $-CH_2-$;

each B is $-CH_2-$;

each R_1 is $-Z^AR_4$, wherein Z^A is -C(O)- and R_4 is a substituted alkyl;

each R_2 is $-Z^BR_5$, wherein each Z^B is independently a bond or an optionally substituted aliphatic wherein up to four carbon units of Z^B are optionally and independently replaced by -C(O)-, $-C(O)NR^B-$, $-C(O)C(O)-NR^B-$, or $-NR^B-$;

each R_5 is independently R^B, halo, -OH, -CN, $-NO_2$, $-NH_2$, alkoxy, or haloalkoxy;

each R^B is independently hydrogen, an optionally substituted aliphatic, or an optionally substituted cycloaliphatic; or

R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted 6-membered heterocycloaliphatic ring;

each R_3 is an optionally substituted amino, $-O-R_{3A}$, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_{3A} is independently an optionally substituted aryl or an optionally substituted heteroaryl; and

each of Y and Y' is H.

B. Specific Compounds

1. Substituent R_1:

[086] Each R_1 is $-Z^AR_4$, wherein each Z^A is independently a bond or an optionally substituted branched or straight C_{1-12} aliphatic chain wherein up to three carbon units of Z^A are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^A-$, $-C(O)NR^ANR^A-$, -C(O)O-, $-NR^AC(O)O-$, -O-, $-NR^AC(O)NR^A-$, $-NR^ANR^A-$, -S-, -SO-, $-SO_2-$, $-NR^A-$, $-SO_2NR^A-$, or $-NR^ASO_2NR^A-$ provided that $-NR^ANR^A-$, $-NR^AC(O)NR^A-$, or $-NR^ASO_2NR^A-$ is not directly bound to the nitrogen ring atom of formula (I).

[087] Each R_4 is independently R^A, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$.

[088] Each R^A is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl.

[089] In several embodiments R_1 is optionally substituted with 1 to 4 substituents.

[090] In certain embodiments, R_1 is $-Q_4-W_4-Q_3-W_3-Q_2-W_2-Q_1$; wherein each of W_2, W_3, and W_4 is independently a bond, -C(O)-, -C(S)-, $-C(O)N(Q_5)-$, -C(O)O-, -O-, $-N(Q_5)C(O)N(Q_5)-$, $-SO_2-$, $-N(Q_5)SO_2-$, -S-, $-N(Q_5)-$, -SO-, -OC(O)-, $-N(Q_5)C(O)O-$, or $-SO_2N(Q_5)-$; each of Q_1, Q_2, Q_3 and Q_4 is independently a bond, an optionally substituted C_{1-4} aliphatic, an optionally substituted cycloaliphatic, an optionally substituted

heterocycloaliphatic, an optionally substituted aryl, an optionally substituted heteroaryl, or a hydrogen when Q_1, Q_2, Q_3, or Q_4 is the terminal group of R_1; and each Q_5 is independently hydrogen or an optionally substituted aliphatic. In some specific embodiments, Q_4 is a bond.

[091] In several embodiments, R_1 is an optionally substituted acyl group. In several examples, R_1 is an optionally substituted alkylcarbonyl. Additional examples of R_1 include (amino)alkylcarbonyl, (halo)alkylcarbonyl, (aryl)alkylcarbonyl, (cycloaliphatic)alkylcarbonyl, or (heterocycloaliphatic)alkylcarbonyl. Included in these examples are embodiments where R_1 is (heterocycloalkyl(oxy(carbonyl(amino))))alkylcarbonyl, (heteroaryl(carbonyl(amino(alkyl(carbonyl(amino)))))alkylcarbonyl, (bicycloaryl(sulfonyl(amino)))alkylcarbonyl, (aryl(alkoxy(carbonyl(amino))))alkylcarbonyl, (alkyl(carbonyl(amino)))alkylcarbonyl, (alkenyl(alkoxy(carbonyl(amino))))alkylcarbonyl, (cycloaliphatic(alkyl(amino(carbonyl(amino)))))alkylcarbonyl, (heteroaryl(carbonyl(amino(alkyl(carbonyl(amino))))))alkylcarbonyl, (alkyl(amino(carbonyl(amino))))alkylcarbonyl, or (bicycloaryl(amino(carbonyl(amino))))alkylcarbonyl, each of which is optionally substituted with 1-3 substituents.

[092] In several embodiments, R_1 is an optionally substituted carboxy group. In one example, R_1 is optionally substituted alkoxycarbonyl. Another example of R_1 includes C_{1-4} alkoxycarbonyl, or (tricyclic aryl)alkoxycarbonyl, each of which is optionally substituted with 1-3 substituents. Other carboxy groups include (aliphatic(oxy))carbonyl, a (heteroaralkyl(oxy))carbonyl, (heterocycloaliphatic(oxy)carbonyl, (aralkyl(oxy))carbonyl, each of which is optionally substituted with 1-3 of halo, alkoxy, aliphatic, cycloaliphatic, heterocycloaliphatic, aryl, heteroaryl, or combinations thereof.

[093] In several embodiments, R_1 is optionally substituted aminocarbonyl. Examples of R_1 include (alkoxy(aryl(alkyl)))aminocarbonyl, (alkyl)aminocarbonyl, or (aryl(alkoxy(carbonyl(alkyl(amino(carbonyl(alkyl))))))aminocarbonyl, each of which is optionally substituted with 1-3 substituents.

[094] In several embodiments, R_1 is optionally substituted heteroaryl. In one example, R_1 is an optionally substituted oxazolyl, pyrrolyl, furyl, thiophenyl, triazinyl, pyridinyl, pyrazinyl, pyrimidinyl, or pyridazinyl.

[095] In several embodiments, R_1 is an alkylsulfonyl, aminosulfonyl, arylsulfonyl, heteroarylsulfonyl, cycloaliphaticsulfonyl, or heterocycloaliphaticsulfonyl, each of which is optionally substituted with 1-4 substituents.

[096] In several embodiments, R_1 is an optionally substituted alkylsulfonyl. Examples of R_1 include (aryl)alkylsulfonyl, or (alkyl(amino))alkylsulfonyl, alkylsulfonyl, aminosulfonyl, arylsulfonyl, heteroarylsulfonyl, cycloaliphaticsulfonyl, or heterocycloaliphaticsulfonyl, each of which is optionally substituted 1-3 substituents. In certain embodiments, R_1 is an optionally substituted alkylsulfonyl.

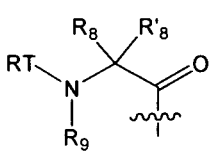
[097] In several embodiments, R_1 is an (aryl)alkylsulfonyl, or (alkyl(amino))alkylsulfonyl, each of which is optionally substituted.

[098] In some specific embodiments, R_1 is (amino)alkylcarbonyl, (halo)alkylcarbonyl, (aryl)alkylcarbonyl, (cycloaliphatic)alkylcarbonyl, or (heterocycloaliphatic)alkylcarbonyl, (heterocycloalkyl(oxy(carbonyl(amino))))alkylcarbonyl, (heteroaryl(carbonyl(amino(alkyl(carbonyl(amino)))))alkylcarbonyl, (bicycloaryl(sulfonyl(amino)))alkylcarbonyl, (aryl(alkoxy(carbonyl(amino))))alkylcarbonyl, (alkyl(carbonyl(amino)))alkylcarbonyl, (alkenyl(alkoxy(carbonyl(amino))))alkylcarbonyl, (cycloaliphatic(alkyl(amino(carbonyl(amino)))))alkylcarbonyl, (heteroaryl(carbonyl(amino(alkyl(carbonyl(amino))))))alkylcarbonyl, (alkyl(amino(carbonyl(amino))))alkylcarbonyl, or (bicycloaryl(amino(carbonyl(amino))))alkylcarbonyl, each of which is optionally substituted.

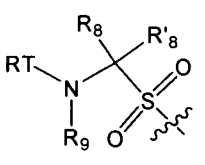
[099] In other specific embodiments, R_1 is a heteroarylcarbonyl, a (cycloaliphatic(alkyl(amido(alkyl))))carbonyl, a (heterocycloaliphatic(oxy(amido(alkyl))))carbonyl, an (aryl(sulfonyl(amino(alkyl))))carbonyl, an (aralkyl(oxy(amido(alkyl))))carbonyl, an (aliphatic(oxy(amido(alkyl))))carbonyl, a (cycloaliphatic(alkyl(amido(alkyl))))carbonyl, a (heterocycloaliphatic)carbonyl, or a (heteroaryl(amido(alkyl(amido(alkyl))))carbonyl, each of which is optionally substituted with 1-4 of halo, aliphatic, cycloaliphatic, acyl, alkoxy, or combinations thereof.

[0100] In still other embodiments, R_1 is amido. For example, R_1 is (alkoxy(aryl(alkyl)))aminocarbonyl, (alkyl)aminocarbonyl, or (aryl(alkoxy(carbonyl(alkyl(amino(carbonyl(alkyl)))))))aminocarbonyl, each of which is optionally substituted.

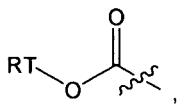
[0101] In several embodiments, R_1 is



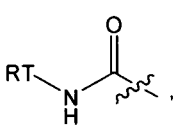
QI



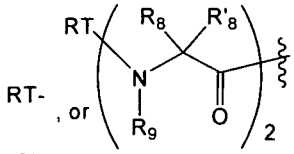
QII



QIII



QIV

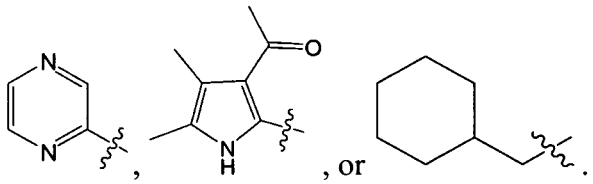


QV QVI

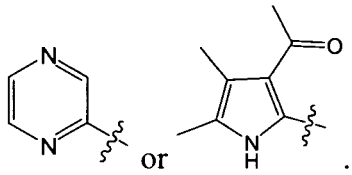
wherein T is a bond, -C(O)-, -OC(O)-, -NHC(O)-, -$S(O)_2N(H)$-, -C(O)C(O)- or -SO_2-; each R is independently hydrogen, amino, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; each R_8 and R'_8 is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; and each R_9 is independently hydrogen, an optionally substituted aliphatic, an optionally substituted heteroaryl, an optionally substituted phenyl, or R_8 and R_9, bound on adjacent atoms, taken together with the atoms to which they are attached form a 5 to 7 membered, optionally substituted monocyclic heterocycloaliphatic, or a 6 to 12 membered, optionally substituted bicyclic heterocycloaliphatic; or R_8 and R'_8, taken together with the atoms to which they are attached form an optionally substituted cycloaliphatic or an optionally substituted heterocycloaliphatic. For clarity, when R_1 is QVI, each of R_8, R'_8 and R_9 in each subunit can be independently selected as described above. The set of R_8, R'_8 and R_9 variables in one subunit need not necessarily be identical to the same set of R_8, R'_8 and R_9 variables in the other subunit.

[0102] In other embodiments, R_1 is QI or QII.

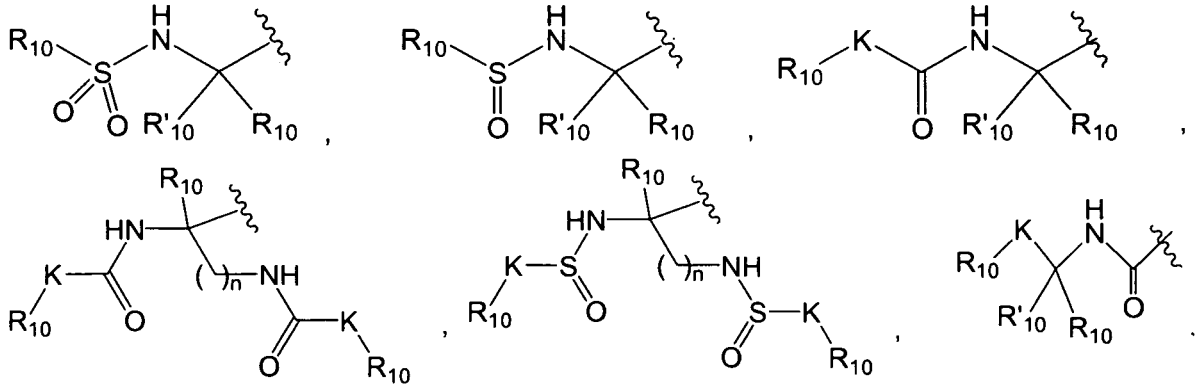
[0103] In some embodiments, R in the substituent in QI, QII, QIII, QIV, QV, or QVI is

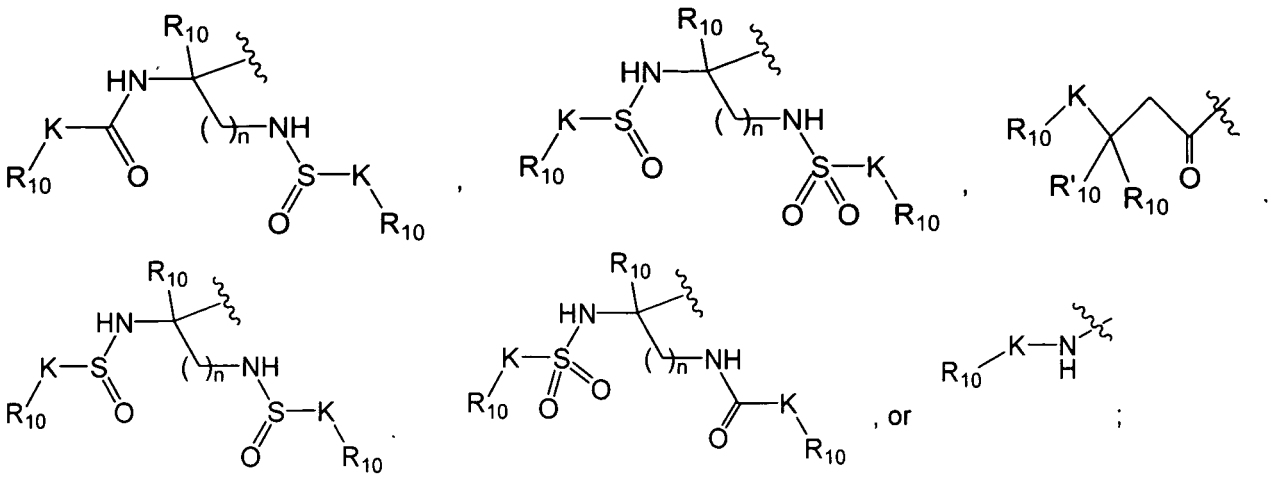


[0104] In other embodiments, R_1 is QVI and R is



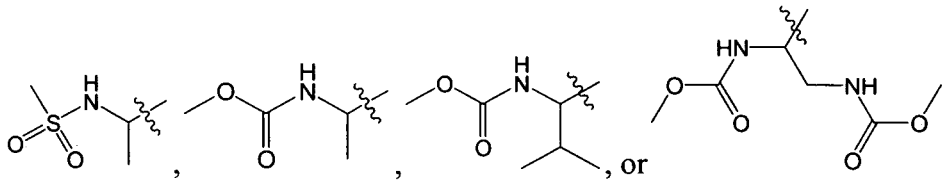
[0105] In other embodiments, R in the substituent QI, QII, QIII, QIV, QV, or QVI is



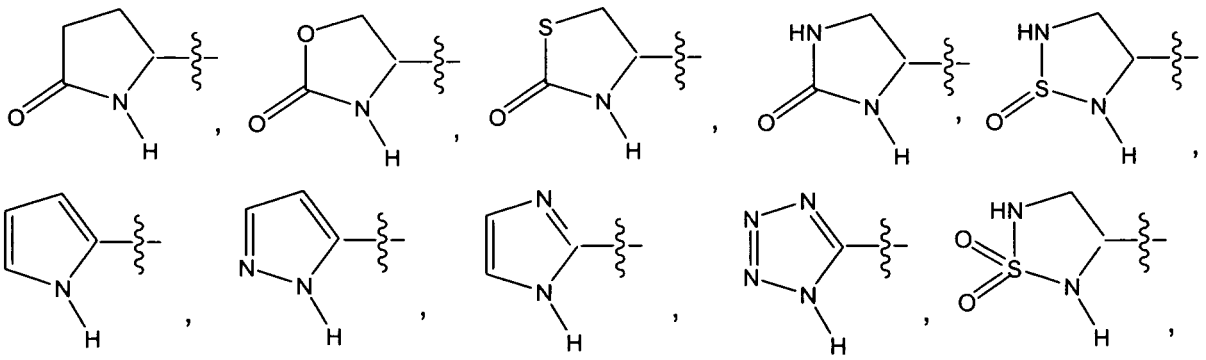


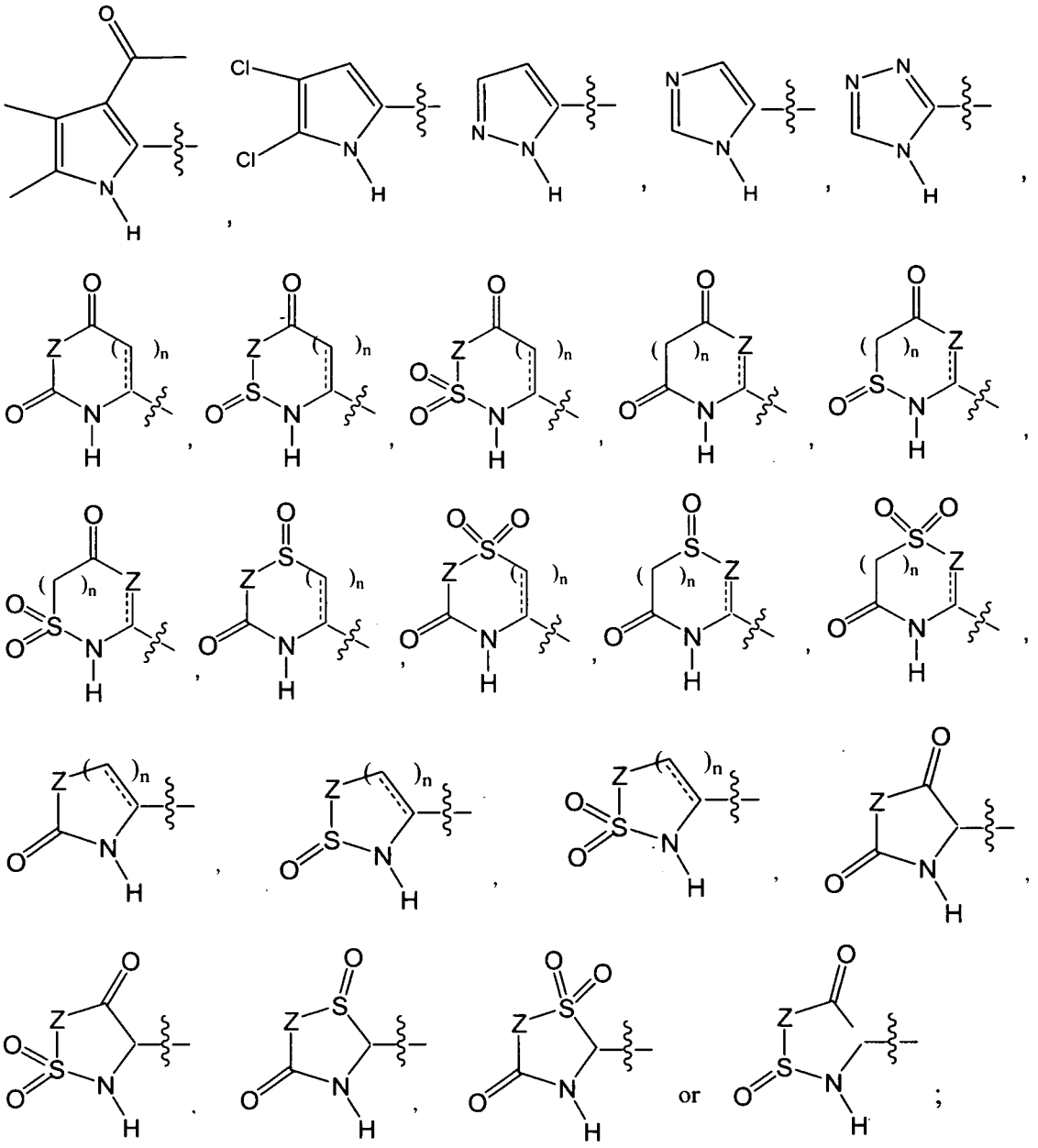
wherein each of R_{10} and R'_{10} is independently hydrogen, optionally substituted aliphatic, optionally substituted aryl, optionally substituted heteroaryl, optionally substituted heterocycloaliphatic, or optionally substituted cycloaliphatic, or R_{10} and R'_{10} together with the atom to which they are both bound form an optionally substituted cycloaliphatic or an optionally substituted heterocycloaliphatic; and each K is independently a bond, C_{1-12} aliphatic, -O-, -S-, $-S(O)_2-$, $-NR_{14}-$, -C(O)-, or $-C(O)NR_{14}-$, wherein R_{14} is hydrogen or an optionally substituted C_{1-12} aliphatic; and n is 1-3. For clarity, when more than one R_{10} is present in QI, QII, QIII, QIV, QV, or QVI, each R_{10} can be the same or different. In several embodiments, R_{10} or R'_{10} is [C_{3-10} cycloalkyl or cycloalkenyl]-C_{1-12} aliphatic, (3 to 10 membered)-heterocycloaliphatic, (3 to 10 membered)-heterocycloaliphatic-C_{1-12} aliphatic-, (5 to 10 membered)-heteroaryl, or (5 to 10 membered)-heteroaryl-C_{1-12} aliphatic-.

[0106] In still other embodiments, R in the substituent in QI, QII, QIII, QIV, QV, or QVI is

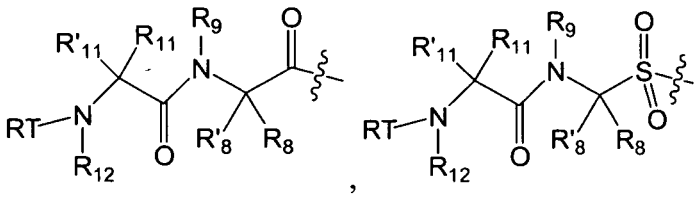


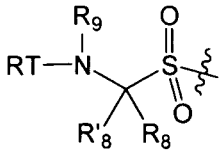
[0107] In further embodiments, R in the substituent in QI, QII, QIII, QIV, QV, or QVI is





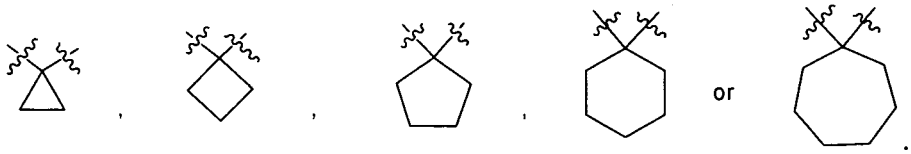
wherein each Z is independently -O-, -S-, -NR_{50}-, or -$C(R_{50})_2$-, ------ is independently a single bond or a double bond, and each R_{50} is independently hydrogen, optionally substituted aliphatic, optionally substituted aryl, optionally substituted heteroaryl, optionally substituted heterocycloaliphatic, or optionally substituted cycloaliphatic; and n is 1 or 2.

[0108] In several embodiments, R_1 is 

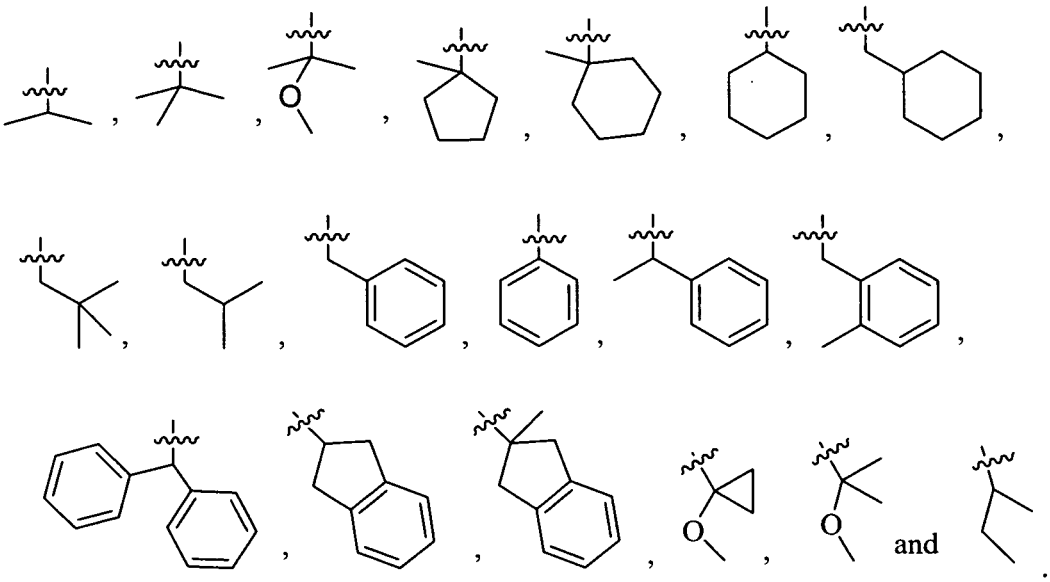
or  , wherein wherein T is a bond, -C(O)-, -OC(O)-, -NHC(O)-,

$-S(O)_2N(H)-$, $-C(O)C(O)-$ or $-SO_2-$; each R is independently hydrogen, amino, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; each R_8 and R'_8 is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; and each R_9 is independently hydrogen, an optionally substituted aliphatic, an optionally substituted heteroaryl, an optionally substituted phenyl, or R_8 and R_9, bound on adjacent atoms, taken together with the atoms to which they are attached form a 5 to 7 membered, optionally substituted monocyclic heterocycloaliphatic, or a 6 to 12 membered, optionally substituted bicyclic heterocycloaliphatic, in which each heterocycloaliphatic ring; or R_8 and R'_8, taken together with the atoms to which they are attached form an optionally substituted cycloaliphatic or an optionally substituted heterocycloaliphatic; each R_{11} and R'_{11} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted heteroaryl, an optionally substituted phenyl, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic; or R_{11} and R'_{11} together with the atom to which they are both attached form an optionally substituted 3 to 7 membered cycloaliphatic or heterocycloaliphatic ring; and each R_{12} is independently hydrogen or a protecting group.

[0109] In some embodiments, R_{11} and R'_{11} together with the atom to which they are attached form a 3 to 7 membered ring. Non-limiting examples include

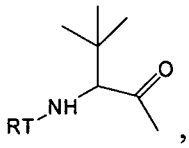


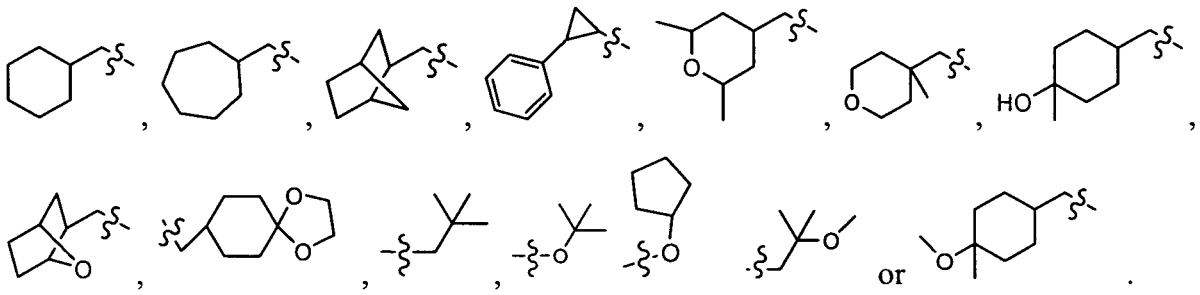
[0110] Non-limiting examples of R_8 and R_{11} include



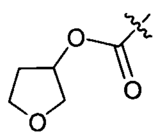
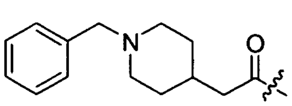
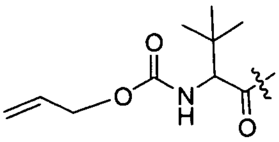
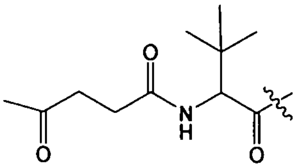
[0111] Alternatively, R_8 and R_{11} together with the atoms to which they are attached may form an optionally substituted 5 to 7 membered monocyclic heterocycloaliphatic or an optionally substituted 6 to 12 membered bicyclic heterocycloaliphatic, in which each heterocycloaliphatic or aryl ring optionally contains an additional heteroatom selected from O, S and N.

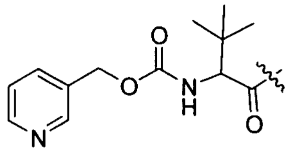
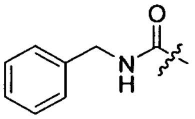
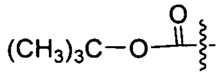
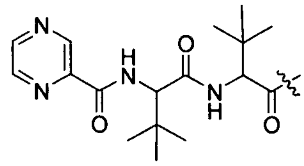
[0112] Also, R_8 and R_9 together to with the atoms to which they are attached can form a ring, R_7 and the ring system formed by R_8 and R_9 form an optionally substituted 8 to 14 membered bicyclic fused ring system, wherein the bicyclic fused ring system is optionally further fused with an optionally substituted phenyl to form an optionally substituted 10 to 16 membered tricyclic fused ring system.

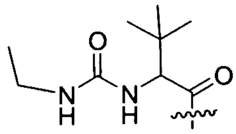
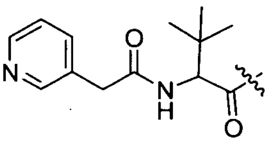
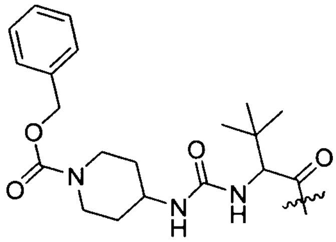
[0113] In several embodiments, R_1 is:  , wherein T is -C(O)-, and R is

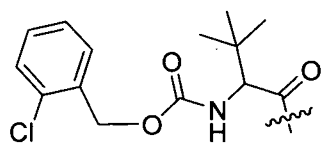
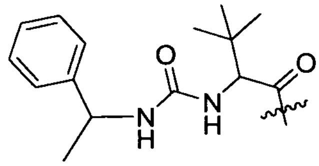
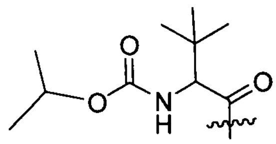


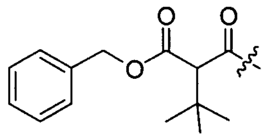
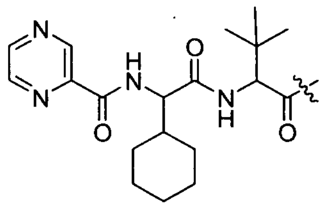
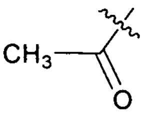
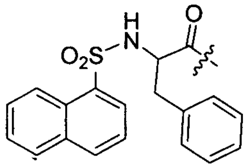
[0114] In several embodiments, R_1 is a group selected from:

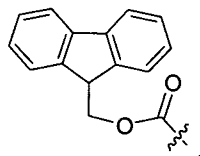
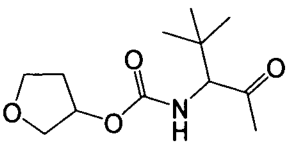
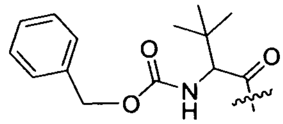
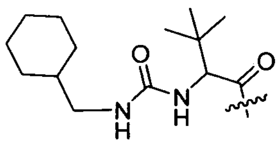
 ,  ,  ,  ,

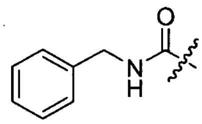
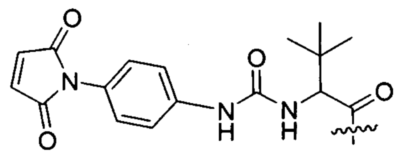
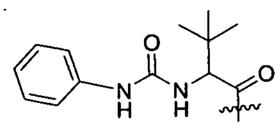
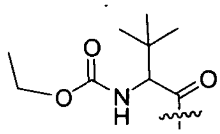
 ,  ,  ,  ,

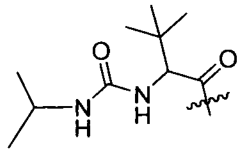
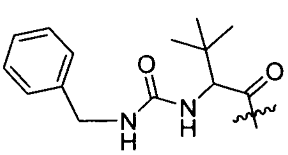
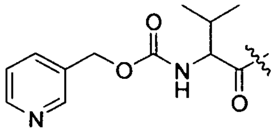
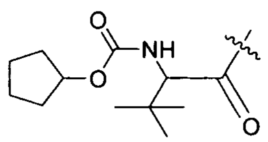
 ,  ,  ,

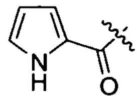
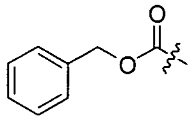
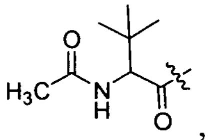
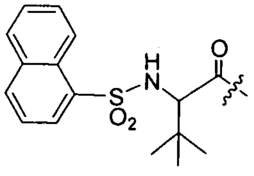
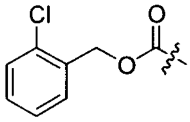
 ,  ,  ,

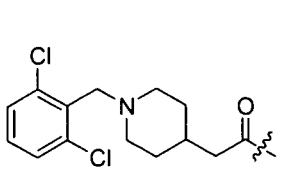
 ,  ,  ,  ,

 ,  ,  ,  ,

 ,  ,  ,  ,

 ,  ,  ,  ,

 ,  ,  ,  ,  ,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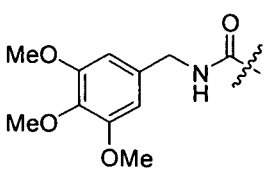
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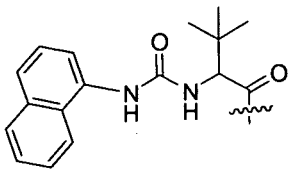
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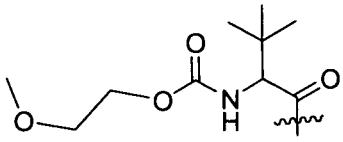
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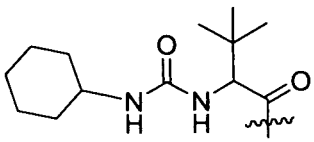
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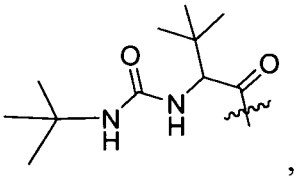
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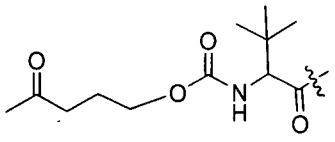
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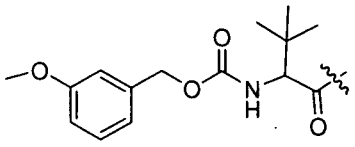
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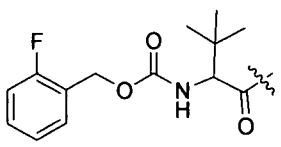
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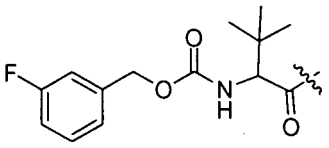
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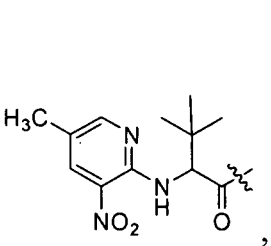
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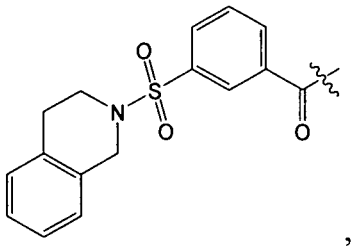
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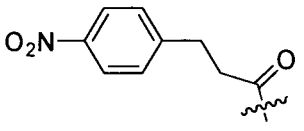
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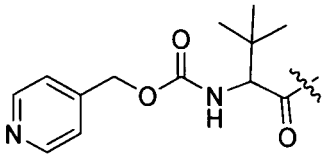
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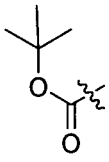
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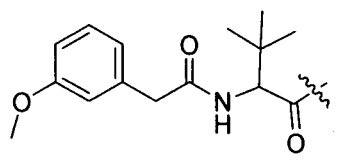
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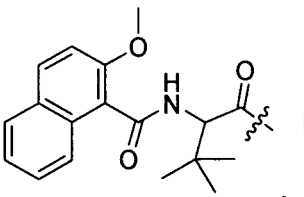
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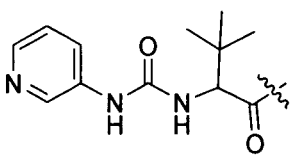
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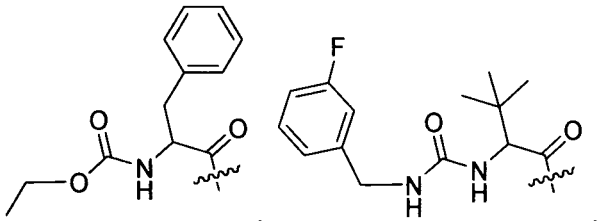
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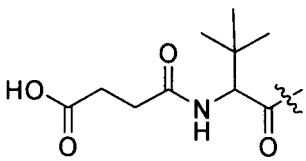
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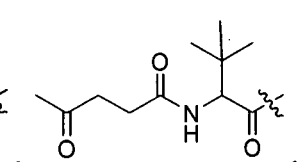
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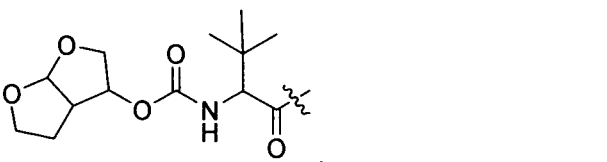
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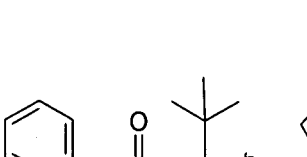
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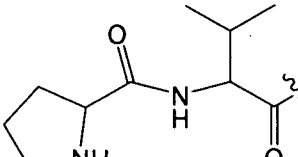
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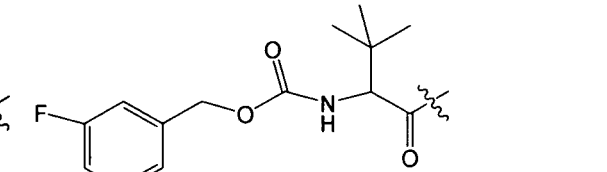
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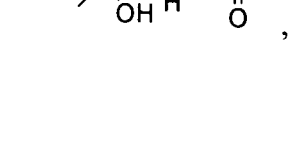
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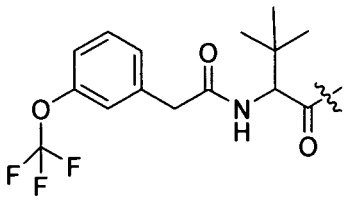
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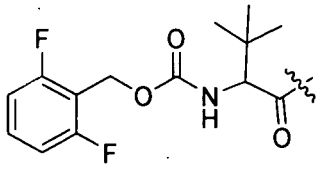
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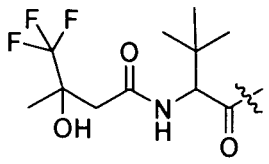
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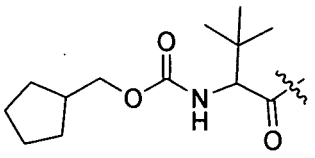
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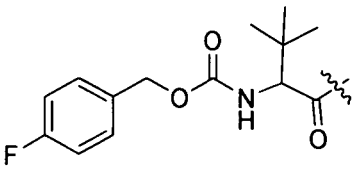
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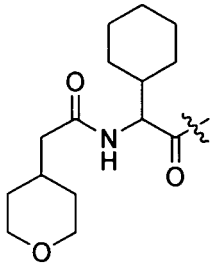
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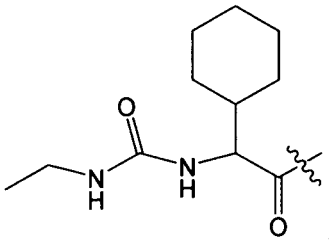
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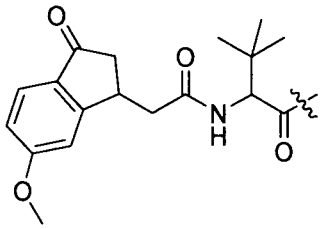
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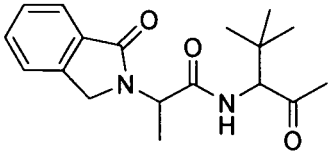
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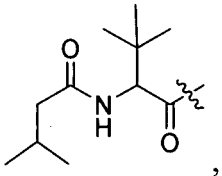
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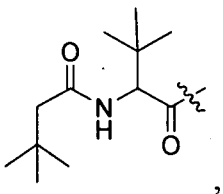
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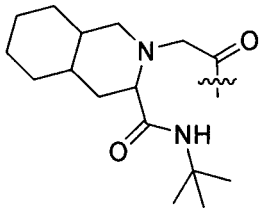
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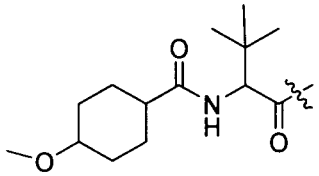
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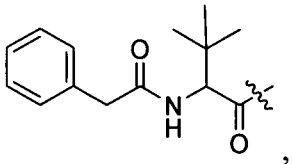
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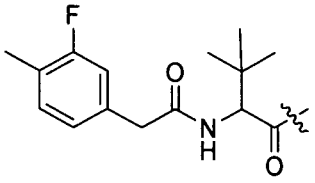
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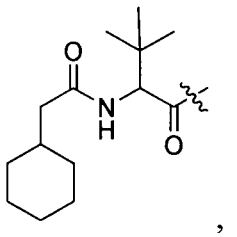
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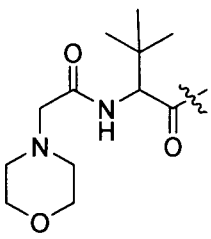
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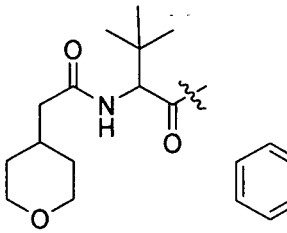
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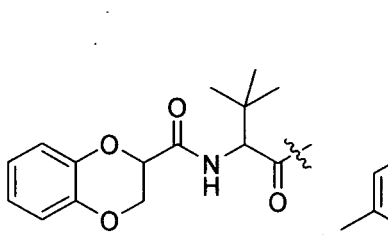
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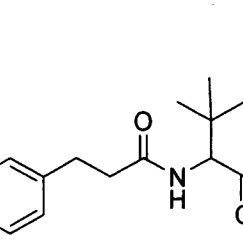
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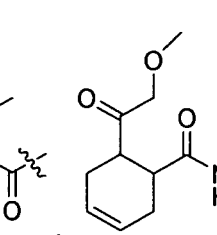
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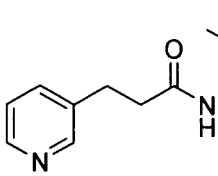
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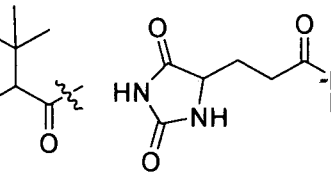
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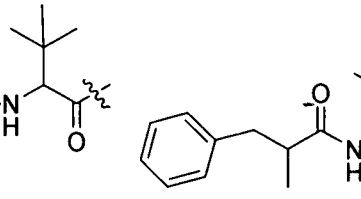
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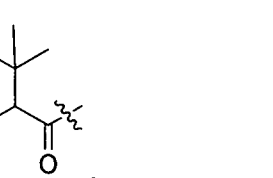
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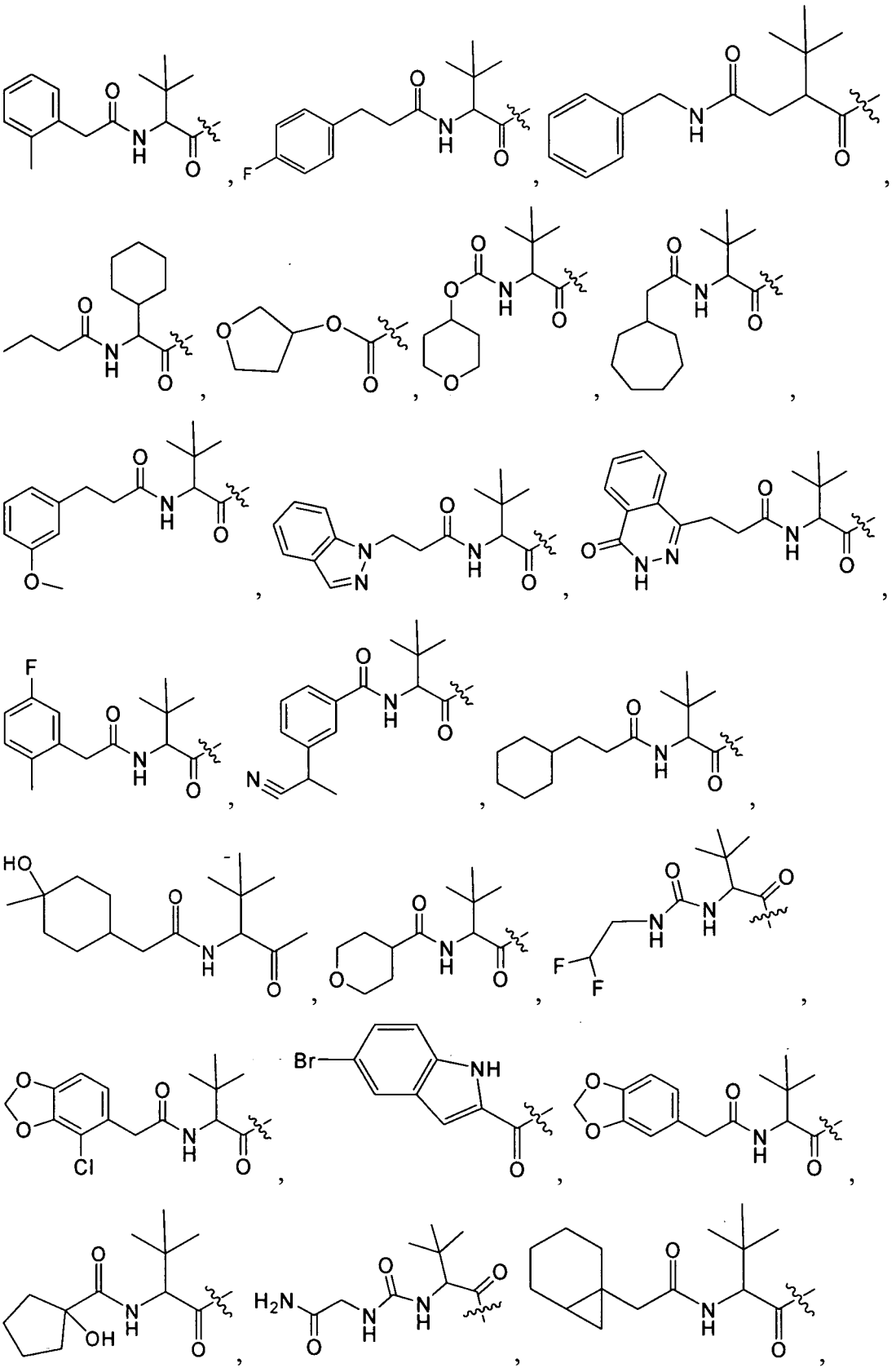

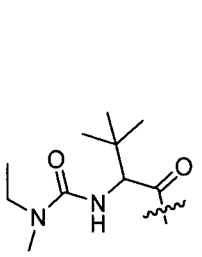
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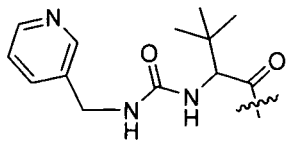
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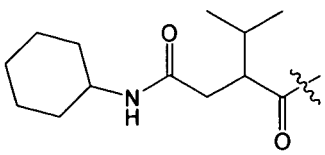
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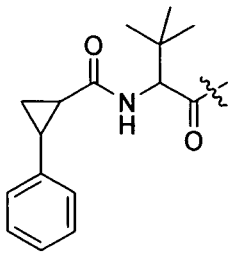
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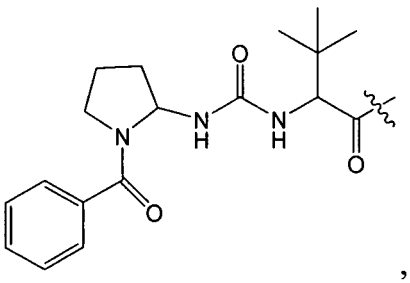
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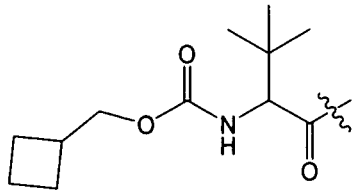
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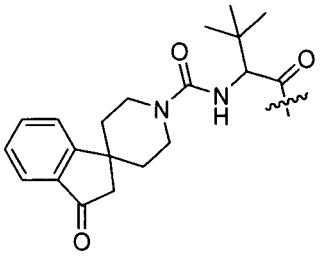
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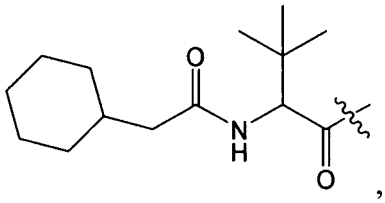
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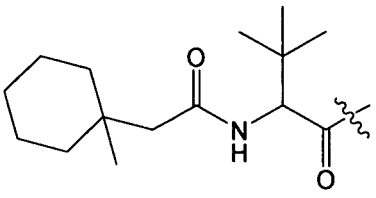
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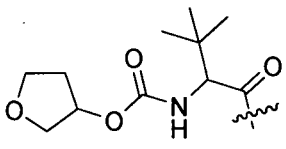
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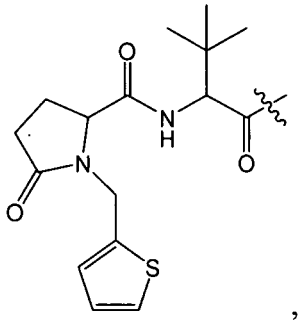
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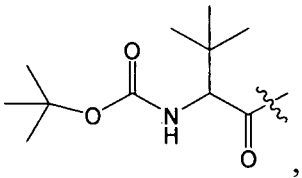
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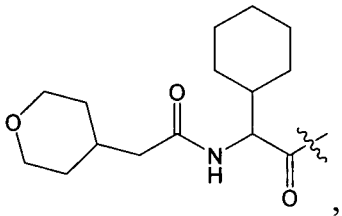
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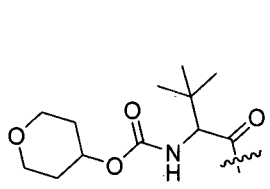
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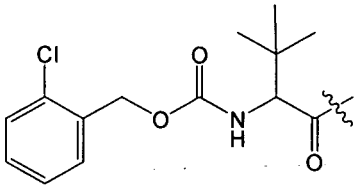
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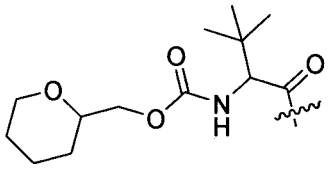
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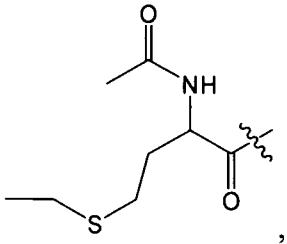
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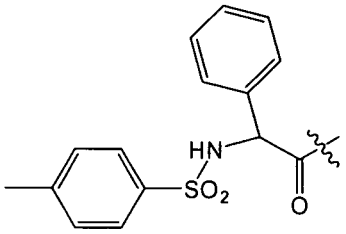
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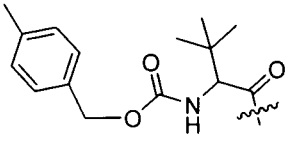
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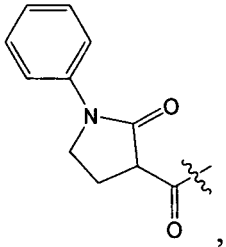
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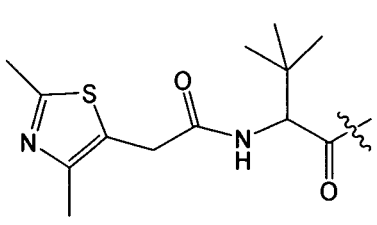
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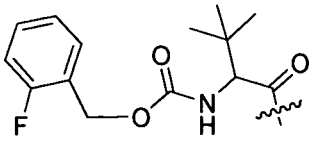
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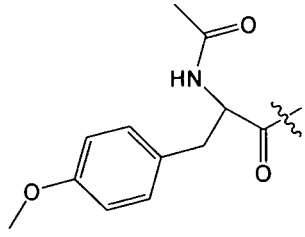
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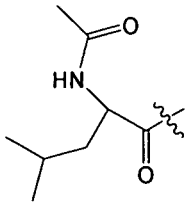
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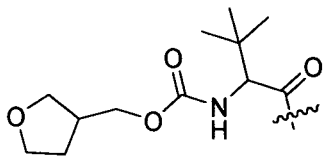
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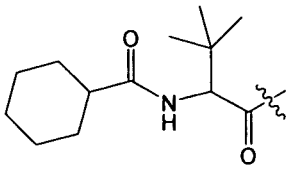
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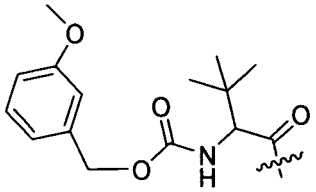
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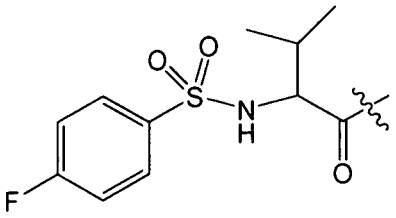
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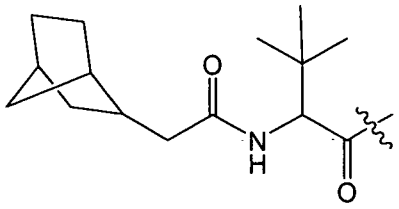
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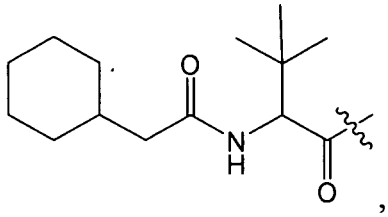
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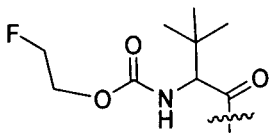
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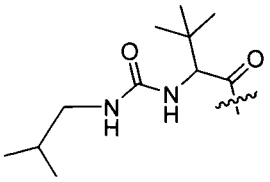
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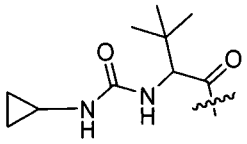
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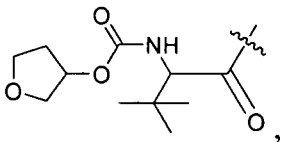
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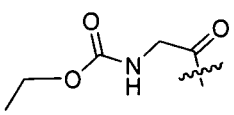
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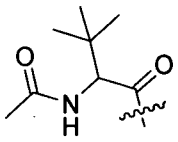
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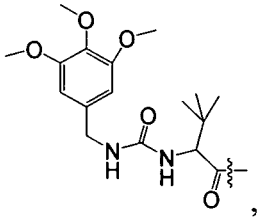
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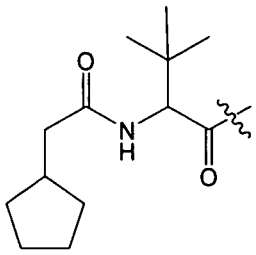
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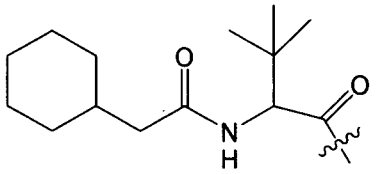
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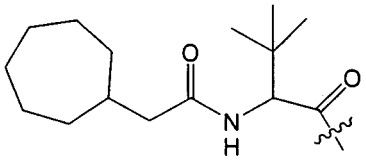
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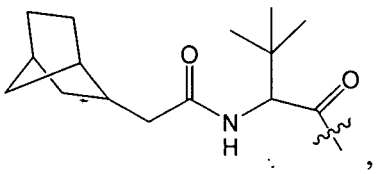
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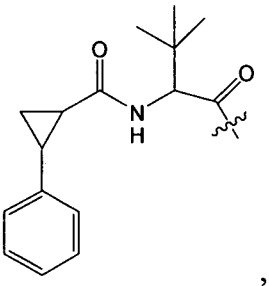
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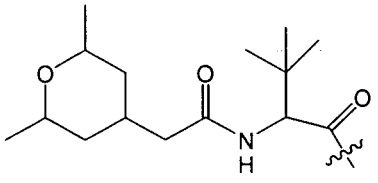
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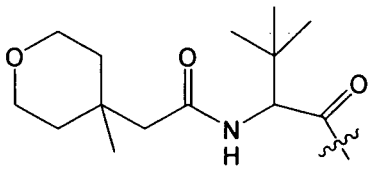
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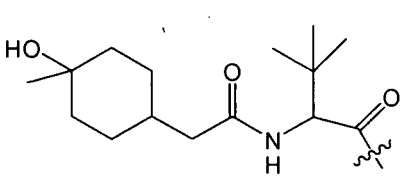
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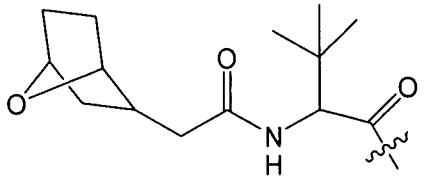
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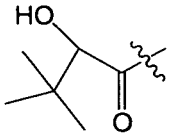
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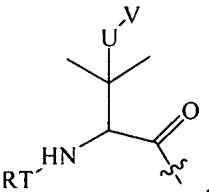
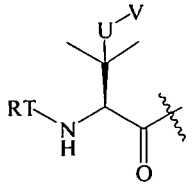
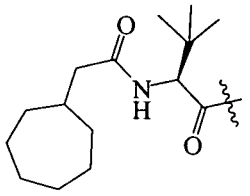
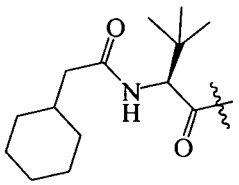
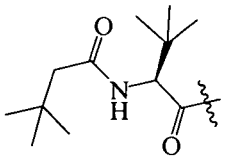
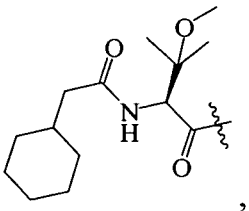
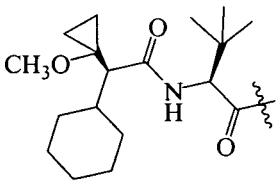
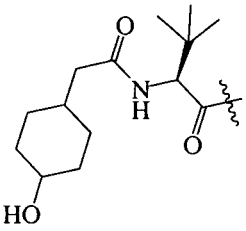
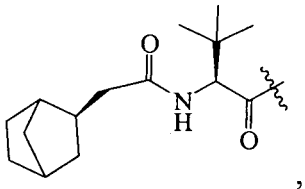
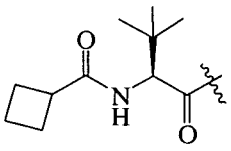
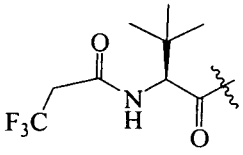
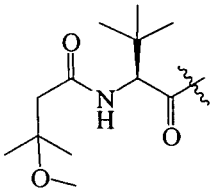
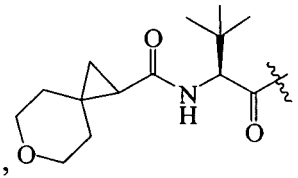
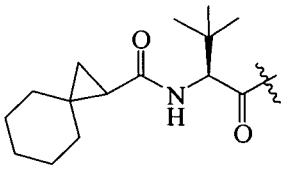
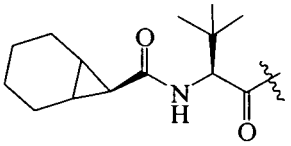
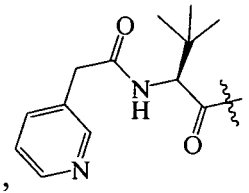
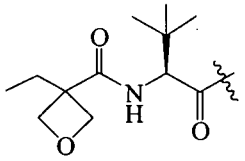
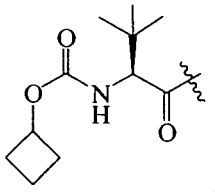
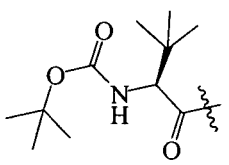
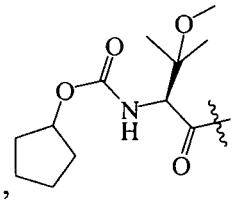
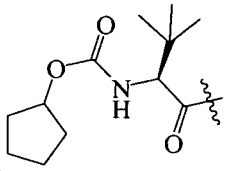
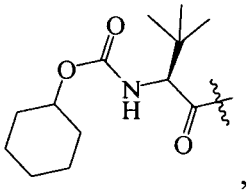
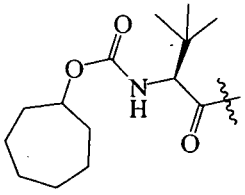
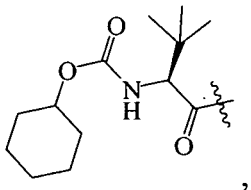
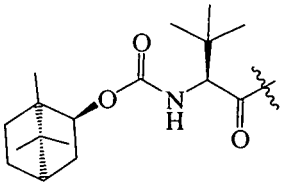
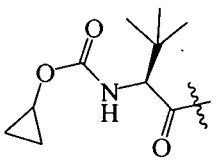


[0115] In several embodiments, R_1 is , wherein U is a bond or -O-; V is an

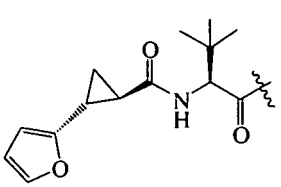
alkyl; T is -C(O)-; and R is an aliphatic, cycloaliphatic, heterocycloaliphatic, aryl, heteroaryl, alkoxy or cycloalkoxy. Examples of R_1 include  in which R, U, and V are as just defined.

[0116] In some embodiments, R is an aliphatic, cycloaliphatic or heterocycloaliphatic such that examples of R_1 include  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  , and  .

[0117] In other embodiments, R is an alkoxy or cycloalkoxy such that examples of R_1 include  ,  ,  ,  ,  ,  ,  ,  , and  .

[0118] In further embodiments, R is cyclopropyl optionally substituted with cyano, halo,

aliphatic, cycloaliphatic, heterocycloaliphatic, aryl, or heteroaryl such that R_1 is selected from the group consisting of :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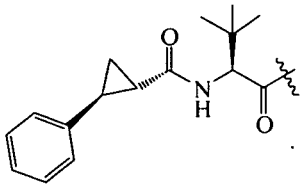
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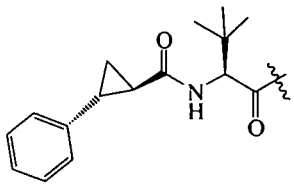
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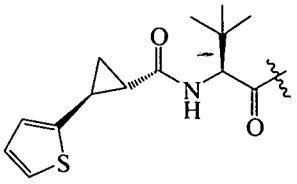
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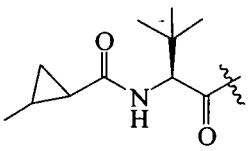
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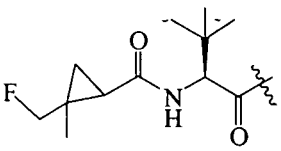
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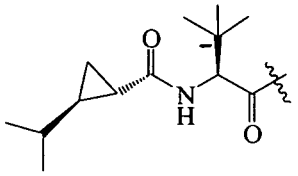
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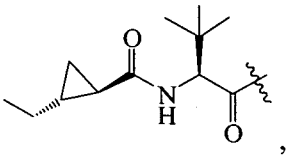
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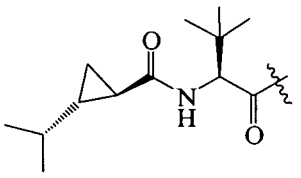
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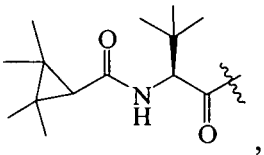
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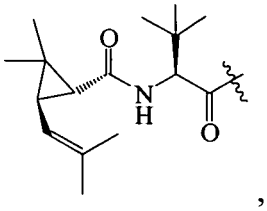
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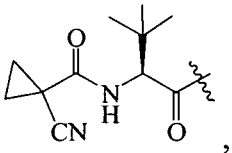
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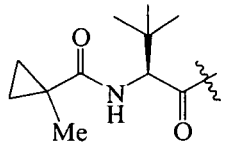
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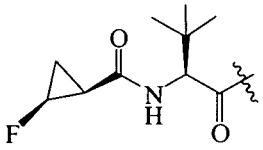
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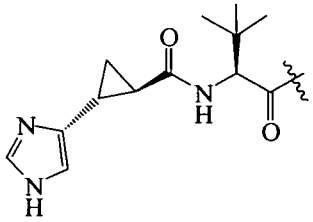
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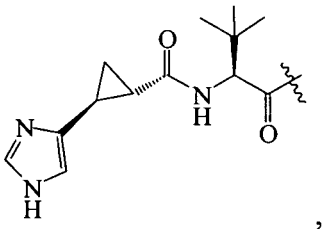
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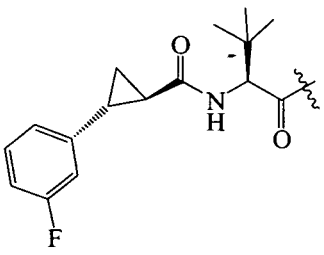
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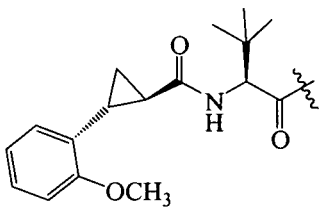
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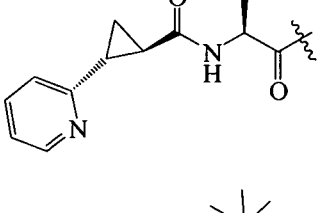
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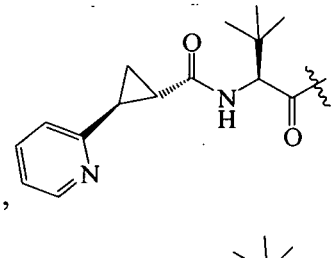
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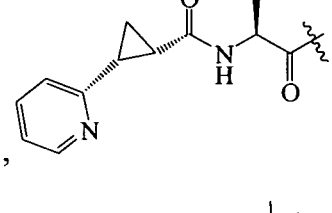
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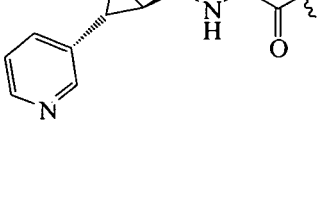
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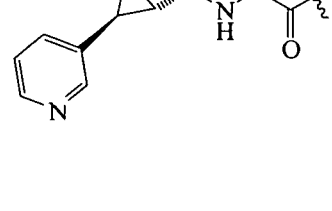
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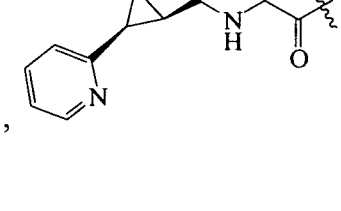
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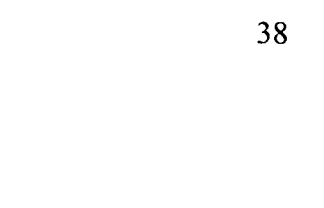
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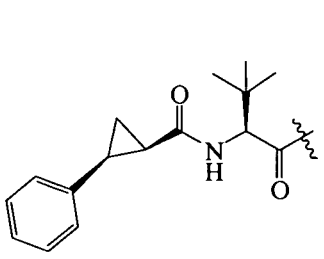
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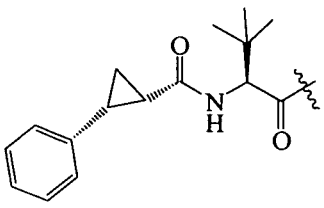
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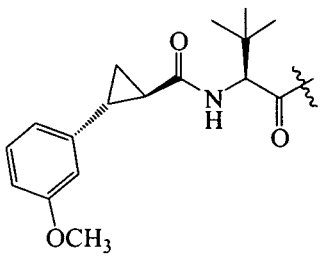
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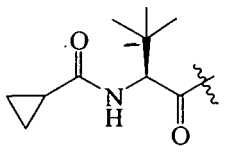
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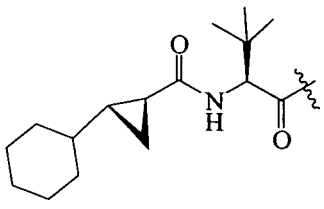
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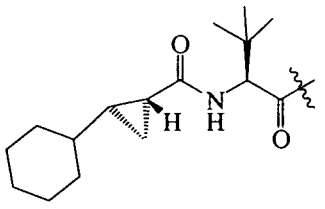
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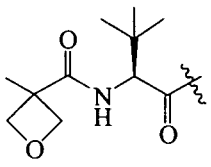
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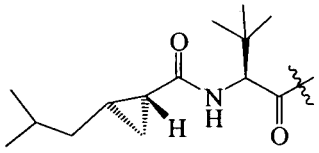
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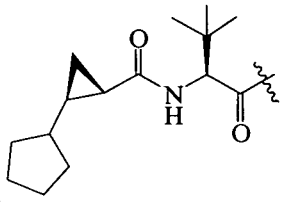
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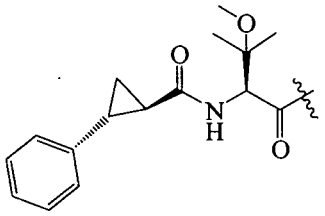
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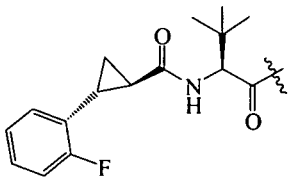
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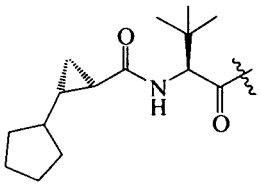
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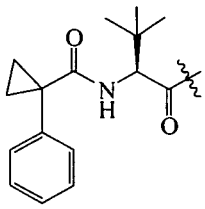
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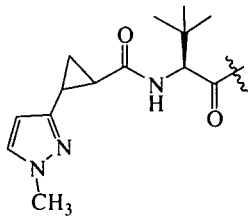
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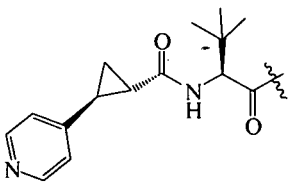
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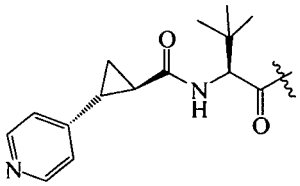
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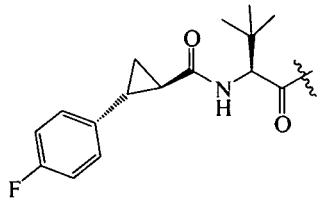
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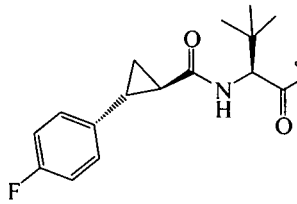
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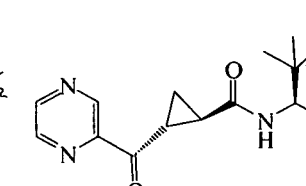
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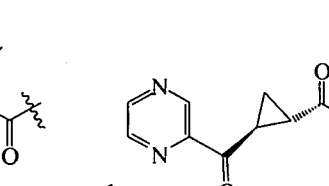
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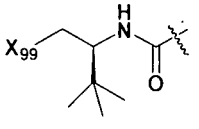
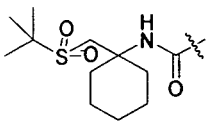
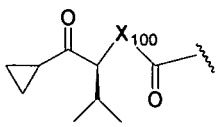
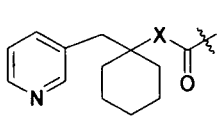
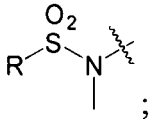
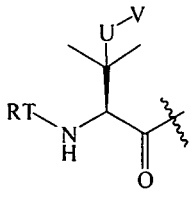
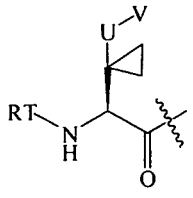
[0119] In some embodiments, R_1 is

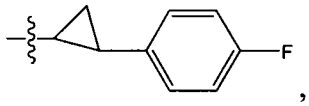





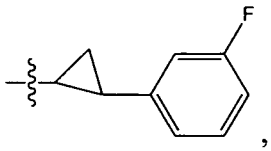
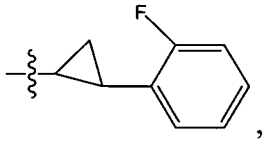
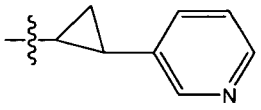
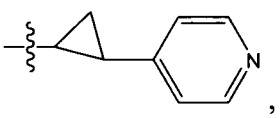
wherein X_{99} is OR, OC(NH)R, or

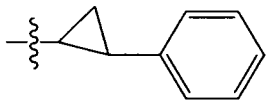
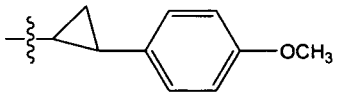
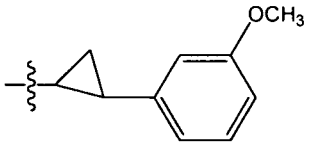

;

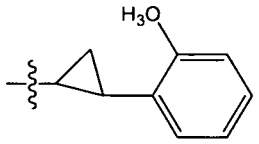
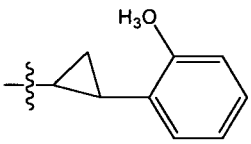
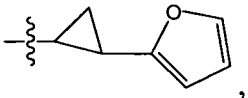
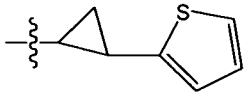
X_{100} is NH, CH_2; and R is defined above.

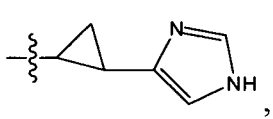
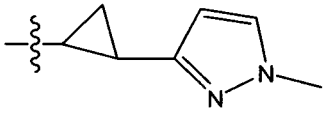
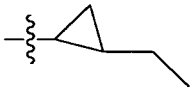
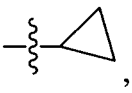
[0120] In some embodiments, R_1 is  or  , wherein U is a bond or -O-; V is an alkyl (e.g., methyl); T is -C(O)-; and R is an alkyl, cycloalkyl, heterocycloalkyl, aryl, heteroaryl, alkoxy, or (cycloalkyl)oxy, each of which is optionally substituted with 1 to 3 substituents each independently selected from the group consisting of halo, hydroxy, cyano, amino, nitro, aliphatic, haloaliphatic, (aliphatic)oxy, (halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, or heterocycloaliphatic.

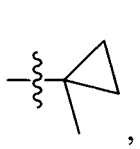
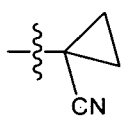
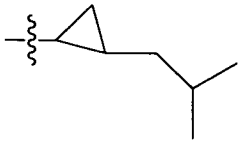
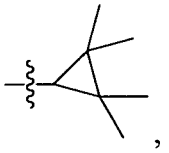
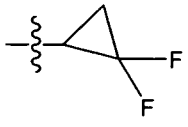
[0121] An example of R is cyclopropyl which has 1 to 4 optional substituents, each is independently aryl, heteroaryl, heterocycloalkyl, cycloalykl, alkyl, alkenyl, cyano, or halo, and attached to cyclopropyl via an optional carbonyl linker; or two of the optional substituents, together with the atom or atoms to which they are attached, form heterocycloalkyl or cycloalkyl. Specific examples of such R include  ,

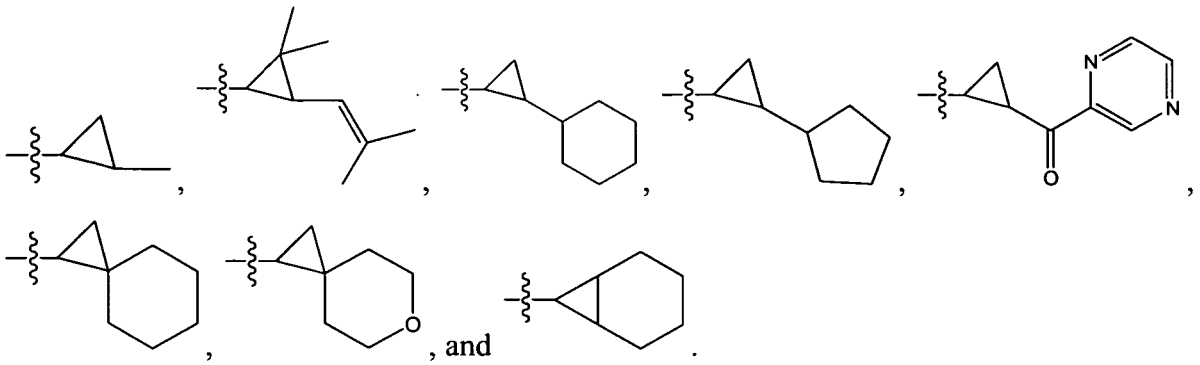
 ,  ,  ,  ,

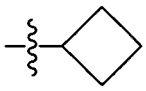
 ,  ,  ,

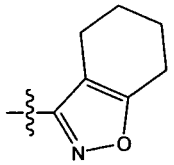
 ,  ,  ,  ,

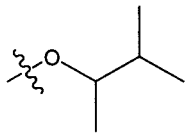
 ,  , ,  ,  ,

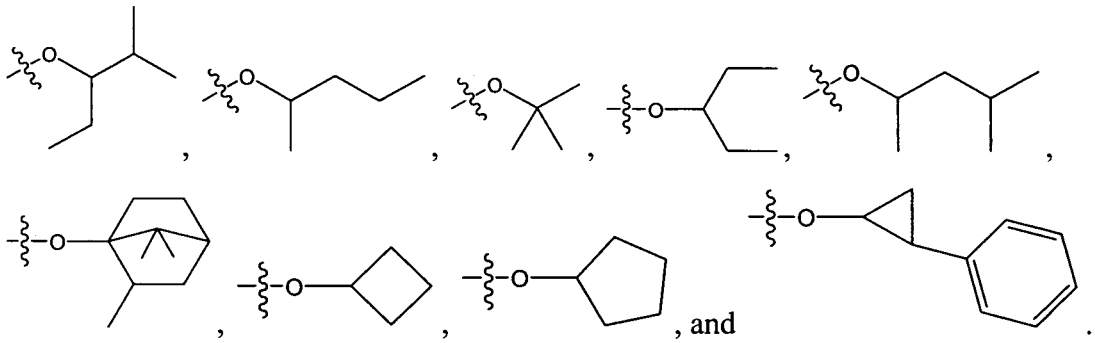
 ,  ,  ,  ,  , ,

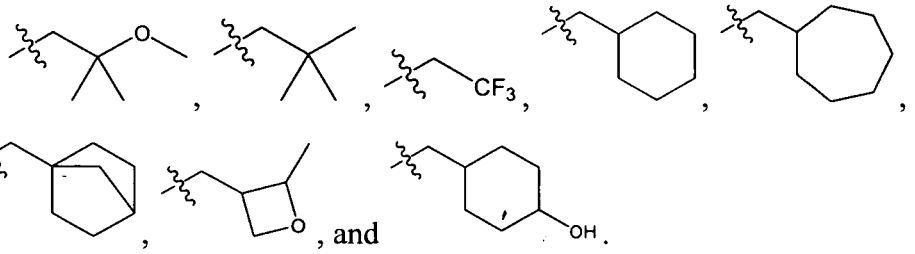


[0122] Another example of R is optionally substituted cyclobutyl (e.g., ) or optionally substituted heterocyclobutyl.

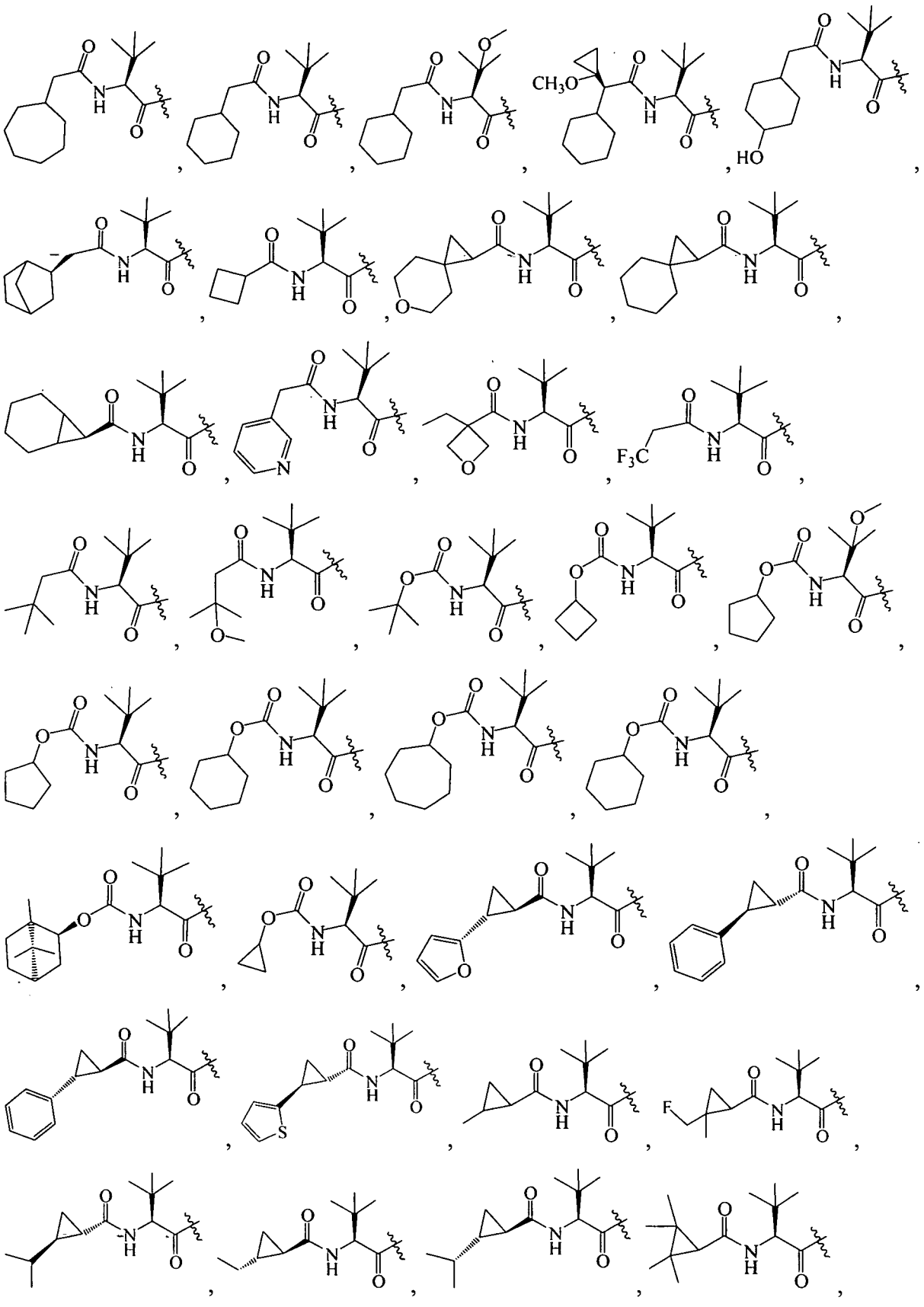
[0123] Another example of R is an optionally suibstituted heteroaryl (e.g., ).

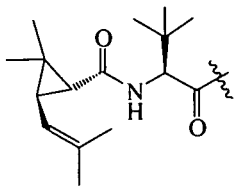
[0124] Another further example of R is an optionally substituted alkoxy or optionally substituted cycloalkyloxy. Specific examples of such R include  ,



[0125] Yet still another example of R is an optionally substituted alkyl. Specific examples of such R include 

[0126] Specific examples of R_1 for the compounds of this invention include

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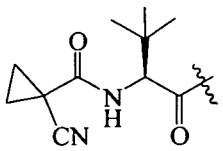
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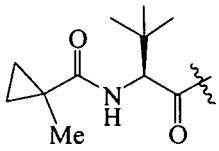
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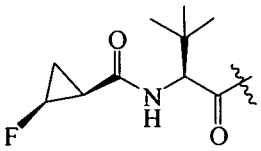
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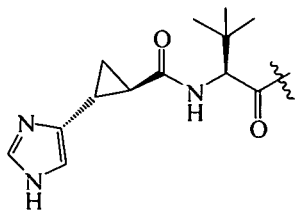
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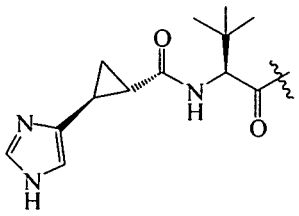
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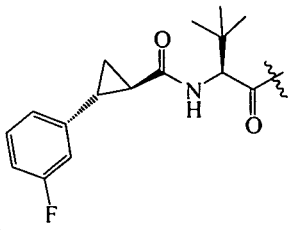
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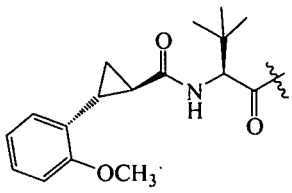
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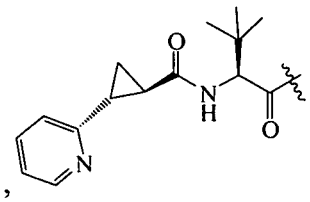
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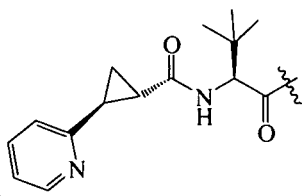
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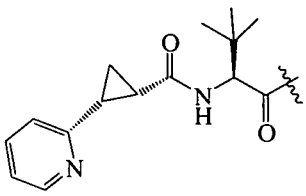
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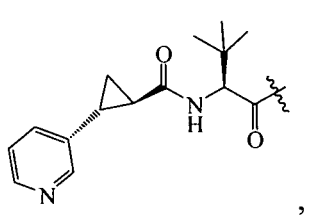
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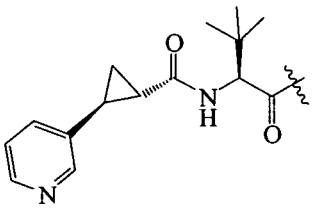
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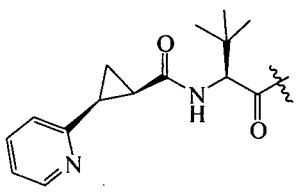
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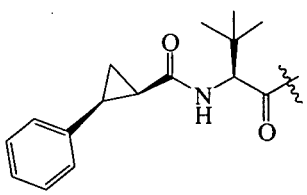
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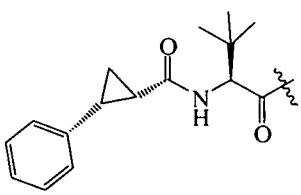
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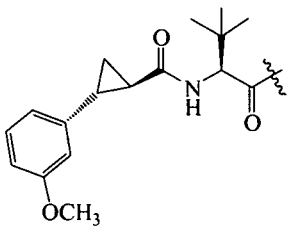
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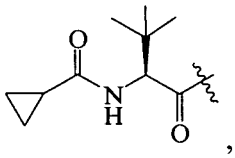
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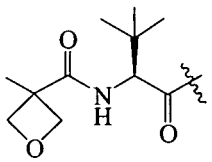
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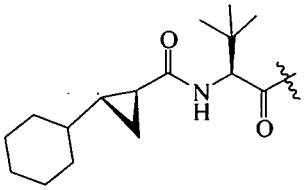
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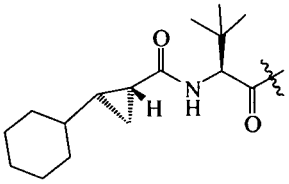
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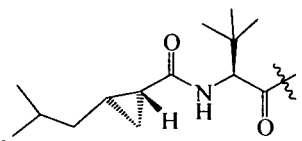
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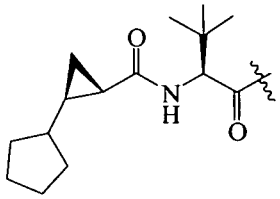
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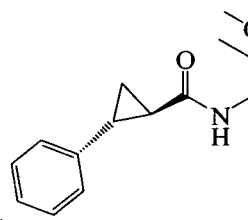
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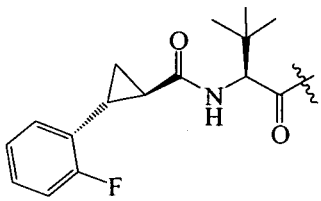
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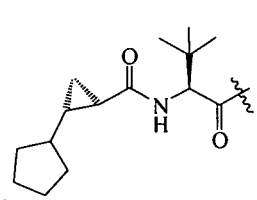
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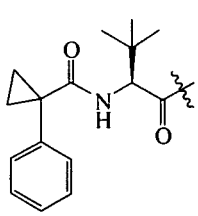
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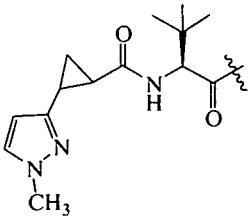
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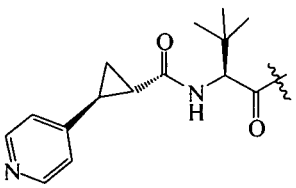
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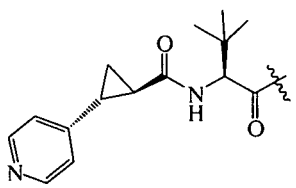
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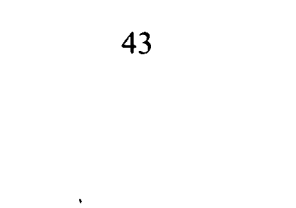
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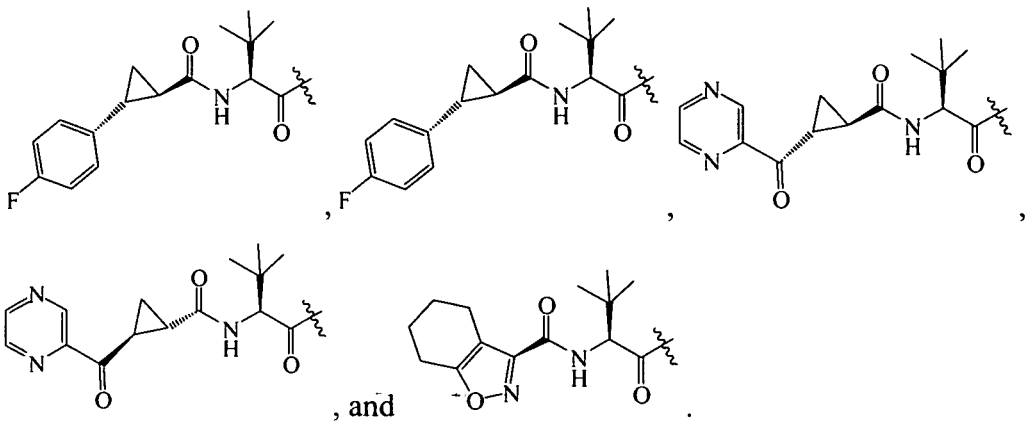


[0127] Additional examples of R_1 are illustrated in PCT publications WO 2004/103996 A1, WO 2004/72243 A2, WO 03/064456 A1, WO 03/64455 A2, WO 03/064416 A1, and U.S. Patent Publication US 2005/0090450, as well as those other publications referenced herein, each of which is incorporated in its entirety by reference.

2. **Substituent R_2:**

[0128] In several embodiments, R_2 is $-Z^BR_5$, wherein each Z^B is independently a bond or an optionally substituted branched or straight C_{1-12} aliphatic chain wherein up to three carbon units of Z^B are optionally and independently replaced by -C(O)-, -C(S)-, $-C(O)NR^B-$, $-C(O)NR^BNR^B-$, -C(O)O-, $-NR^BC(O)O-$, $-NR^BC(O)NR^B-$, $-NR^BNR^B-$, -S-, -SO-, $-SO_2-$, $-NR^B-$, $-SO_2NR^B-$, or $-NR^BSO_2NR^B-$, provided that SO, SO_2, or $-SO_2NR^B-$ is not directly bound to the carbonyl of formula (I). Each R_5 is independently R^B, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$. Each R^B is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl.

[0129] In still further embodiments, R_2 is $-Z_1-V_1-Z_2-V_2-Z_3-V_3$ each of V_1, V_2, and V_3 is independently a bond, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, an optionally substituted heteroaryl, or a hydrogen when V_1, V_2, V_3 is the terminal group of R_2; each of Z_1, Z_2, and Z_3 is independently a bond, -C(O)-, -C(O)C(O)-, -C(S)-, $-C(O)N(Q_6)-$, $-N(Q_6)C(O)-$, $-C(O)C(O)N(Q_6)-$, -O-, , SO-, $-SO_2-$, $-N(Q_6)SO_2-$, $-N(Q_6)C(O)N(Q_6)-$, $-N(Q_6)C(S)N(Q_6)-$, $-N(Q_6)-$, $-N(Q_6)SO_2-$, $-SO_2N(Q_6)-$, $-C(O)N(Q_6)SO_2-$, $-SO_2N(Q_6)C(O)-$, or hydrogen when Z_1, Z_2, or Z_3 is the terminal group of R_2; and each Q_6 is independently hydrogen, or an optionally substituted aliphatic.

[0130] In other embodiments, R_2 is an optionally substituted (aliphatic)amino wherein the aliphatic portion of R_2 is $-Z_2-V_2-Z_3-V_3$ or $-Z_3-V_3$ wherein each of Z_2 and Z_3 is independently a bond, -C(O)-, $-N(Q_5)-$, -CH(OH)-, $-C(O)N(Q_6)-$, or $-C(O)C(O)N(Q_6)-$; V_2 is independently

a bond, an optionally substituted aliphatic, or an optionally substituted cycloaliphatic; and V_3 is hydrogen, an optionally substituted aliphatic, or an optionally substituted cycloaliphatic.

[0131] In still further embodiments, Z_2 is -CH(OH)-, V_2 is a bond, and Z_3 is -C(O)N(Q_6)- such that R_2 is -N(Q_6)-CH(OH)-C(O)-N(V_3)(Q_6).

[0132] In certain embodiments, R_2 is an optionally substituted (aliphatic)amino, optionally substituted (cycloaliphatic)amino, an optionally substituted alkoxy, or hydroxy.

[0133] In still another embodiment, R_2 is an alkoxy optionally substituted with 1 to 3 of halo, hydroxy, aliphatic, cycloaliphatic, or heterocycloaliphatic.

[0134] In several embodiments, R_2 is amino. Examples of R_2 include a mono-substituted amino. Additional examples of R_2 include
(cycloaliphatic(carbonyl(carbonyl(alkyl))))amino
(amino(carbonyl(carbonyl(aliphatic))))amino,
(aliphatic(carbonyl(carbonyl(aliphatic))))amino, or
(aryl(amino(carbonyl(carbonyl(aliphatic)))))amino, each of which is optionally substituted with 1 to 3 substituents.

[0135] In several embodiments, R_2 is -$NR_{2Z}R'_{2Z}$, -SR_{2Y}, or -NR_{2Y}-$CR_{2X}R'_{2X}$-L_1-NR_{2Z}-R_{2W}, wherein R_{2Y} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; each R_{2W} is independently hydrogen, optionally substituted aliphatic, optionally substituted aryl, optionally substituted heteroaryl, optionally substituted heterocycloaliphatic, or optionally substituted cycloaliphatic; each R_{2X} and R'_{2X} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted heteroaryl, an optionally substituted phenyl, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic; or R_{2X} and R'_{2X} together with the atom to which they are both attached form an optionally substituted 3 to 7 membered cycloaliphatic or heterocycloaliphatic ring; each L_1 is -CH_2-, -C(O)-, -CF_2-, -C(O)C(O)-, -C(O)O-, -S(O)-, or -SO_2-; each R_{2Z} or R'_{2Z} is hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or R_{2Z} and R'_{2Z} together with the nitrogen to which they are both attached may form an optionally substituted 3 to 7 membered heterocycloaliphatic ring.

[0136] In several embodiments, each R_{2X} and R'_{2X} is independently hydrogen, or optionally substituted aliphatic, optionally substituted cycloaliphatic, or optionally substituted (cycloaliphatic)aliphatic.

[0137] In several embodiments, L_1 is -C(O)C(O)- or -SO_2-.

[0138] In several other embodiments, each R_{2W} is hydrogen or optionally substituted cycloaliphatic.

[0139] In several embodiments, R_2 is -NH-CHR_{2X}-C(O)-C(O)-$N(R_{2Z})R_{2W}$.

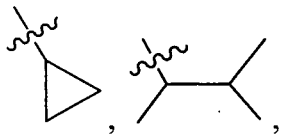
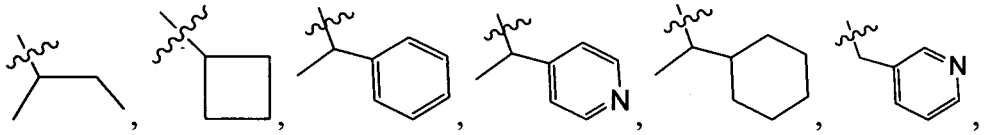
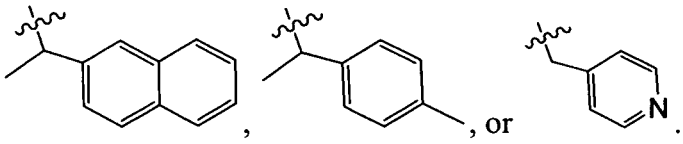
[0140] In several embodiments, R_2 is -NH-CHR_{2X}-CH(OH)-C(O)-$N(R_{2Z})R_{2W}$.

[0141] In several embodiments, R_2 is -NH-CHR_{2X}-C(O)-C(O)-NHR_{2Z} wherein –NHR_{2Z} is NH-cyclopropyl, -NH-Me, -NH-Et, -NH-iPr, -NH-nPr.

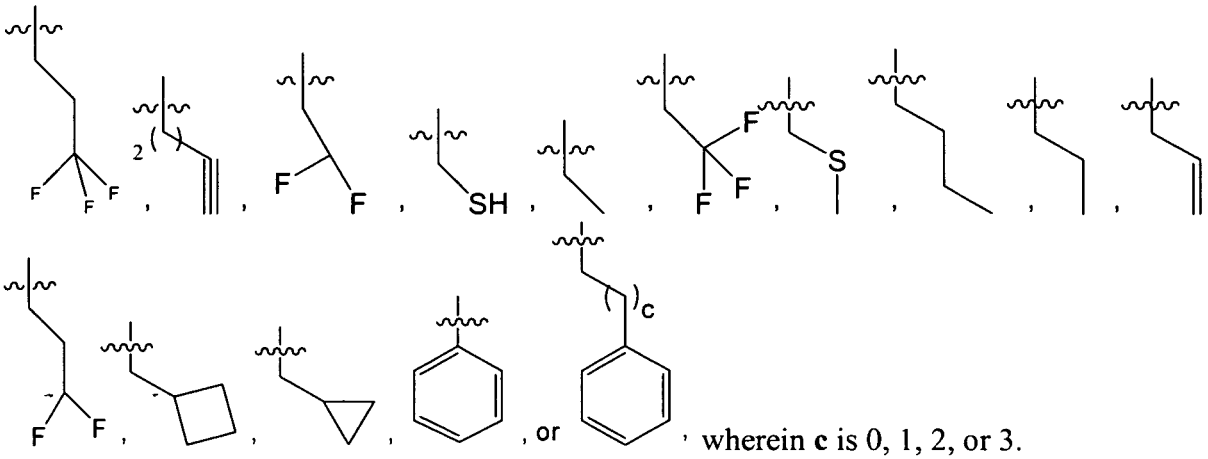
[0142] In several embodiments R_2 is -$NR_{2Z}R'_{2Z}$, -SR_{2Z} wherein each R_{2Z} and R'_{2Z} is independently hydrogen, alkyl, cycloalkyl or aralkyl. Non-limiting examples of R_{2Z} include methyl, ethyl, *t*-butyl, cyclopentyl, cyclohexyl and benzyl.

[0143] In other embodiments R_2 is (-NH-$CR_{2X}R'_{2X}$- L_1-C(O))$_i$-M; wherein each M is independently -OH, R_{2X}, -$NR_{2Z}R'_{2Z}$, or -OR_{2X}, each i is 1 or 2, and L_1, R_{2Z}, R_{2X}, and R'_{2Z} are defined above.

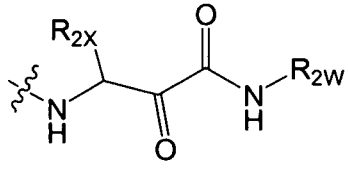
[0144] In several embodiments R_2 is (-NH-$CR_{2Z}R'_{2Z}$- L_1-C(O))$_i$-M wherein L_1 is -C(O)-, i is 1, and M is independently R_{2X}, -$N(R_{2X}R'_{2X})$, -OR_{2X}, -$NHSO_2R_{2X}$, or -SR_{2X}.

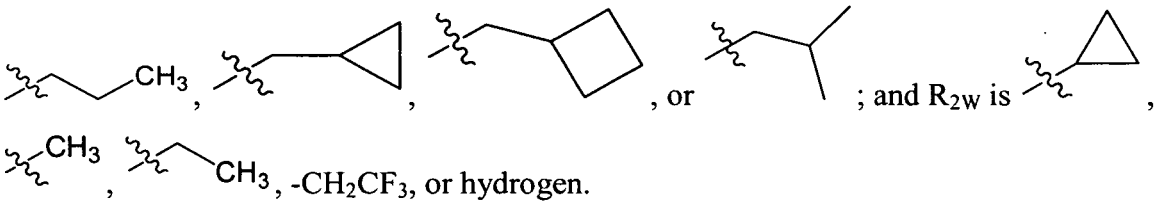
[0145] In some embodiments, R'_{2Z} is H and R_{2Z} is an aliphatic, (aryl)aliphatic or cycloaliphatic. Non-limiting examples of R_{2X} include hydrogen,  ,  ,  , or .

[0146] In some embodiments, R'_{2X} is H and R_{2X} is an optionally substituted aliphatic, optionally substituted aryl, optionally substituted aralkyl, optionally substituted heteroaliphatic or optionally substituted heteroaralkyl. Some non-limiting examples of R_{2X} include



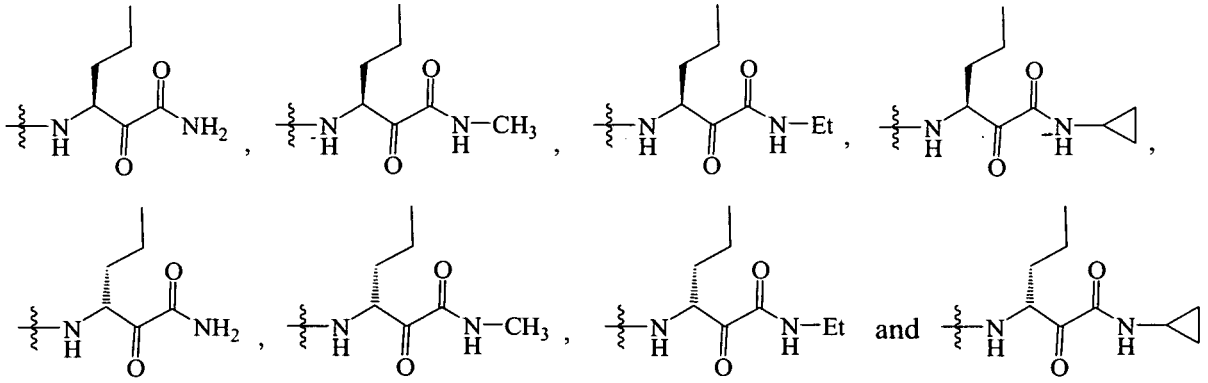
wherein **c** is 0, 1, 2, or 3.

[0147] In several embodiments, R_2 is  , wherein R_{2X} is

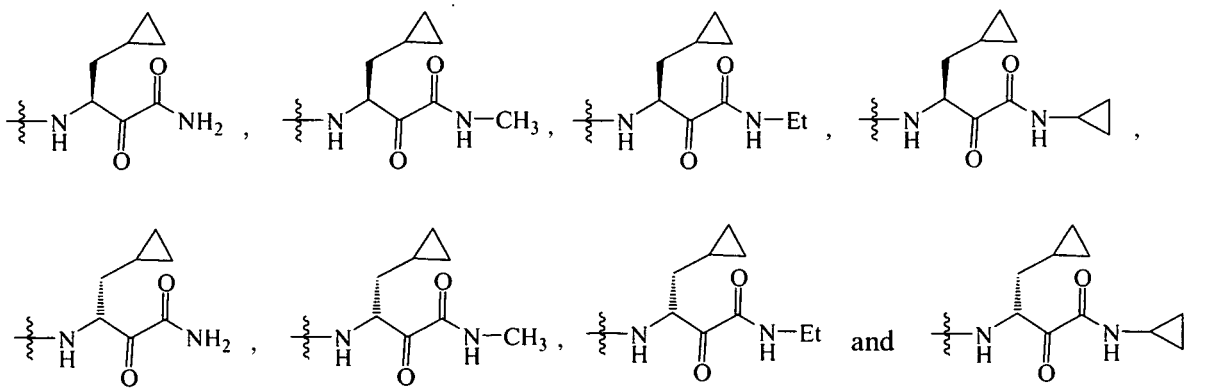


; and R_{2W} is , , , $-CH_2CF_3$, or hydrogen.

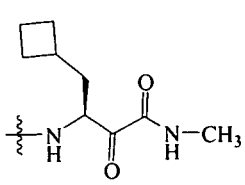
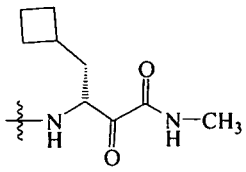
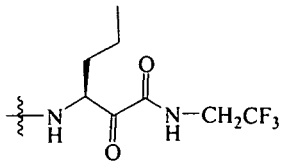
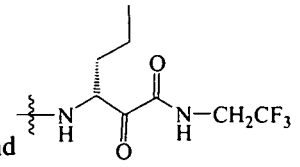
[0148] In some embodiments, R_2 is selected from the group consisting of:

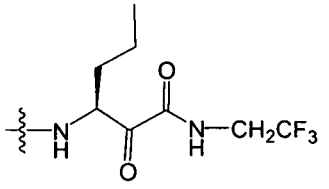
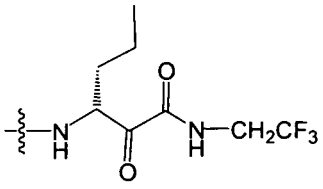


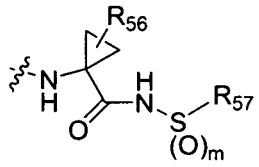
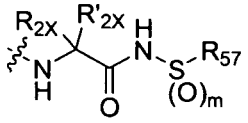
[0149] In some embodiments, R_2 is selected from the group consisting of:

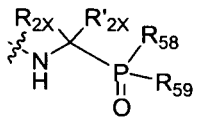


[0150] In some embodiments, R_2 is selected from the group consisting of:

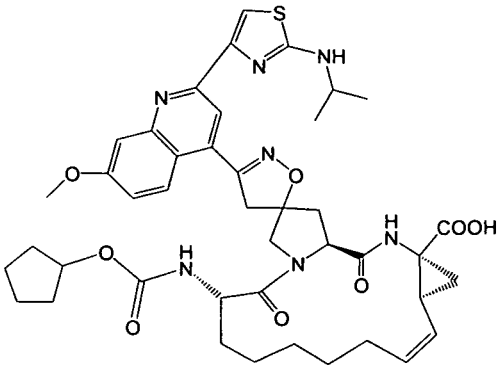
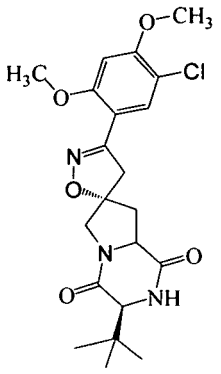
 ,  ,  and 

[0151] In some embodiments, R_2 is  or  .

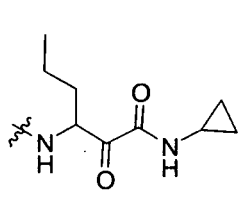
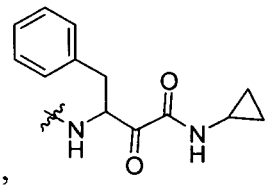
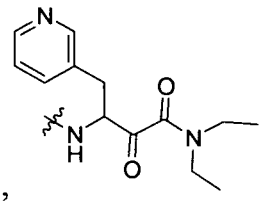
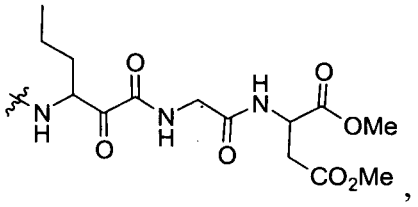
[0152] In some embodiments, R_2 is  or  , wherein each R_{56} is independently optionally substituted C_{1-6} aliphatic; optionally substituted aryl, optionally substituted heteraryl, optionally substituted cycloaliphatic, or optionally substituted heterocycloaliphatic; each R_{57} is independently optionally substituted aliphatic, optionally substituted aryl, optionally substituted aliphatic, optionally substituted heteroaryl, optionally substituted aliphatic, optionally substituted cycloaliphatic or optionally substituted amino; and m is 1 or 2; and each R_{2X} and R'_{2X} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or R_{2X} and R'_{2X} together with the atom to which they are both attached form an optionally substituted 3 to 7 membered cycloaliphatic or heterocycloaliphatic ring.

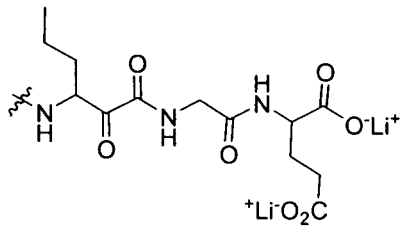
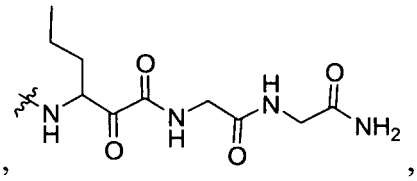
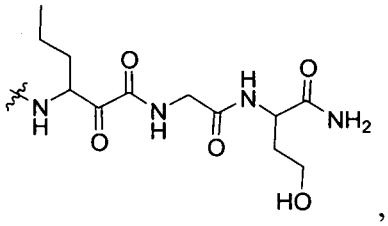
[0153] In some other embodiments, R_2 is  , wherein R_{58} and R_{59} are each independently selected from optionally substituted aliphatic, optionally substituted alkoxy, optionally substituted aryloxy, optionally substituted heteroaryloxy, optionally substituted (cycloaliphatic)oxy, optionally substituted (heterocycloaliphatic)oxy optionally substituted aryl, optionally substituted heteroaryl, optionally substituted cycloaliphatic or optionally substituted amino; and each R_{2X} and R'_{2X} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; or R_{2X} and R'_{2X} together with the atom to which they are both attached form an optionally substituted 3 to 7 membered cycloaliphatic or heterocycloaliphatic ring.

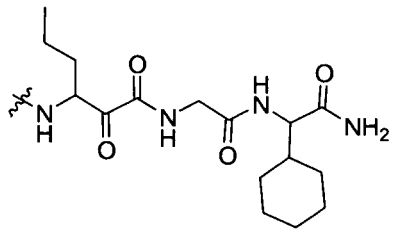
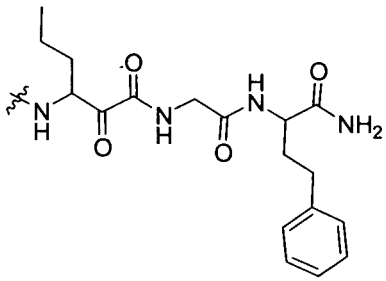
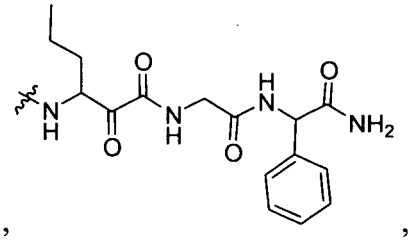
[0154] In several embodiments, a portion of R_1 and a portion of R_2 together can form a cyclic structure, e.g., a 5- to 18-membered cycloheteroaliphatic ring (e.g., an optionally

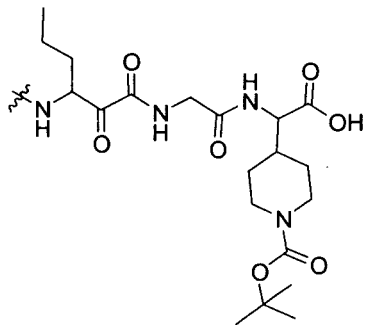
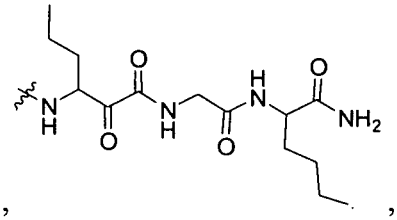
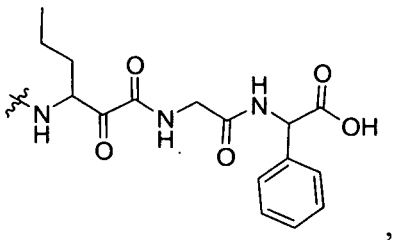
substituted 6-membered heterocycloalkyl ring). Some non-limiting examples of compounds thus formed include  and  .

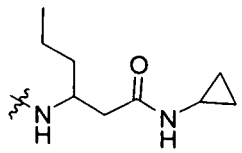
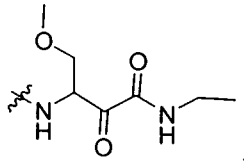
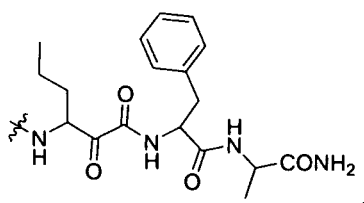
[0155] In several embodiments, R_2 is one selected from

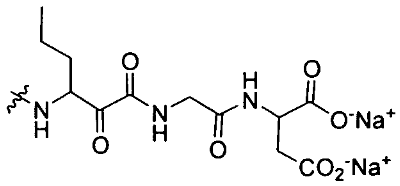
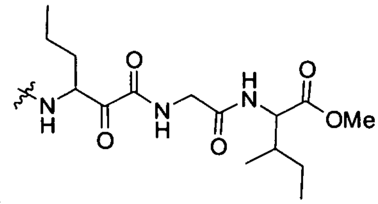
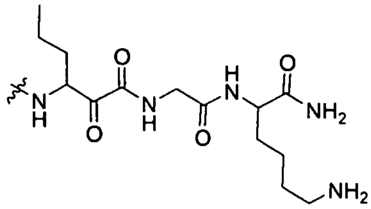
 ,  ,  ,  ,

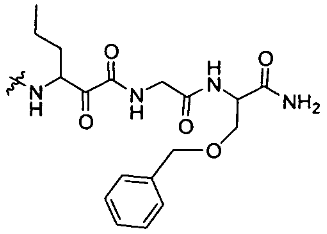
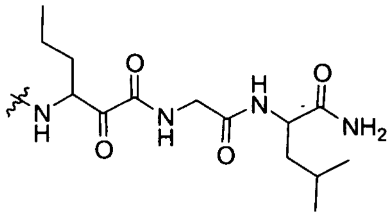
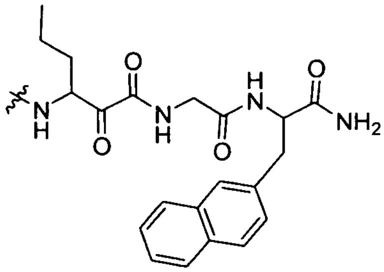
 ,  ,  ,

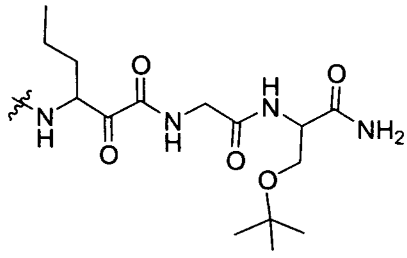
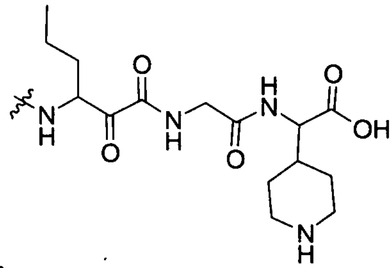
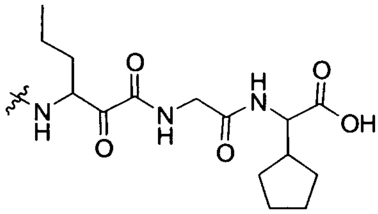
 ,  ,  ,

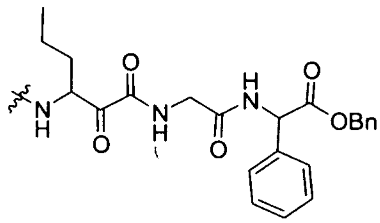
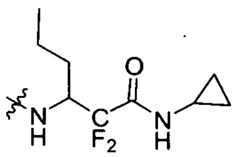
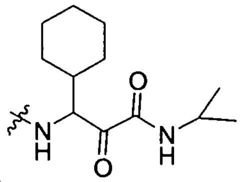
 ,  ,  ,

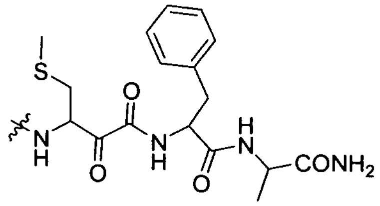
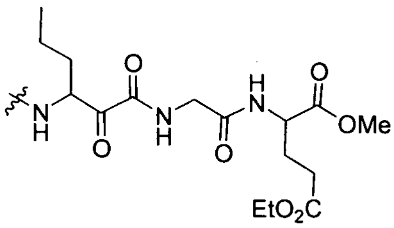
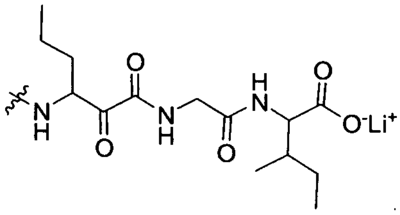
 ,  ,  .

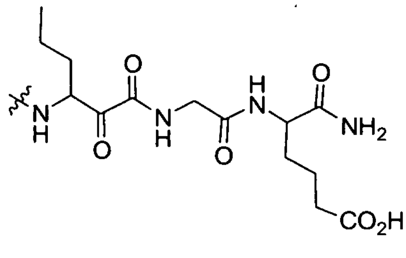
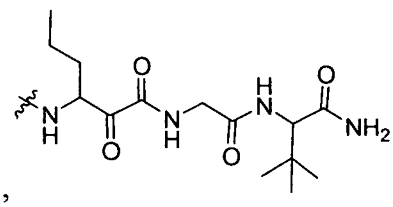
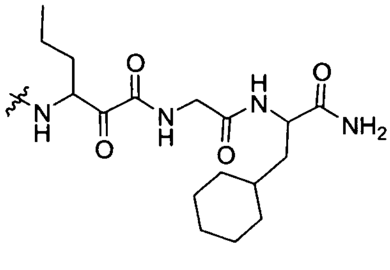
 ,
 ,
 ,

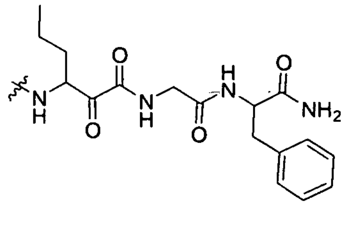
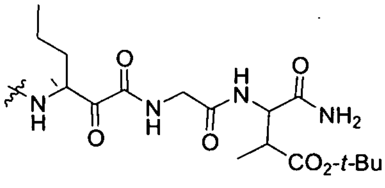
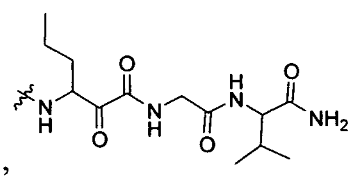
 ,
 ,
 ,

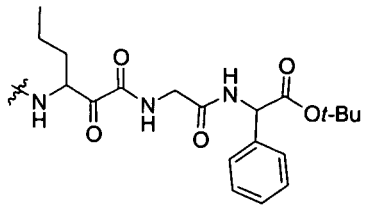
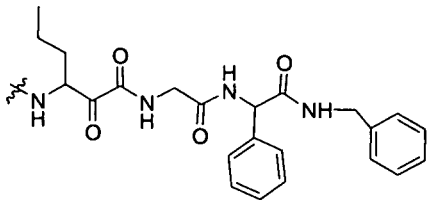
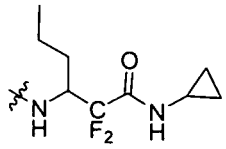
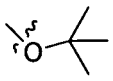
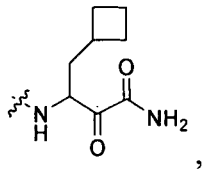
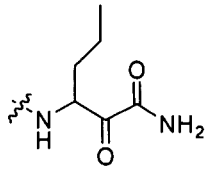
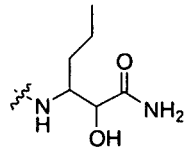
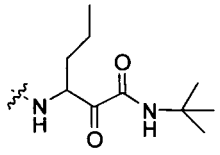
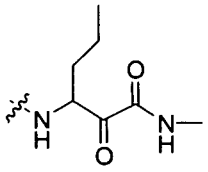
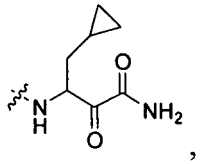
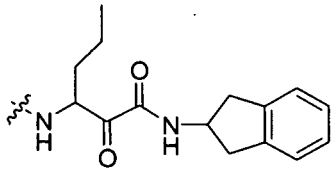
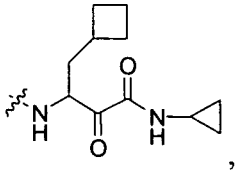
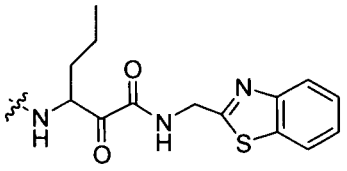
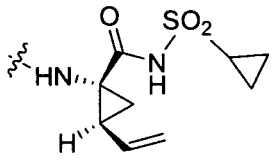
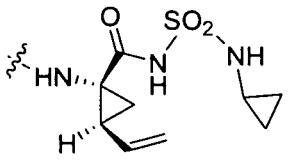
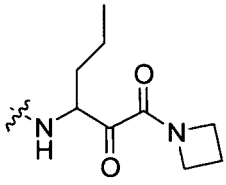
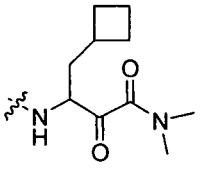
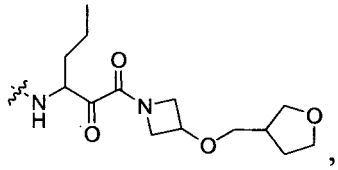
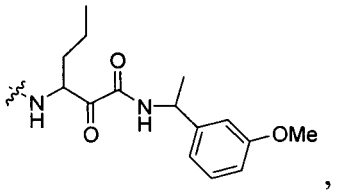
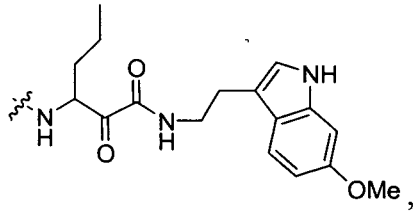
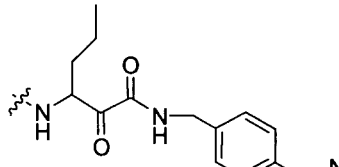
 ,
 ,
 ,

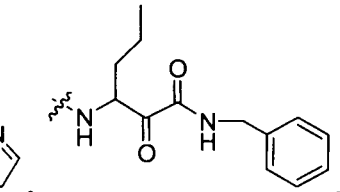
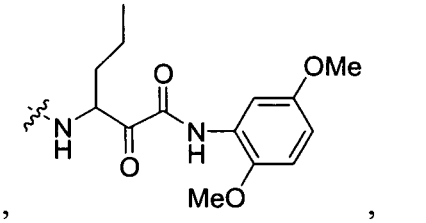
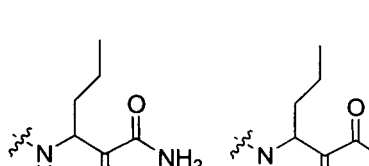
 ,
 ,
 ,

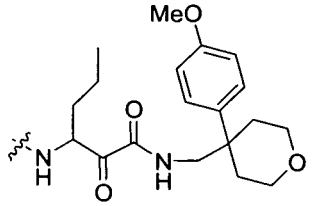
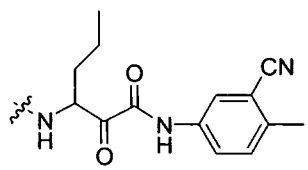
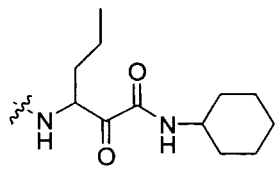
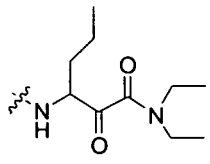
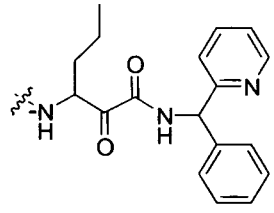
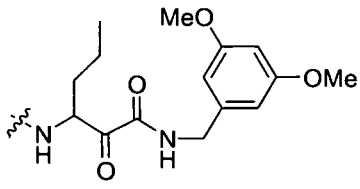
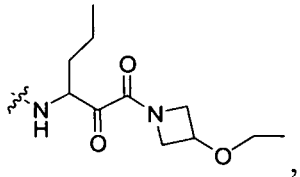
 ,
 ,
 ,

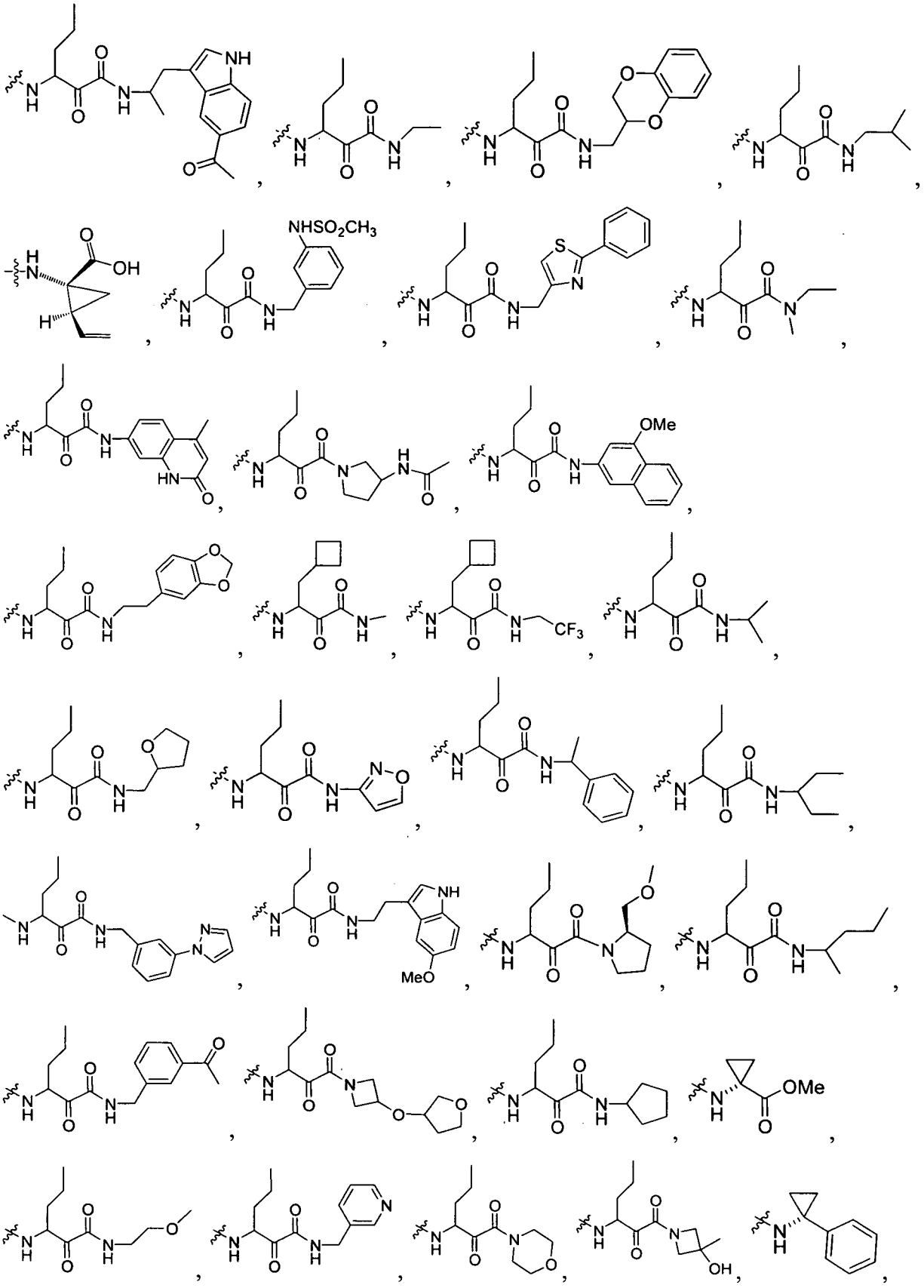
 ,
 ,
 ,

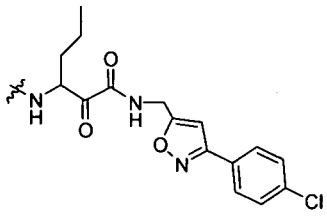
 ,
 ,
 ,

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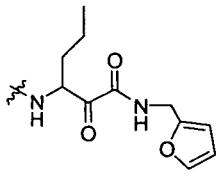
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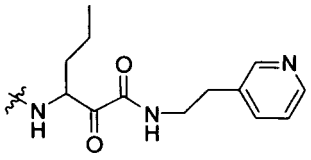
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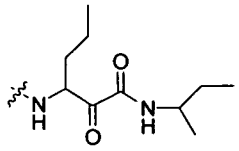
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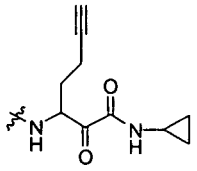
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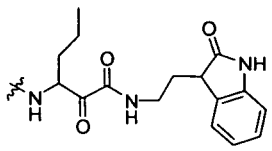
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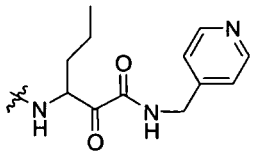
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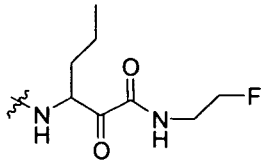
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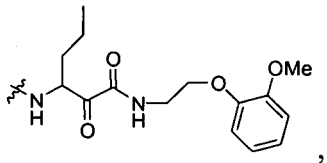
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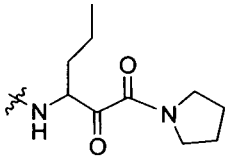
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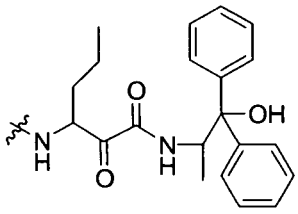
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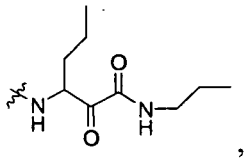
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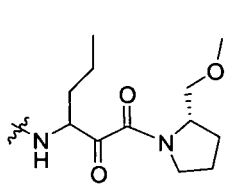
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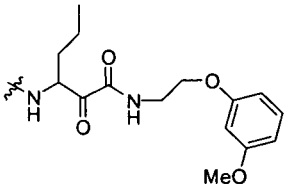
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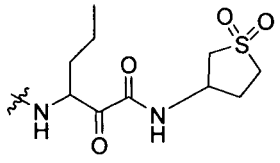
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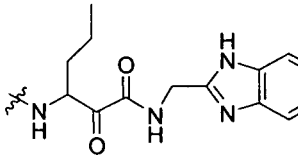
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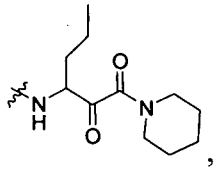
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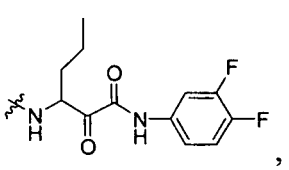
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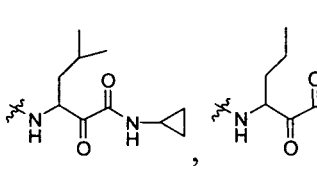
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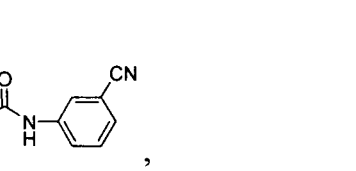
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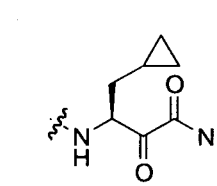
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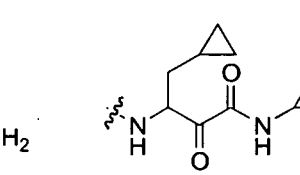
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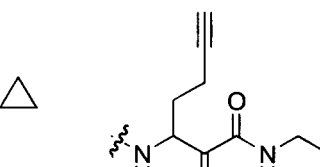
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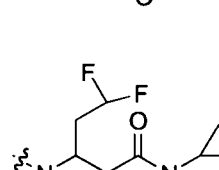
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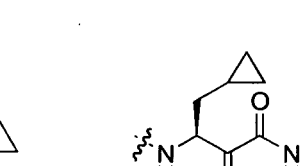
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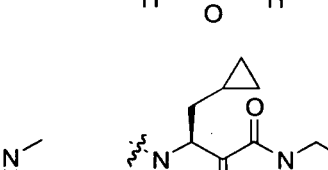
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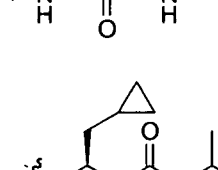
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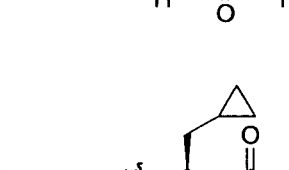
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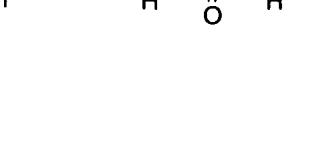
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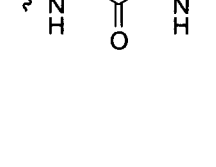
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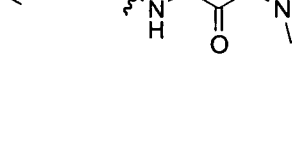
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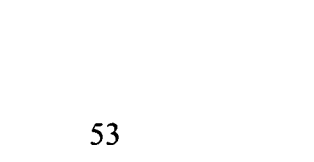
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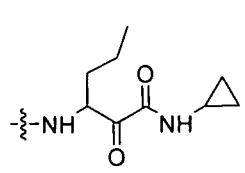
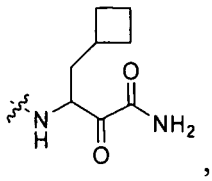
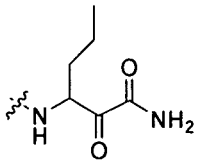
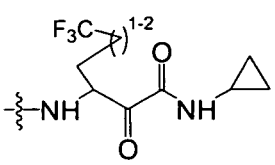
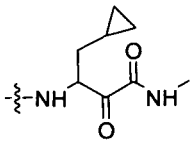
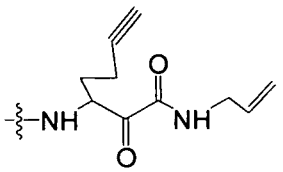
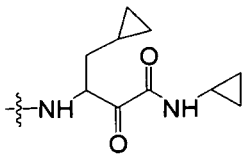
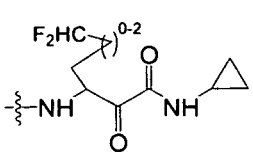
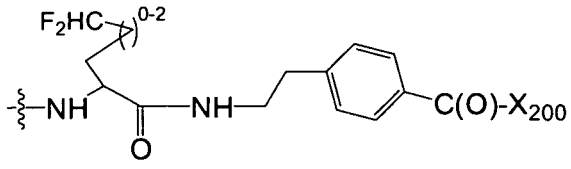
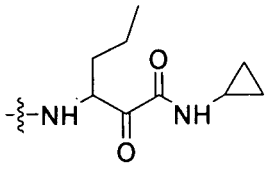
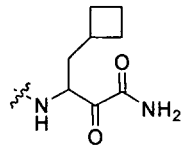
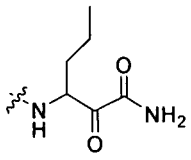
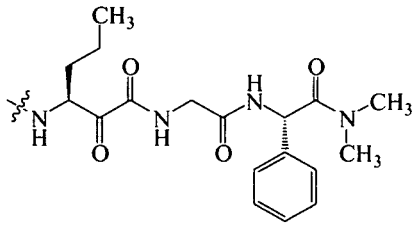
[0156] In some specific embodiments, R_2 is  ,  ,  ,  ,  ,  ,  ,  , or  ,

where X_{200} is $-OX_{202}$ or $-X_{202}$, and X_{202} is aliphatic, cycloaliphatic, heterocycloaliphatic, aryl, or heteroaryl.

[0157] In other embodiments, R_2 is  ,  , or  .

[0158] In some other embodiments, R_2 is  .

[0159] Additional examples of R_2 are illustrated in PCT publications WO 2004/103996 A1, WO 2004/72243 A2, WO 03/064456 A1, WO 03/64455 A2, WO 03/064416 A1, and U.S. Patent Publication US 2005/0090450, as well as those other publications referenced herein, each of which is incorporated in its entirety by reference.

3. Substituent R_3:

[0160] Each R_3 is an aliphatic, a cycloaliphatic, a heterocycloaliphatic, an aryl, or a heteroaryl, each of which is optionally substituted.

[0161] In several embodiments, each R_3 is independently $-Z^CR_6$, wherein each Z^C is independently a bond or an optionally substituted aliphatic wherein up to two carbon units of Z^C are optionally and independently replaced by -C(O)-, -CS-, $-C(O)NR^C-$, $-C(O)NR^CNR^C-$, -C(O)O-, $-NR^CC(O)O-$, -O-, $-NR^CC(O)NR^c-$, $-NR^cNR^c-$, -S-, -SO-, $-SO_2-$, $-NR^C-$, $-SO_2NR^C-$, or $-NR^CSO_2NR^C-$. Each R_6 is independently R^C, halo, -OH, -CN, $-NO_2$, $-NH_2$, alkoxy, or haloalkoxy. Each R^C is independently hydrogen, an optionally substituted aliphatic group, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an

optionally substituted aryl, or an optionally substituted heteroaryl. However, in many embodiments, when Z^C is a bond and R_6 is R^C, then R^C is independently an optionally substituted aliphatic group, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl.

[0162] In still other embodiments, each R_3 is an optionally substituted aliphatic, amino, sulfonyl, sulfanyl, sulfinyl, sulfonamide, sulfamide, sulfo, -O-R_{3A}, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; and each R_{3A} is independently an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl.

[0163] In some embodiment, R_3 is an optionally substituted amino, -O-R_{3A}, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; and R_{3A} is an optionally substituted aryl or an optionally substituted heteroaryl;

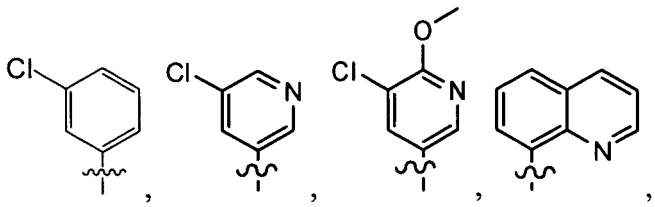
[0164] In several embodiments, R_3 is an optionally substituted aryl. In some examples, R_3 is a monocyclic, bicyclic, or tricyclic aryl, each of which is optionally substituted. For example, R_3 is an optionally substituted phenyl, an optionally substituted naphthyl, or an optionally substituted anthracenyl. In other examples, R_3 is a monocyclic, bicyclic, or tricyclic aryl, each of which is optionally substituted with 1 to 4 of halo, hydroxy, cyano, nitro, aliphatic, haloaliphatic, (aliphatic)oxy, (halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, heterocycloaliphatic, or combinations thereof. In several examples, R_3 is an optionally substituted fused bicyclic aryl. In several examples, R_3 is an optionally substituted fused tricyclic aryl.

[0165] In several embodiments, R_3 is an optionally substituted heteroaryl. In several examples, R_3 is a monocyclic or bicyclic heteroaryl, each of which is optionally substituted with 1 to 4 of halo, hydroxy, cyano, nitro, aliphatic, haloaliphatic, (aliphatic)oxy, (halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, heterocycloaliphatic, or combinations thereof.

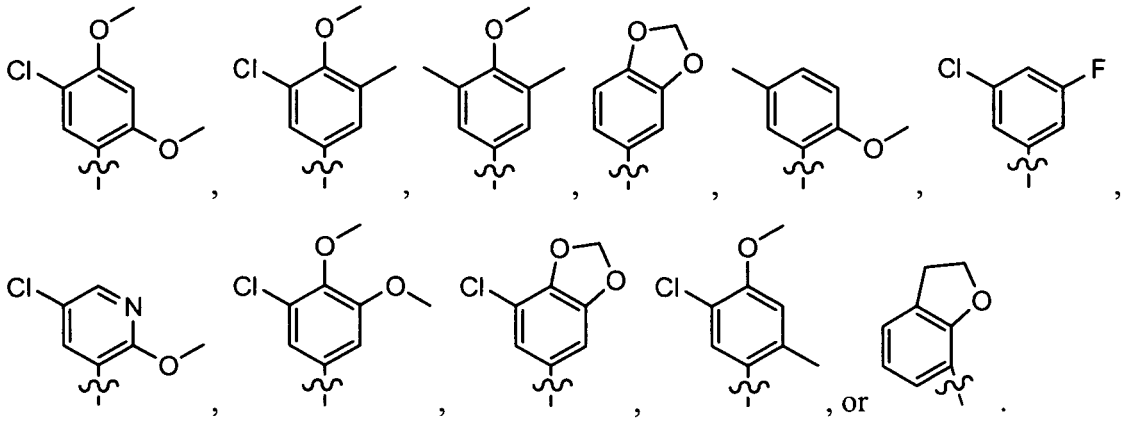
[0166] In some embodiments, R_3 is optionally substituted aliphatic such as methyl, ethyl or propyl, each of which is optionally substituted.

[0167] According to other embodiments, R_3 is an optionally substituted aliphatic.

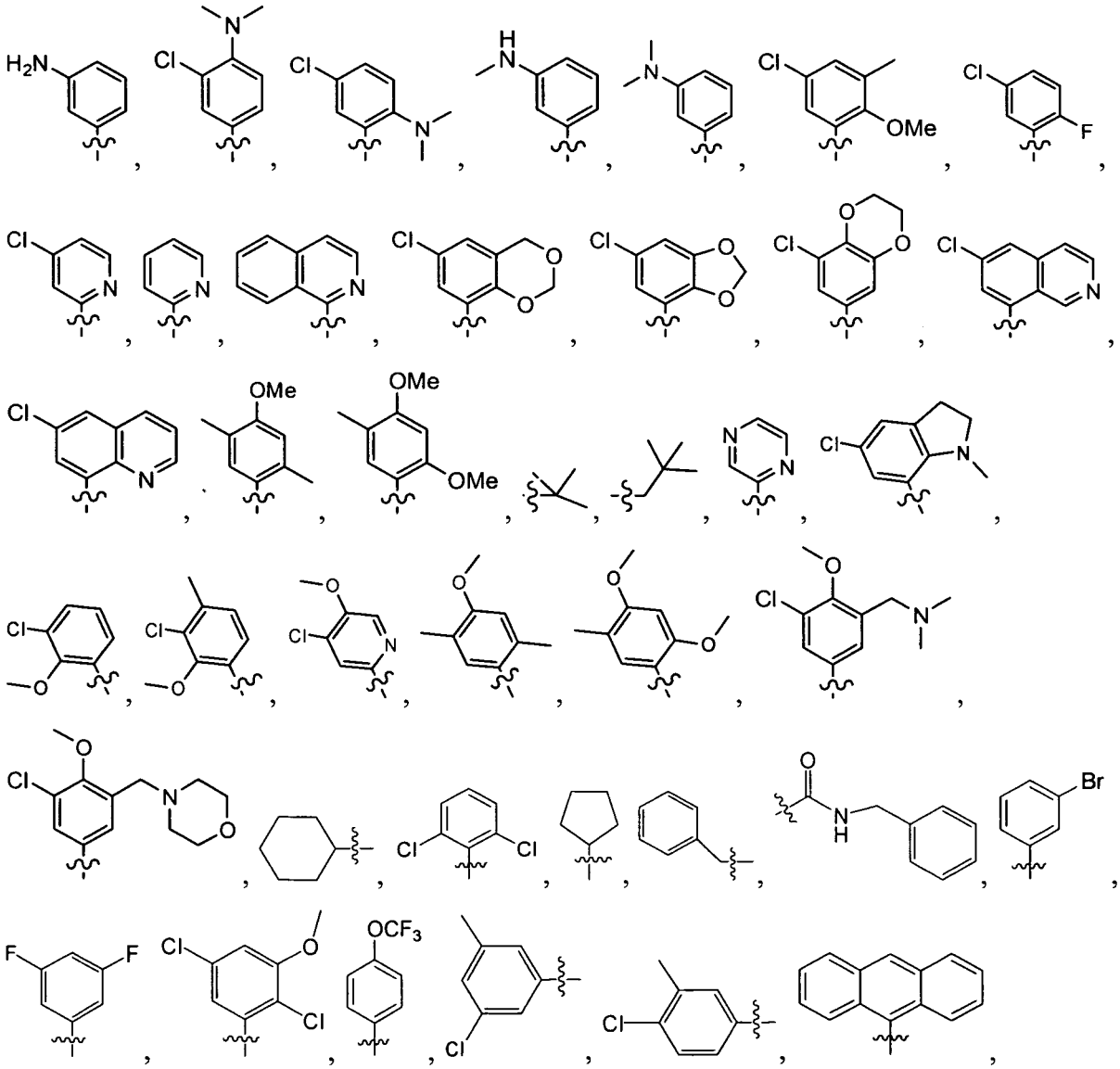
[0168] According to other embodiments, R_3 is an optionally substituted C_{1-5} aliphatic.

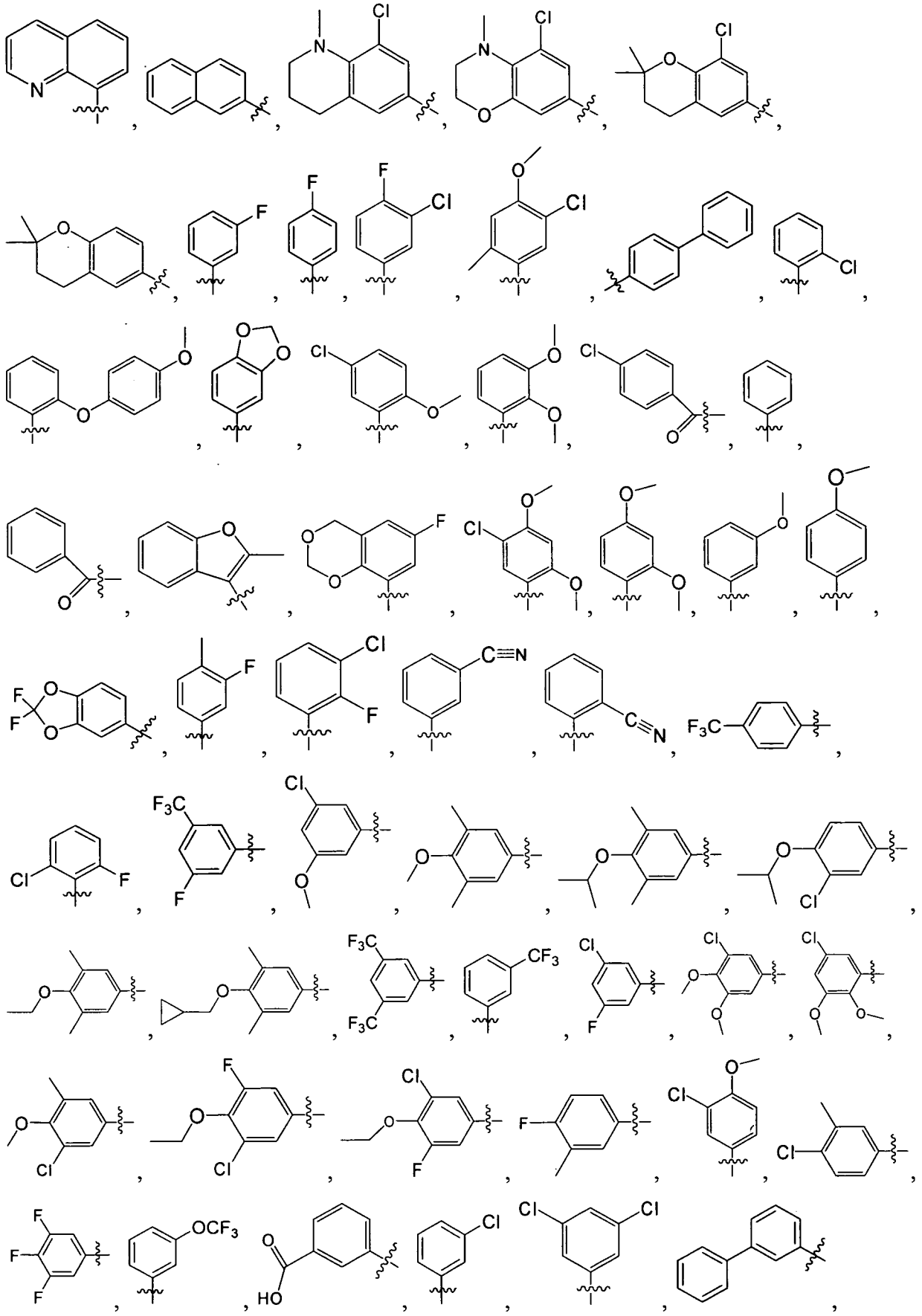


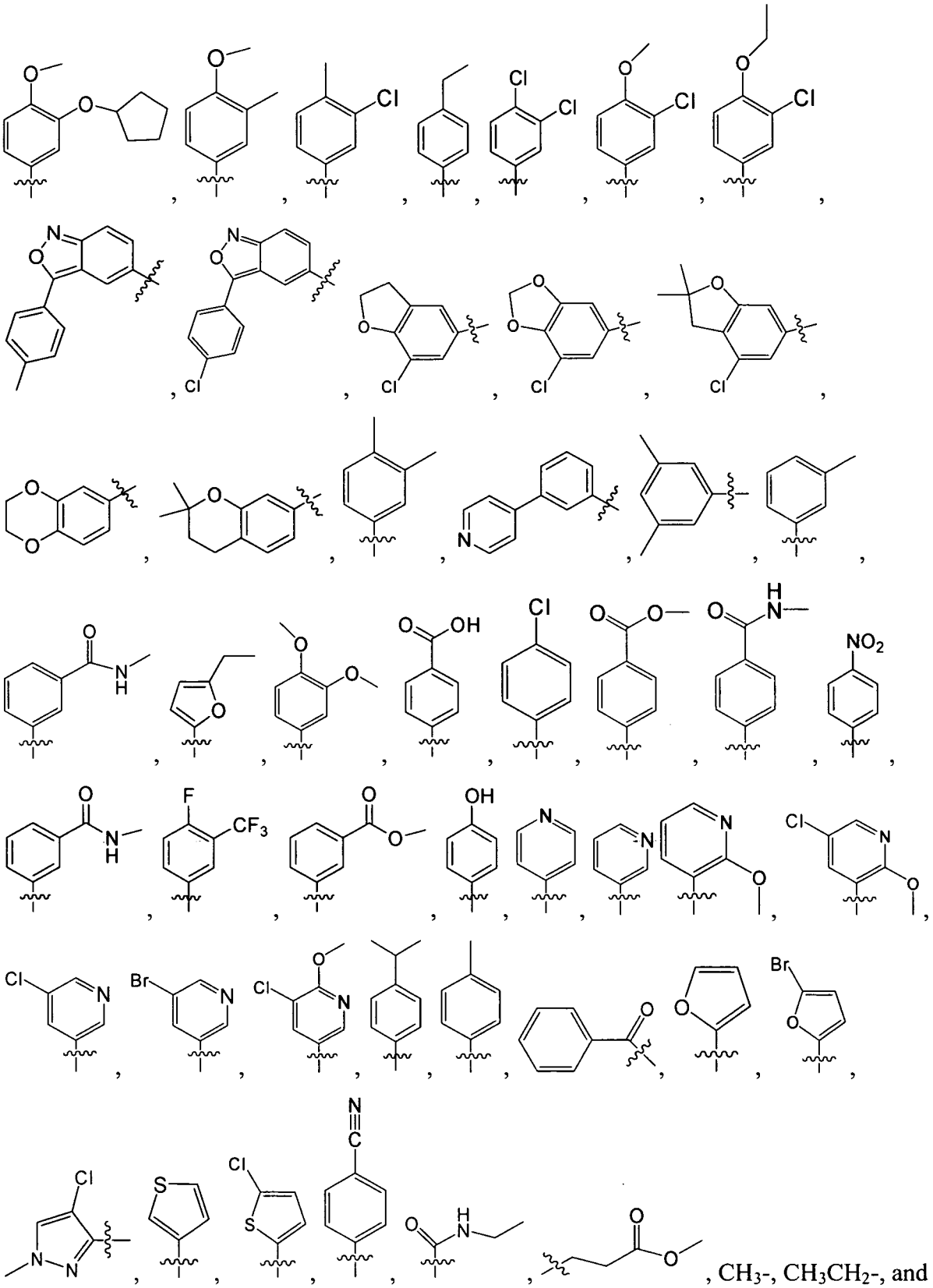
[0169] In several examples, R_3 is



[0170] In several embodiments, R_3 is one selected from:

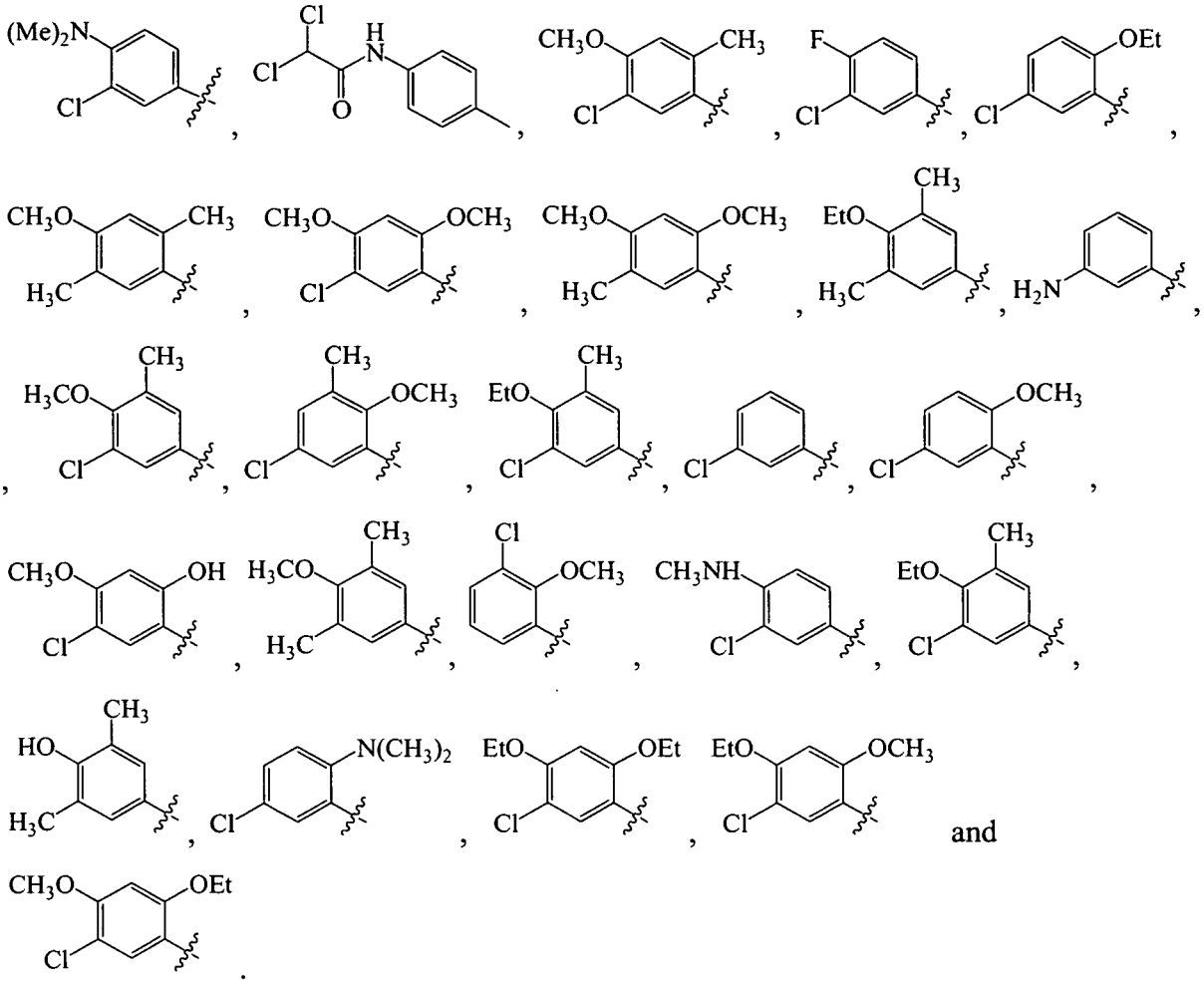






, CH_3-, CH_3CH_2-, and $CH_3CH_2CH_2$-.

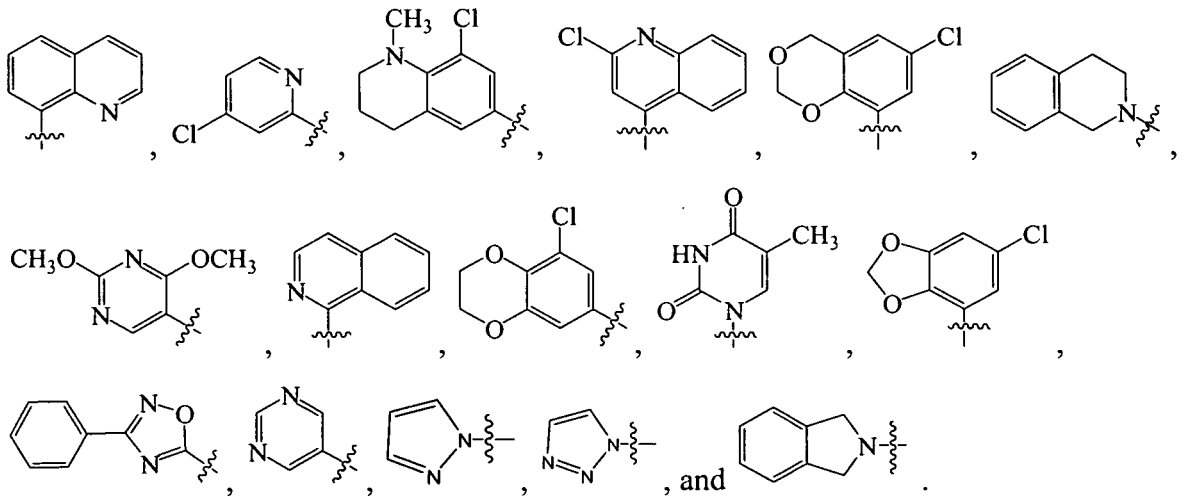
[0171] In some embodiments, R_3 is selected from the group consisting of:



and

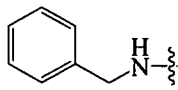
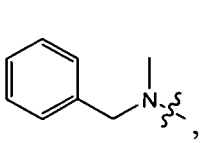
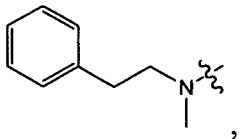
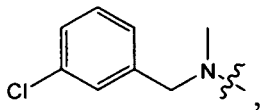
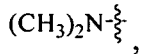
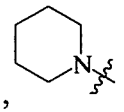
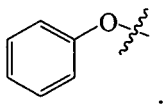
[0172] In some embodiments, R_3 is an optionally substituted monocyclic, bicyclic, or tricyclic heteroaryl, each of which is optionally substituted with 1-3 of halo, hydroxy, cyano, nitro, aliphatic, haloaliphatic, (aliphatic)oxy, (halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, heterocycloaliphatic, or combinations thereof.

[0173] In further embodiments, R_3 is selected from the group consisting of:



, and

[0174] In other embodiments, R_3 is an optionally substituted amino or an optionally substituted aryloxy.

[0175] In some embodiments, R_3 is selected from the group consisting of  ,  ,  ,  ,  ,  , and  .

4. **Group A:**

[0176] Each A is $-(CX_1X_2)_a-$, wherein each X_1 and X_2 is independently hydrogen, optionally substituted C_{1-4} aliphatic, or optionally substituted aryl; or X_1 and X_2 taken together form an oxo group; and each a is 0 to 3.

[0177] In several embodiments, X_1 or X_2 is hydrogen.

[0178] In several embodiments, X_1 or X_2 is optionally substituted C_{1-4} aliphatic. Examples of X_1 or X_2 include trifluoromethyl, or optionally substituted ethyl, propyl, butyl, or isomers thereof.

[0179] In several embodiments, X_1 or X_2 is an optionally substituted aryl. Examples of X_1 or X_2 include optionally substituted phenyl, naphthyl, or azulenyl.

5. **Group B:**

[0180] Each B is $-(CX_1X_2)_b-$, wherein each X_1 and X_2 is independently hydrogen, optionally substituted C_{1-4} aliphatic, or optionally substituted aryl; or X_1 and X_2 taken together form an oxo group; and each b is 0 to 3.

[0181] In several embodiments, X_1 or X_2 is hydrogen.

[0182] In several embodiments, X_1 or X_2 is optionally substituted C_{1-4} aliphatic. Examples of X_1 or X_2 include trifluoromethyl, or optionally substituted ethyl, propyl, butyl, or isomers thereof. In several additional examples, X_1 or X_2 is an optionally substituted mono- or di-substituted (amino)-C_{1-4} aliphatic.

[0183] In several embodiments, X_1 or X_2 is an optionally substituted aryl. Examples of X_1 or X_2 include optionally substituted phenyl, naphthyl, indenyl, or azulenyl.

6. **Substituents Y and Y'**

[0184] In several embodiments, each Y and Y' is independently hydrogen, optionally substituted aliphatic, or optionally substituted aryl.

[0185] Each Y and Y' is independently $-Z^DR_7$, wherein each Z^D is independently a bond or an optionally substituted straight or branched (C_{1-6})-aliphatic chain wherein up to two carbon units of Z^D are optionally and independently replaced by -C(O)-, -CS-, $-C(O)NR^D-$, $-C(O)NR^DNR^D-$, -C(O)O-, -OC(O)-, $-NR^DC(O)O-$, -O-, $-NR^DC(O)NR^D-$, $-OC(O)NR^D-$, $-NR^DNR^D-$, $-NR^DC(O)-$, -S-, -SO-, $-SO_2-$, $-NR^D-$, $-SO_2NR^D-$, $-NR^DSO_2-$, or $-NR^DSO_2NR^D-$.

Each R_7 is independently R^D, halo, -OH, -CN, $-NO_2$, $-NH_2$, or $-OCF_3$. Each R^D is independently hydrogen, or optionally substituted aryl.

[0186] In several embodiments, one selected from Y and Y' is hydrogen.

[0187] In several embodiments, one selected from Y and Y' is optionally substituted aliphatic.

[0188] In several embodiments, one selected from Y and Y' is optionally substituted aryl.

[0189] In several embodiments, both Y and Y' are hydrogen.

[0190] In several embodiments, one of Y or Y' is hydrogen and the other is fluorine.

[0191] In several embodiments, both of Y and Y' are fluorine.

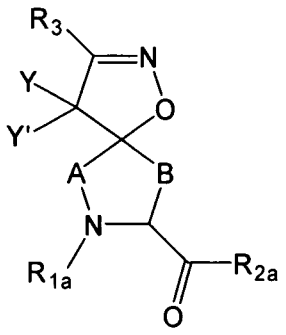
[0192] In additional of examples, one of Y or Y' is hydrogen and the other is methoxycarbonyl; one of Y or Y' is hydrogen and the other is hydroxy; or together, Y and Y' form an oxo group or form =S.

7. **Exceptions:**

[0193] In compounds of formula (I), the sum of **a** and **b** is 2 or 3. For example, **a** is 0 and **b** is 3; **a** is 1 and **b** is 2; **a** is 2 and **b** is 1; or **a** is 3 and **b** is 0.

C. Sub-generic Compounds:

[0194] Another aspect of the present invention provides compounds of formula (I)a useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (Ia) include:



(Ia)

or a pharmaceutically acceptable salt thereof wherein R_3, A, B, Y, and Y' are defined above in formula (I).

[0195] Each R_{1a} is $-Q_4-W_4-Q_3-W_3-Q_2-W_2-Q_1$; wherein each of W_2, W_3, and W_4 is independently a bond, -C(O)-, -C(S)-, $-C(O)N(Q_5)-$, -C(O)O-, -O-, $-N(Q_5)C(O)N(Q_5)-$, $-SO_2-$, $-N(Q_5)SO_2-$, -S-, $-N(Q_5)-$, -SO-, $-N(Q_5)C(O)-$, -OC(O)-, $-N(Q_5)C(O)O-$, or $-SO_2N(Q_5)-$; each of Q_1, Q_2, Q_3 and Q_4 is independently a bond, an optionally substituted C_{1-4} aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, an optionally substituted heteroaryl, or a hydrogen when Q_1, Q_2,

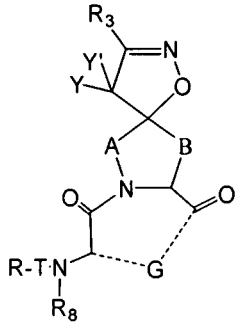
Q_3, or Q_4 is the terminal group of R_1; and each Q_5 is independently hydrogen or an optionally substituted aliphatic.

[0196] Each R_{2a} is $-Z_1-V_1-Z_2-V_2-Z_3-V_3$ each of V_1, V_2, and V_3 is independently a bond, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, an optionally substituted heteroaryl, or a hydrogen when V_1, V_2, V_3 is the terminal group of R_2; each of Z_1, Z_2, and Z_3 is independently a bond, -C(O)-, -C(O)C(O)-, -C(S)-, $-C(O)N(Q_5)-$, $-N(Q_5)C(O)-$, $-C(O)C(O)N(Q_5)-$, -O-, , SO-, $-SO_2-$, $-N(Q_5)SO_2-$, $-N(Q_5)C(O)N(Q_5)-$, $-N(Q_5)C(S)N(Q_5)-$, $-N(Q_5)-$, $-N(Q_5)SO_2-$, $-SO_2N(Q_5)-$, $-C(O)N(Q_5)SO_2-$, $-SO_2N(Q_5)C(O)-$, or hydrogen when Z_1, Z_2, or Z_3 is the terminal group of R_2; and each Q_5 is independently hydrogen, or an optionally substituted aliphatic.

[0197] In several examples, R_{2a} is an optionally substituted (aliphatic)amino, an optionally substituted alkoxy, or hydroxy.

[0198] In several examples, R_{2a} is an (aliphatic)amino wherein the nitrogen atom is optionally substituted with $-Z_2-V_2-Z_3-V_3$ or $-Z_3-V_3$ wherein each of Z_2 and Z_3 is independently a bond, -C(O)-, $-N(Q_5)-$, or $-C(O)C(O)N(Q_5)-$; and each of V_2 and V_3 is independently a bond, an optionally substituted aliphatic, or an optionally substituted cycloaliphatic.

[0199] Another aspect of the present invention provides compounds of formula (I)b useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (I)b include:

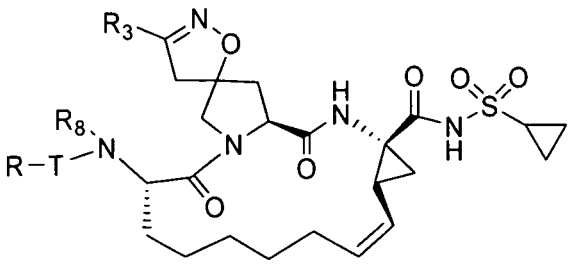
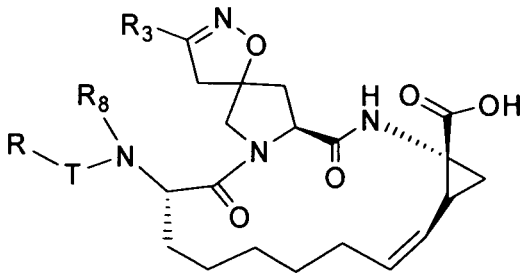


Ib

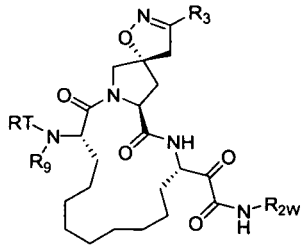
or a pharmaceutically acceptable salt thereof, wherein R_3, R_8, R, T, A, B, Y and Y' are defined above in formula (I).

[0200] Each G is a 2 to 15 atom optionally substituted aliphatic chain optionally containing 1 to 3 heteroatoms selected from O, S and N.

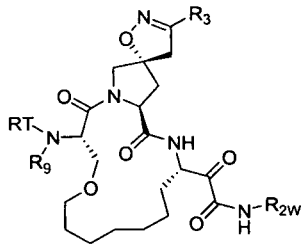
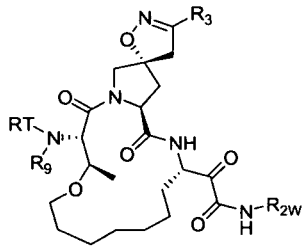
[0201] Examples of compounds of formula (I)b include:


or

,

wherein T, R, and R_3 are defined above in formula (I).

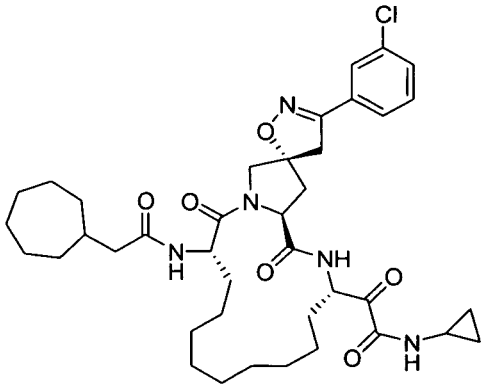
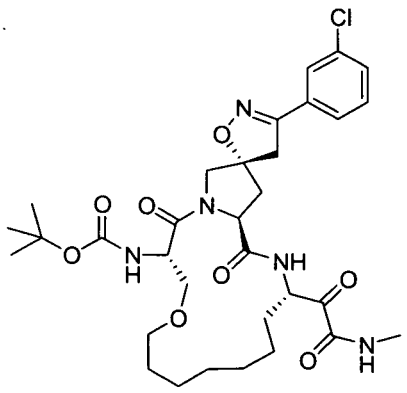

,

[0202] Still other examples of formula (I)b are

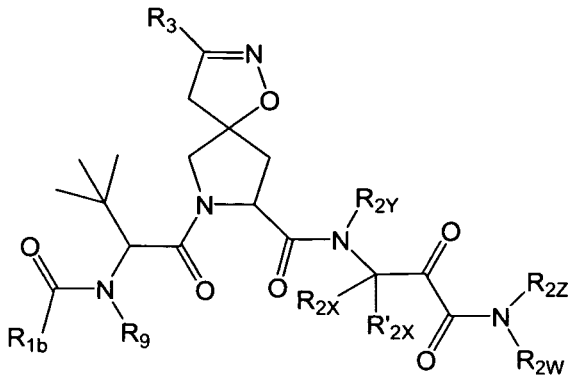

, or

, wherein each R_{2W} is independently

, , , or hydrogen; each T is independently a bond, -C(O)-, -OC(O)-, -NHC(O)-, $-S(O)_2N(H)-$, -C(O)C(O)- or $-SO_2-$; each R is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl; and each R_9 is independently hydrogen, an optionally substituted aliphatic, an optionally substituted heteroaryl, an optionally substituted phenyl.

[0203] Further specific examples of compounds of formula (I)b are


or

.

[0204] Other examples of compounds of formula (I)b include:



(II)

or a pharmaceutically acceptable salt thereof, wherein

each R_3 is an optionally substituted aryl or an optionally substituted heteroaryl;

each R_{2Y} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_9 is independently hydrogen, optionally substituted aliphatic, optionally substituted aryl, optionally substituted heteroaryl, optionally substituted heterocycloaliphatic, or optionally substituted cycloaliphatic;

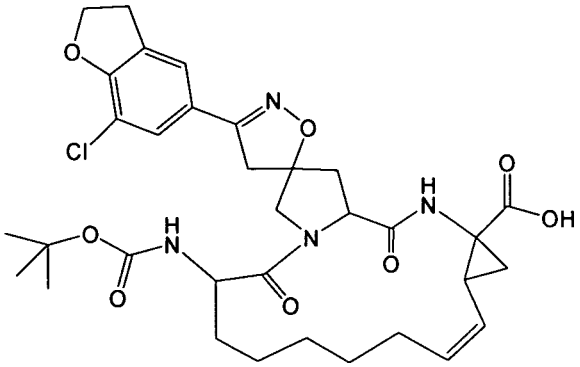
each of R_{2X} and R'_{2X} is independently hydrogen, an optionally substituted aliphatic, an optionally substituted heteroaryl, an optionally substituted phenyl, an optionally substituted cycloaliphatic, an optionally substituted heterocycloaliphatic; or R_{2X} and R'_{2X} together with the atom to which they are both attached form an optionally substituted 3 to 7 membered cycloaliphatic or heterocycloaliphatic ring, or R_{2X} and R_{2Y} together with the atoms to which they are attached form an optionally substituted 5 to 7 membered heterocycloaliphatic ring;

each R_{1b} is $-Z^ER_{21}$, wherein Z^E is $-CH_2-$, -NH-, $-CH(R_{1Z})-$, or -O-, and R_{21} is optionally substituted 6-7 membered cycloaliphatic or optionally substituted *tert*-butyl;

each R_{1Z} is optionally substituted aliphatic, optionally substituted cycloaliphatic, optionally substituted heterocycloaliphatic, optionally substituted aryl , or optionally substituted heteroaryl;

each R_{2Z} is hydrogen, optionally substituted cycloaliphatic, optionally substituted heterocycloaliphatic, or optionally substituted aliphatic; and

each R_{2W} is hydrogen, optionally substituted cycloaliphatic, optionally substituted heterocycloaliphatic, or optionally substituted aliphatic, or R_{2Z} and R_{2W}, together with the nitrogen atom to which they are attached form an optionally substituted heterocycloaliphatic.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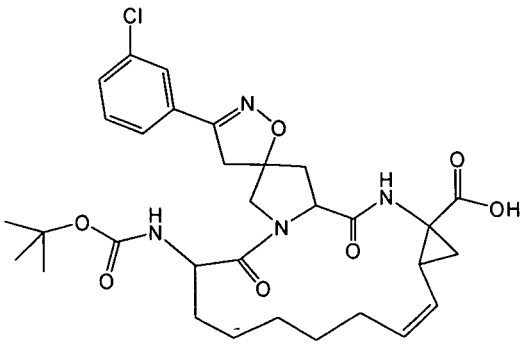
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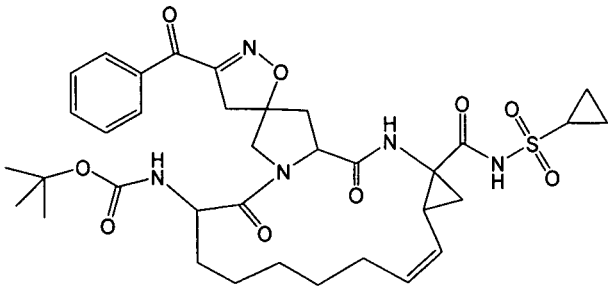
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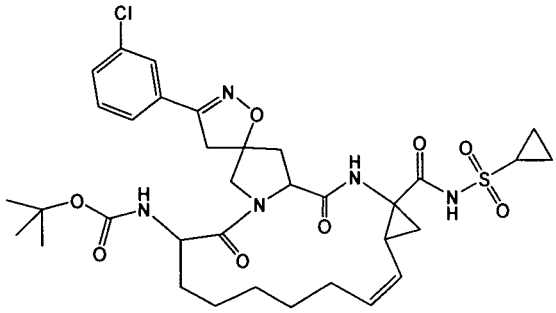
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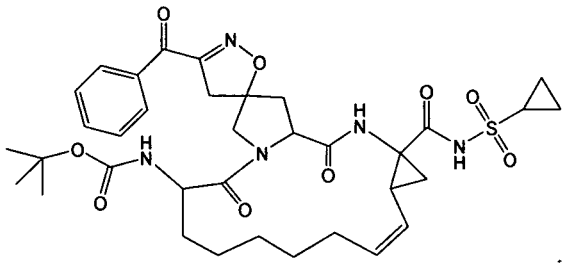
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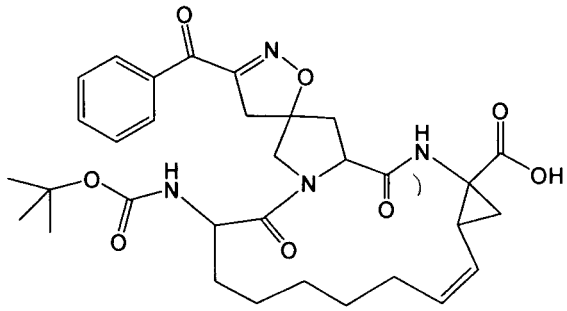
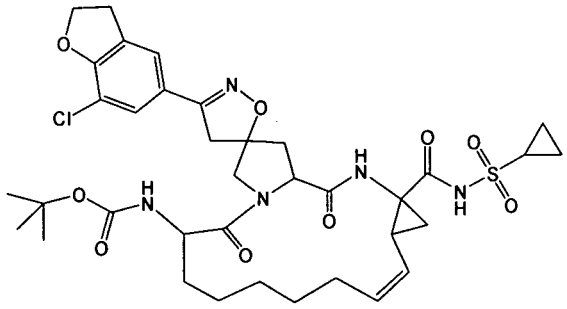
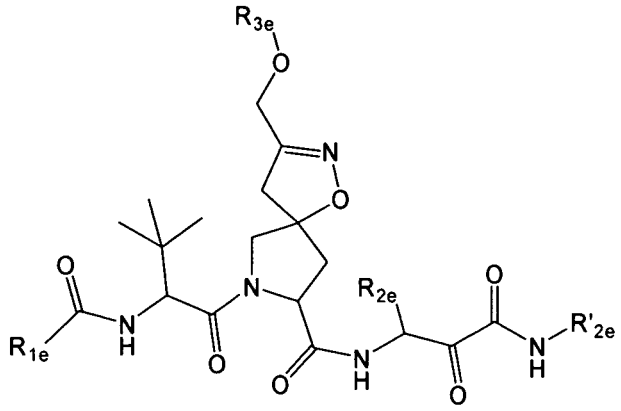

, and

.

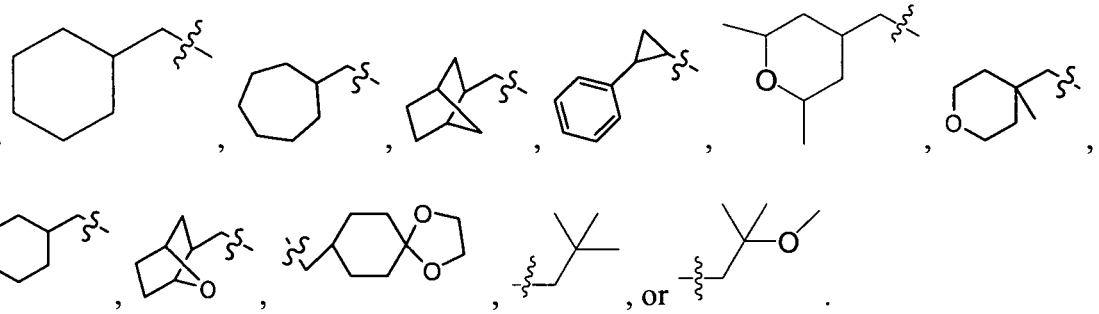
[0205] Another aspect of the present invention provides compounds of formula (I)I useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (II) include:

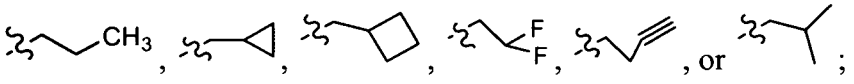
[0206] Another aspect of the present invention provides compounds of formula (I)II useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (III) include:

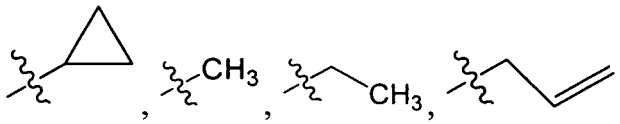


(III)

or a pharmaceutically acceptable salt thereof, wherein

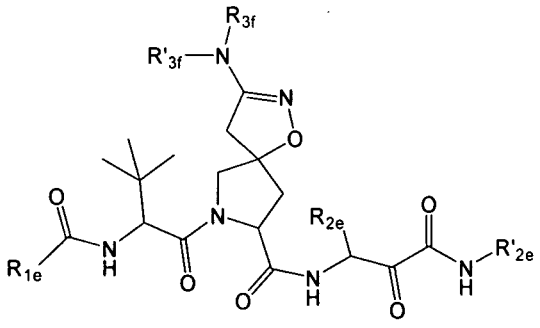
R_{1e} is 

R_{2e} is  ;

R'_{2e} is  , or hydrogen; and

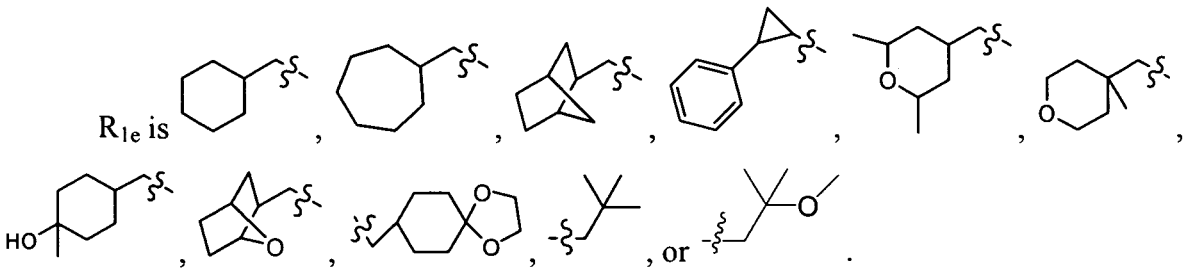
R_{3e} is optionally substituted aryl or optionally substituted heteroaryl.

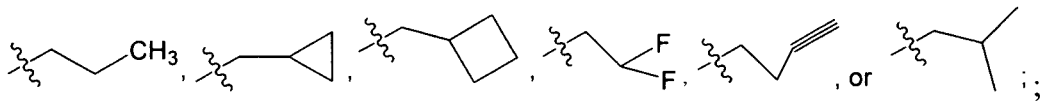
[0207] Another aspect of the present invention provides compounds of formula (IV) useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (IV) include:

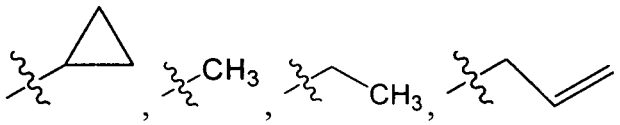


(IV)

or pharmaceutically acceptable salts thereof, wherein

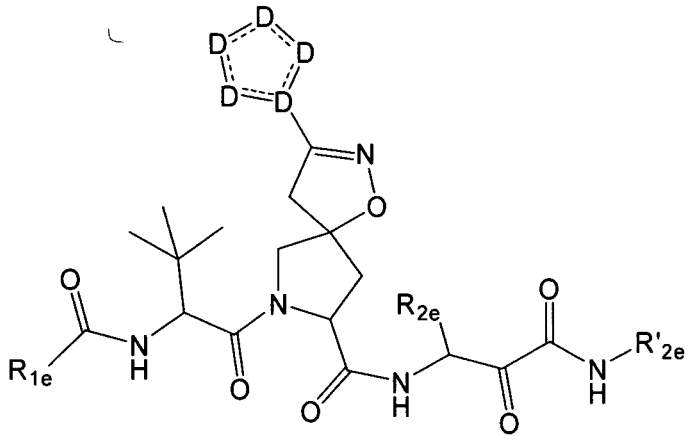
R_{1e} is 

R_{2e} is 

R'_{2e} is  , or hydrogen; and

each of R_{3f} and R'_{3f} is independently hydrogen, sulfonamide, sulfonyl, sulfinyl, optionally substituted acyl, optionally substituted aliphatic, optionally substituted cycloaliphatic, optionally substituted heterocycloaliphatic, optionally substituted aryl, or optionally substituted heteroaryl, or R_{3f} and R'_{3f} together with the nitrogen atom to which they are attached form an optionally substituted, saturated, partially unsaturated, or full unsaturated, 5- to 8-membered heterocycloaliphatic or heteroaryl.

[0208] Another aspect of the present invention provides compounds of formula V useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (V) include:

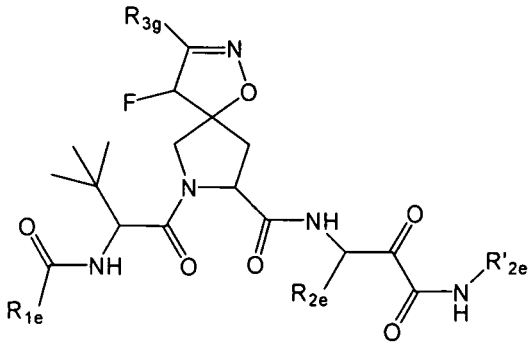


(V)

or pharmaceutically acceptable salts thereof, wherein R_{1e}, R_{2e}, and R'_{2e} are defined above in formula (III).

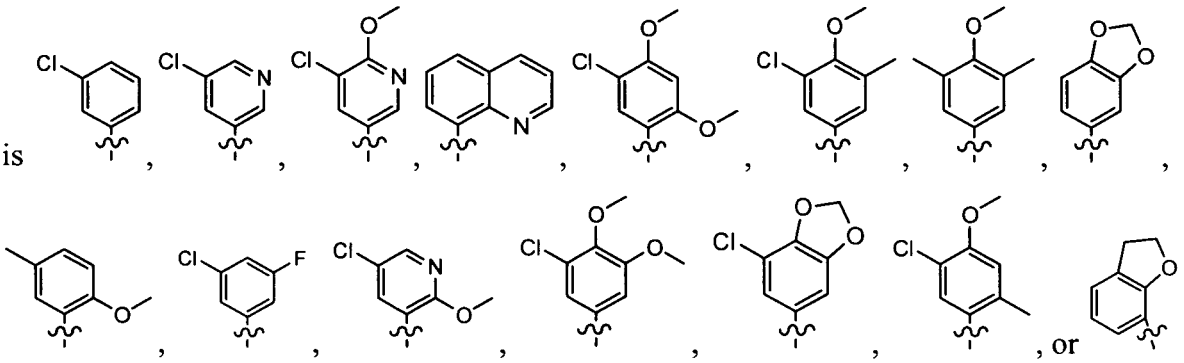
[0209] Each D is independently $-CR_8-$, N, S, or O, provided that no more than two D are independently, S, or O, and R_8 is defined above in formula (I).

[0210] Another aspect of the present invention provides compounds of formula VI useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (VI) include:

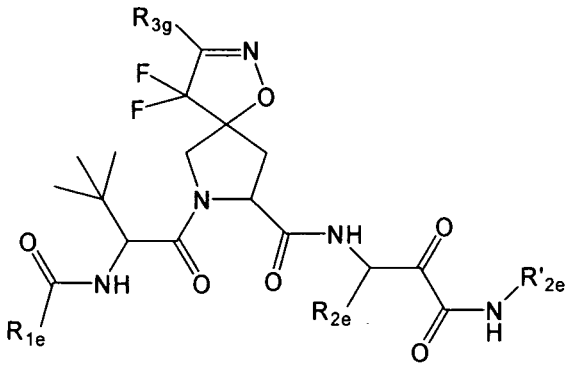


(VI)

or pharmaceutically acceptable salts thereof, wherein R_{1e}, R_{2e}, and R'_{2e} are defined above in formula (III).

[0211] Each R_{3g} is a substituted aryl or a substituted heteroaryl. In some embodiments, R_{3g} is  , or .

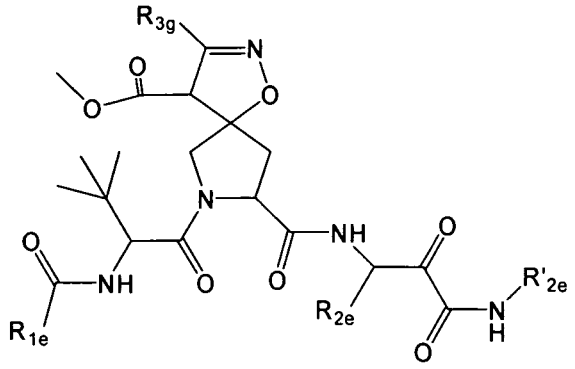
[0212] Another aspect of the present invention provides compounds of formula VII useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (VII) include:



(VII)

or pharmaceutically acceptable salts thereof, wherein R_{1e}, R_{2e}, and R'_{2e} are defined above in formula (III), and R_{3g} is defined in formula (VI).

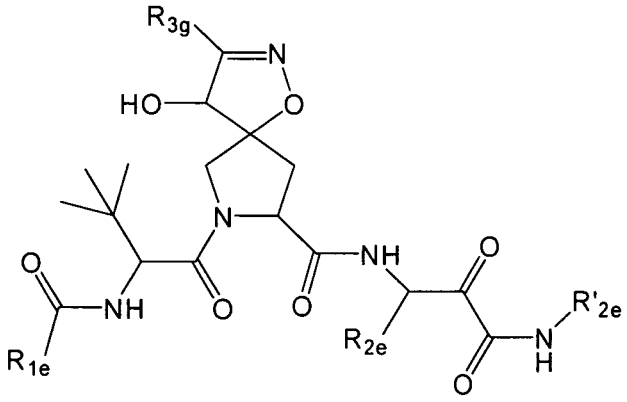
[0213] Another aspect of the present invention provides compounds of formula VIII useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (VIII) include:



(VIII)

or pharmaceutically acceptable salts thereof, wherein R_{1e}, R_{2e}, and R'_{2e} are defined above in formula (I)II, and R_{3g} is defined in formula VI.

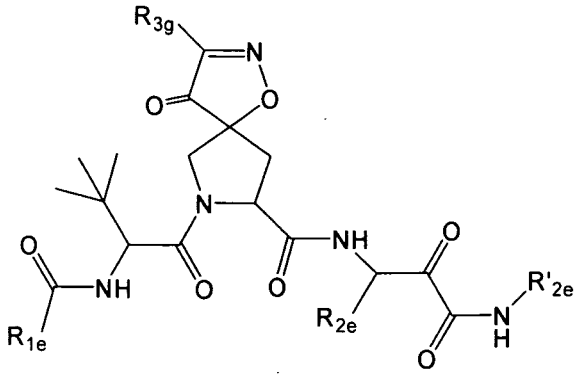
[0214] Another aspect of the present invention provides compounds of formula (I)X useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (IX) include:



(IX)

or pharmaceutically acceptable salts thereof, wherein R_{1e}, R_{2e}, and R'_{2e} are defined above in formula (III), and R_{3g} is defined in formula (VI).

[0215] Another aspect of the present invention provides compounds of formula (X) useful for inhibiting serine protease activity and methods inhibiting serine protease activity. Compounds of formula (X) include:



(X)

or a pharmaceutically acceptable salt thereof, wherein R_{1e}, R_{2e}, and R'_{2e} are defined above in formula (III), and R_{3g} is defined in formula (VI).

D. Combinations of Embodiments

[0216] Other embodiments include any combination of the aforementioned substituents R_1, R_2, R_3, A, B, Y, and Y'.

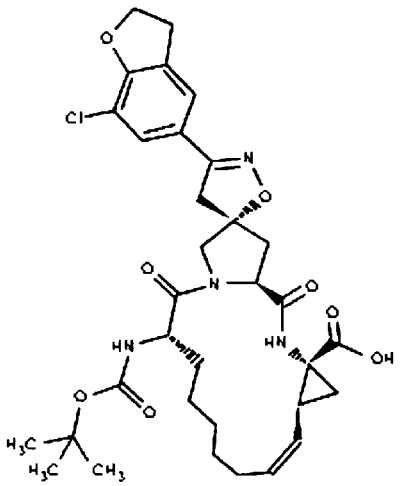
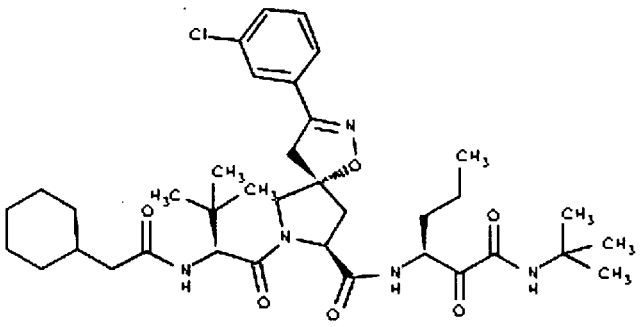
E. Exemplary Compounds

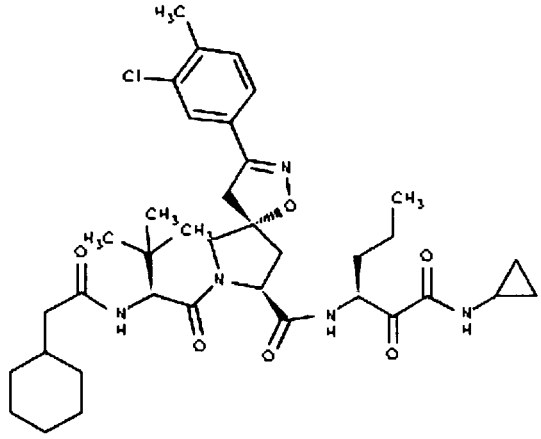
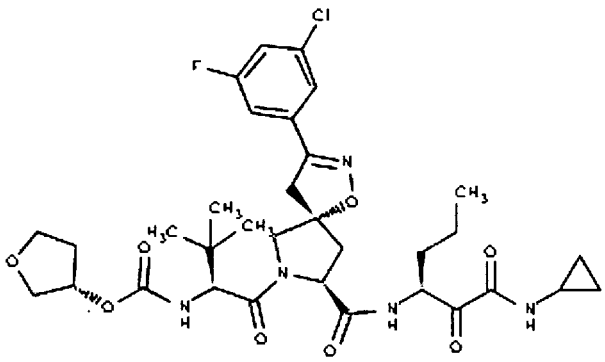
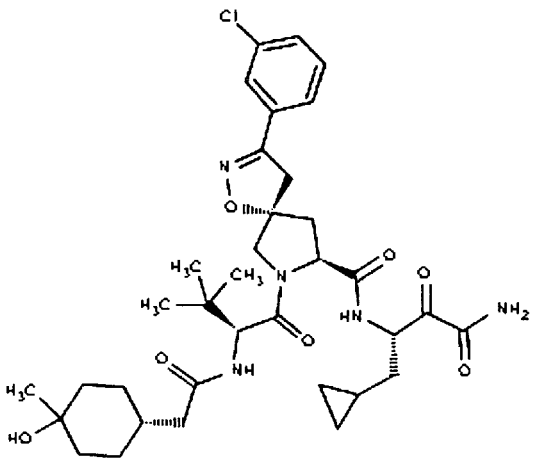
[0217] The invention is intended to include compounds wherein R_1 and R_2 contain structural elements of a serine protease inhibitor. Compounds having the structural elements of a serine protease inhibitor include, but are not limited to, the compounds of the following publications: WO 97/43310, US 20020016294, WO 01/81325, WO 01/58929, WO 01/32691, WO 02/08198, WO 01/77113, WO 02/08187, WO 02/08256, WO 02/08244, WO 03/006490, WO 01/74768, WO 99/50230, WO 98/17679, WO 02/48157, WO 02/08251, WO 02/07761, WO 02/48172, WO 02/08256, US 20020177725, WO 02/060926, US 20030008828, WO 02/48116, WO 01/64678, WO 01/07407, WO 98/46630, WO 00/59929, WO 99/07733, WO 00/09588, US 20020016442, WO 00/09543, WO 99/07734, US 6,018,020, US 6,265,380, US 6,608,027, US 20020032175, US 20050080017, WO 98/22496, WO 05/028502, US 5,866,684, WO 02/079234, WO 00/31129, WO 99/38888, WO 99/64442, WO 2004/072243, WO 02/18369, US 2006/046956, US 2005/197301, WO 2005058821, WO 2005051980, WO 2005030796, WO2005021584, WO2005113581, WO2005087731, WO2005087725, WO2005087721, WO2005085275, WO2005085242, US2003216325, WO2003062265, WO2003062228, WO2002008256, WO 2002008198, WO2002008187, WO 2002048172, WO 2001081325, WO 2001077113, US 6251583, US 5990276, US20040224900, US20040229818, WO2004037855, WO2004039833, WO200489974, WO2004103996, WO2004030670, WO2005028501, WO2006007700, WO2005070955, WO2006007708, WO2006000085, WO2005073195, WO2005073216, WO2004026896, WO2004072243, WO2004113365, WO2005010029, US20050153877,

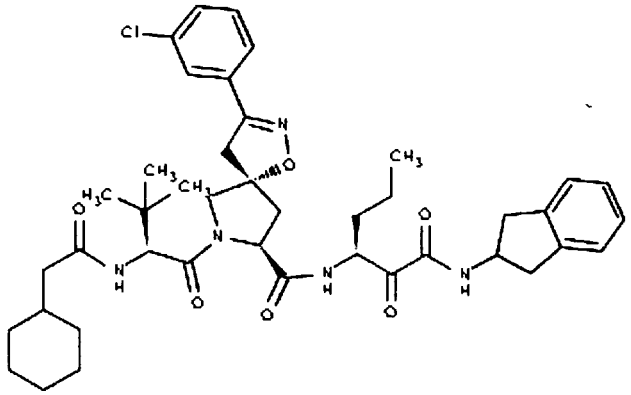
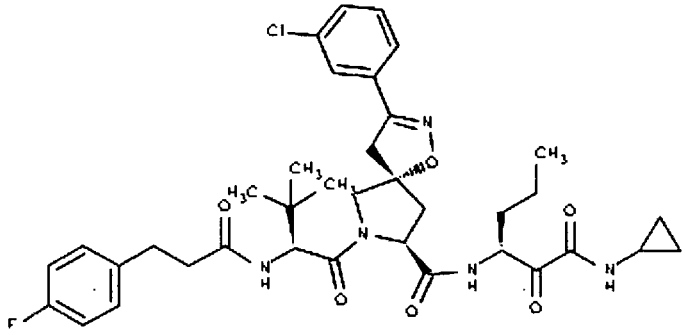
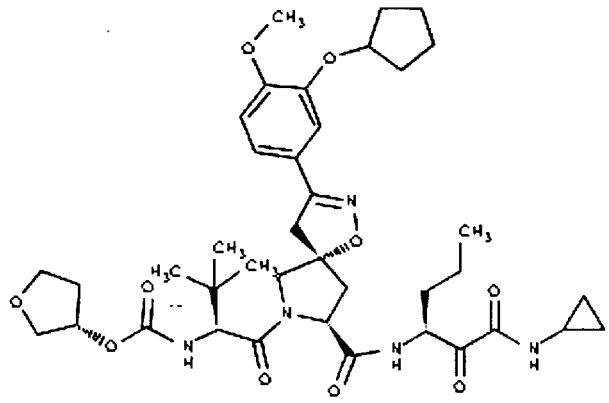
WO2004093798, WO2004094452, WO2005046712, WO2005051410, WO2005054430, WO2004032827, WO2005095403, WO2005077969, WO2005037860, WO2004092161, WO2005028502, WO2003087092, and WO2005037214, each of which is incorporated herein by reference.

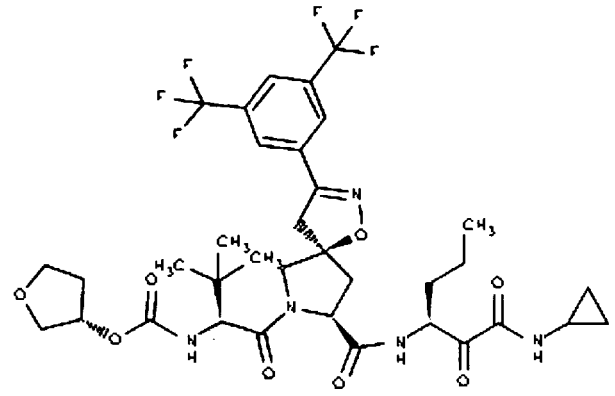
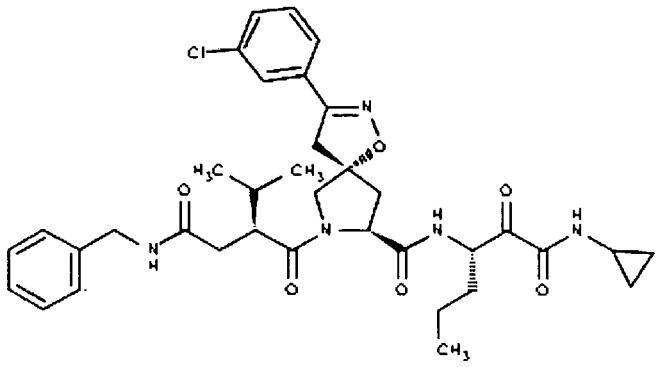
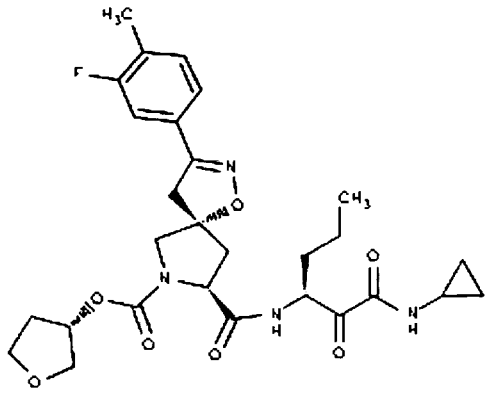
[0218] Specific exemplary compounds of the invention are shown below in Table 1.

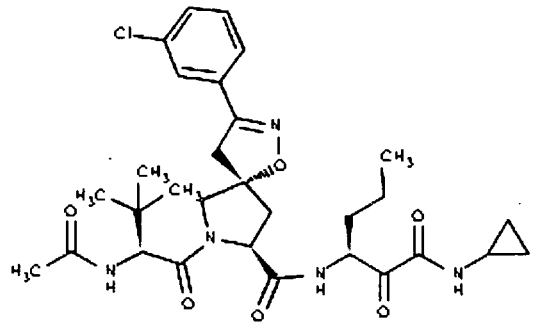
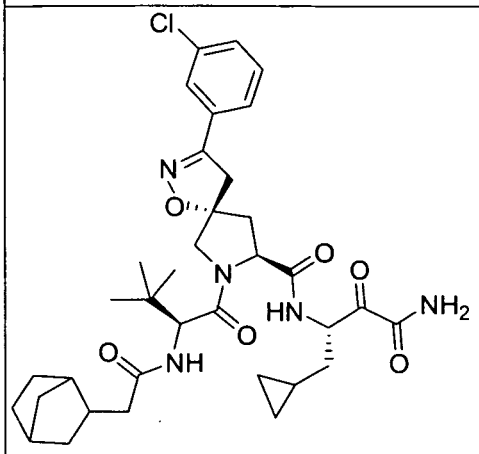
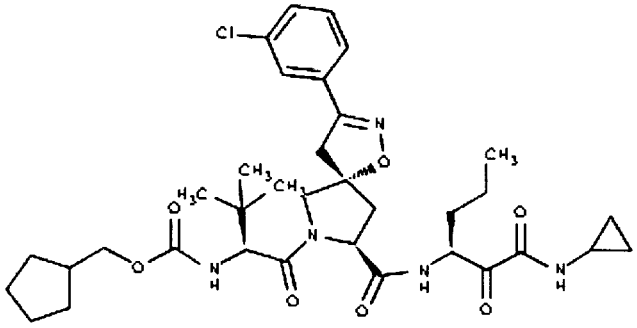
Table 1: Exemplary compounds of Formula (I).

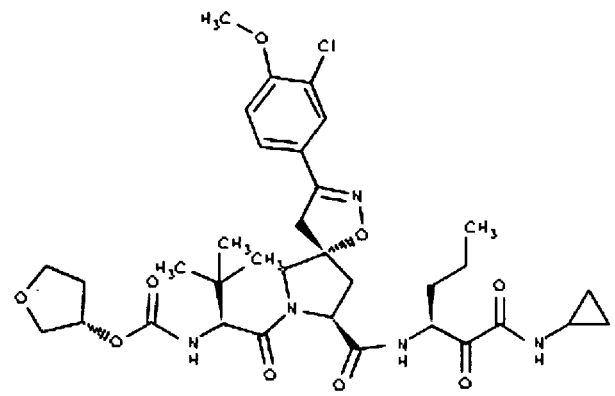
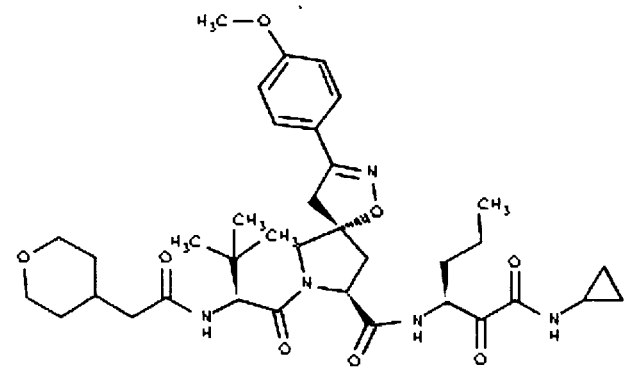
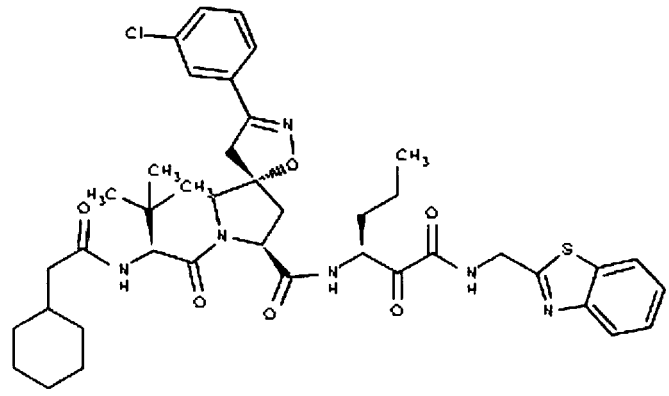
Structure	Compound No.
	1
	2

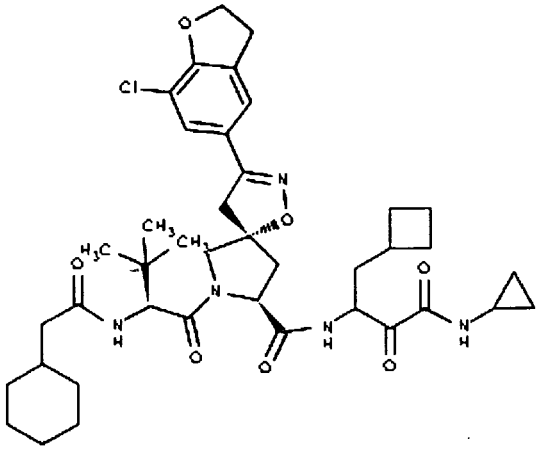
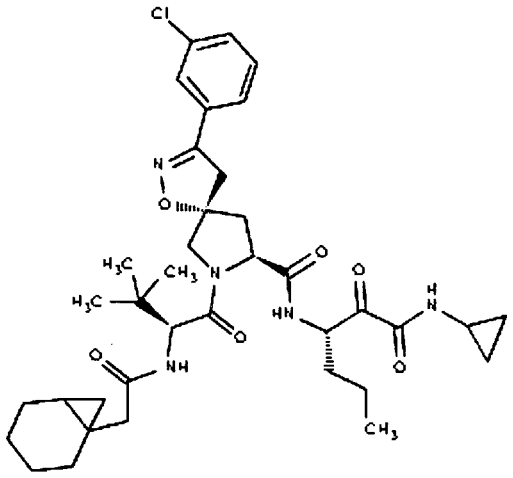
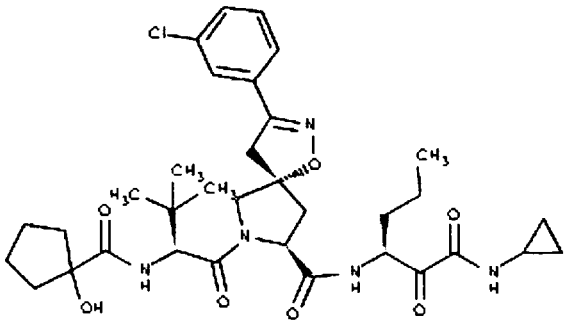
	3
	4
	5

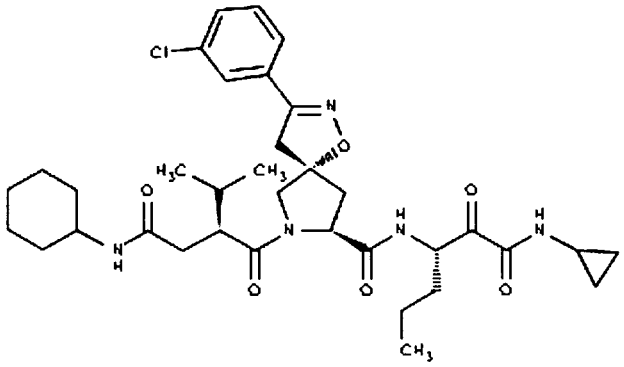
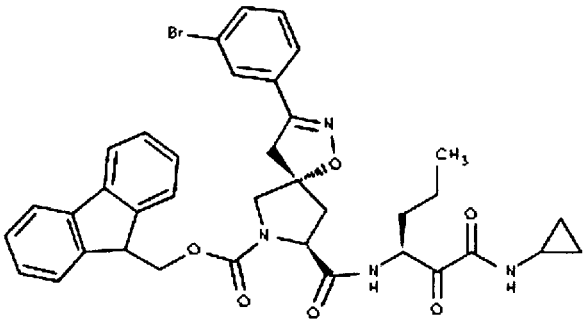
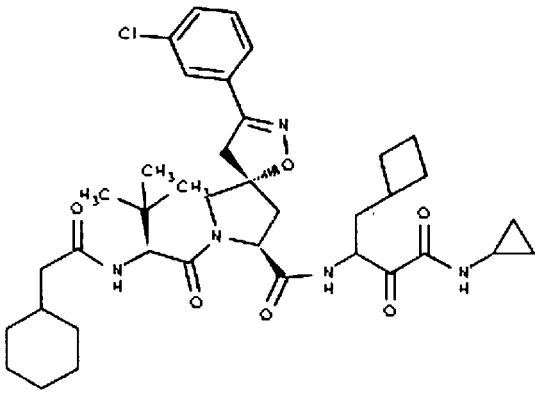
	6
	7
	8

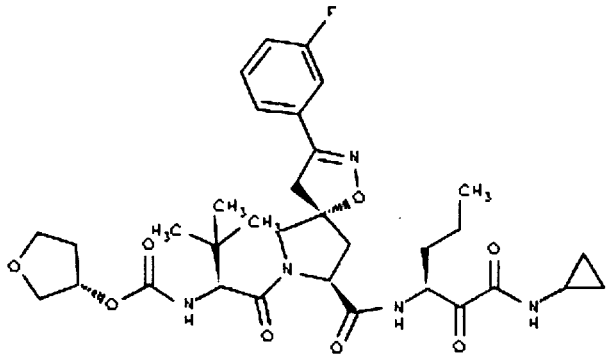
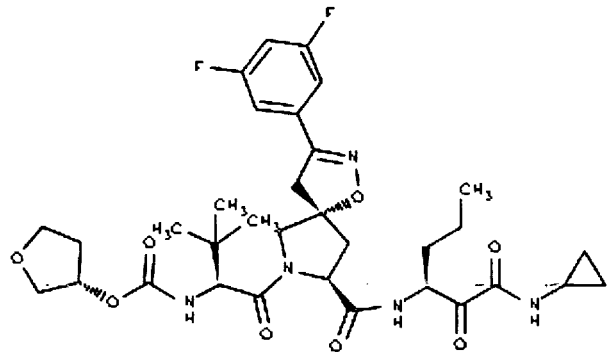
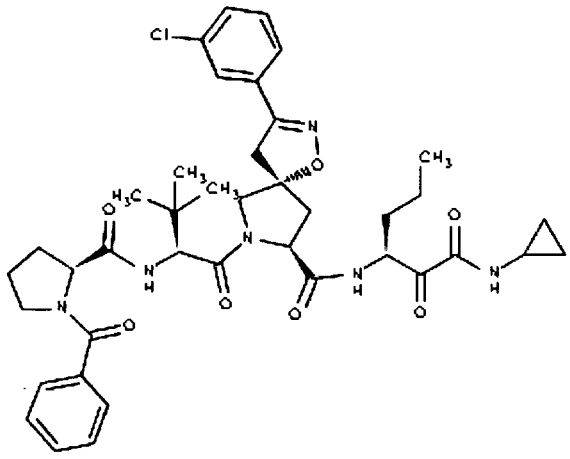
	9
	10
	11

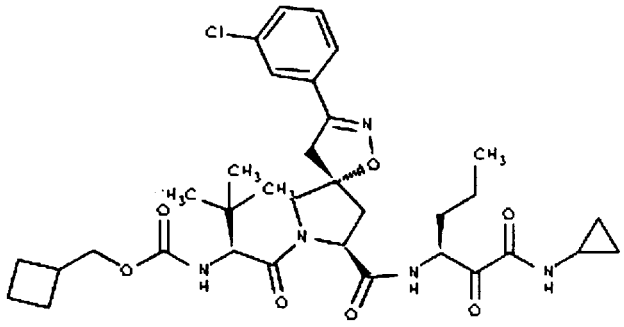
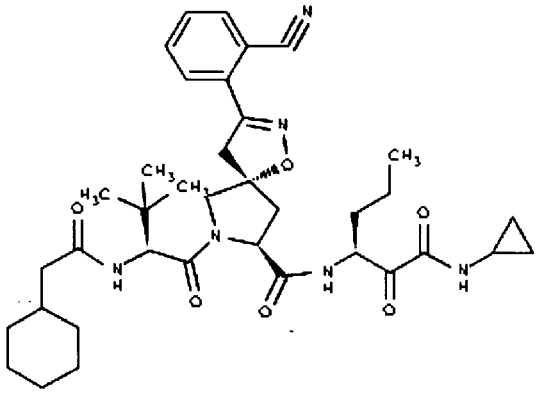
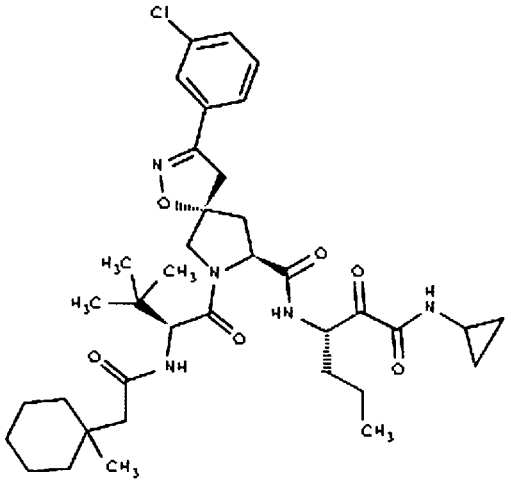
	12
	13
	14

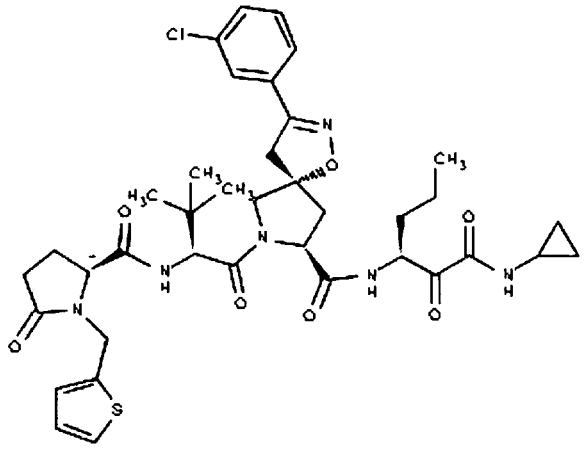
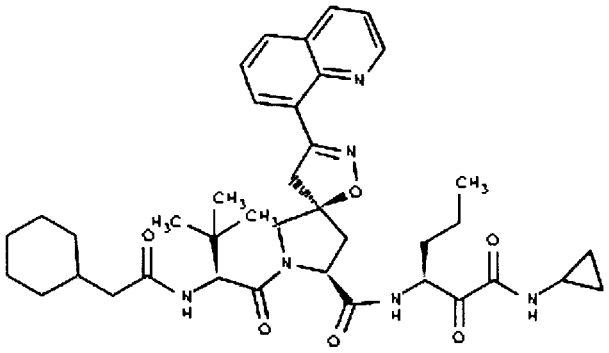
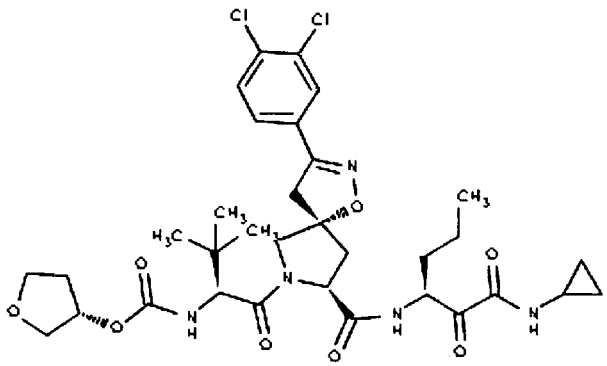
	15
	16
	17

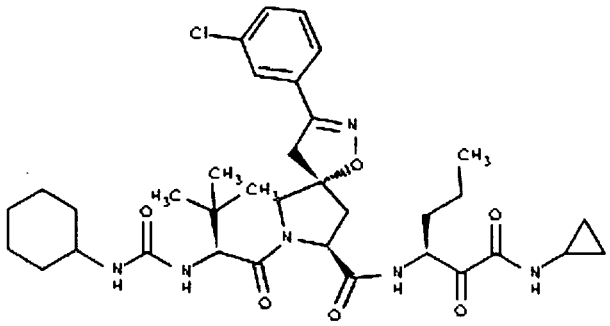
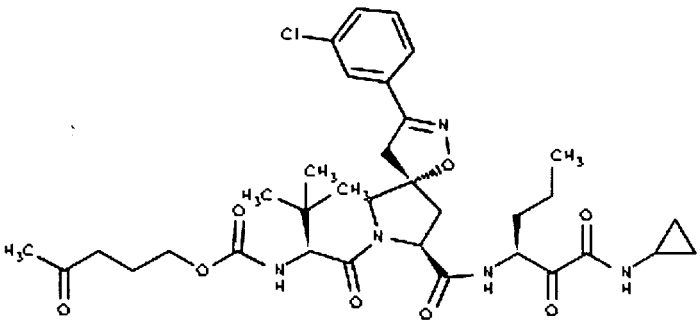
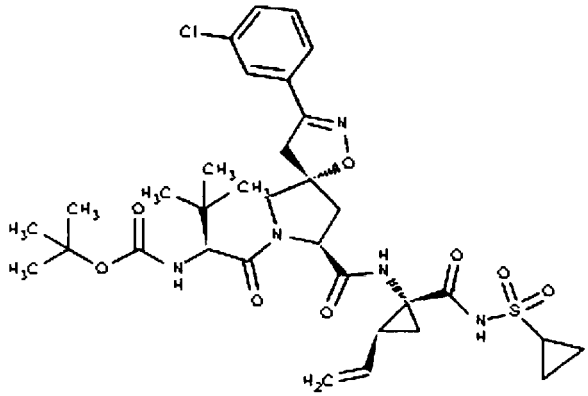
	18
	19
	20

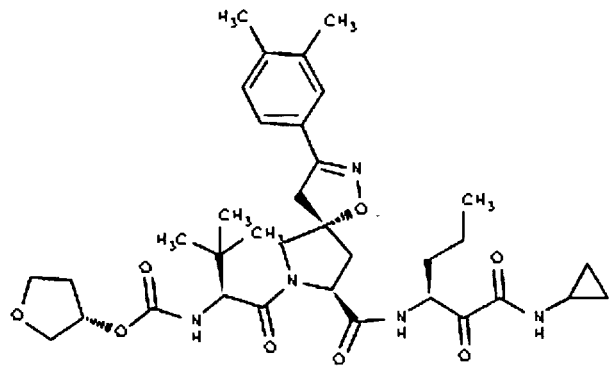
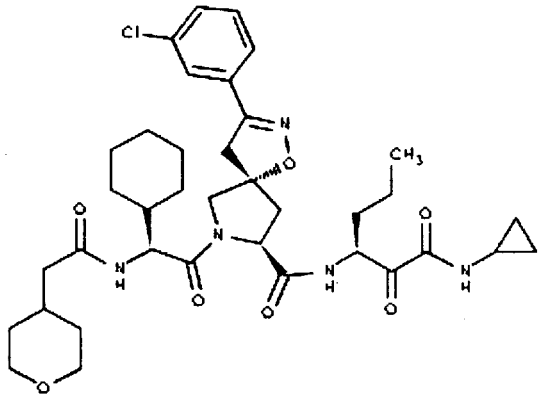
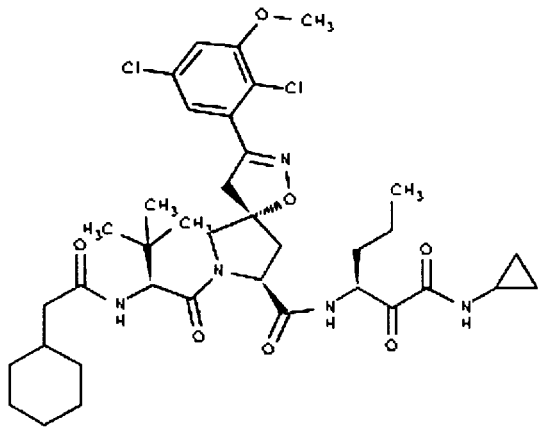
	21
	22
	23

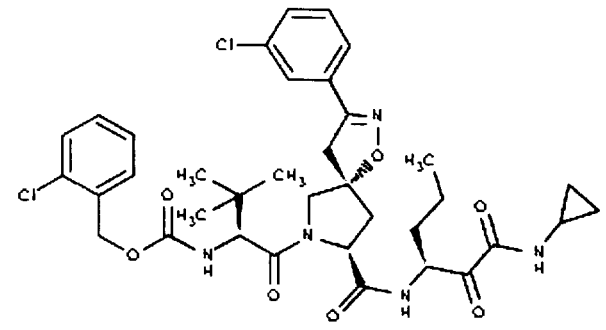
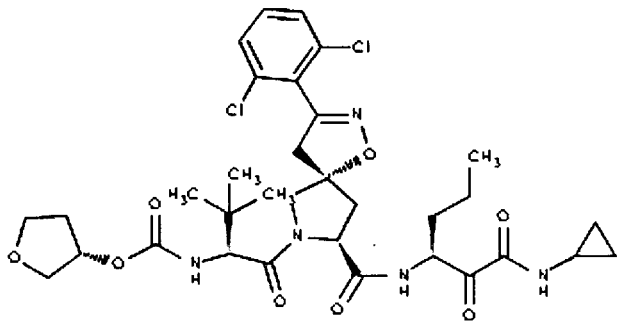
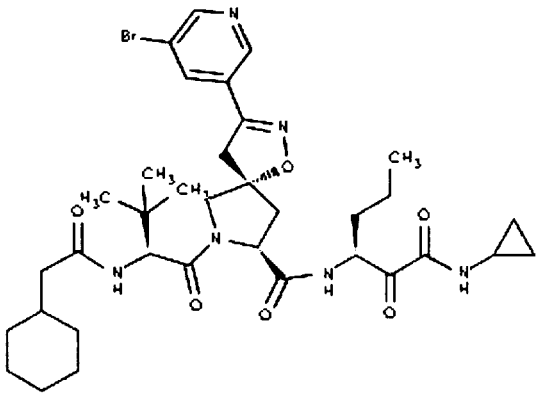
	24
	25
	26

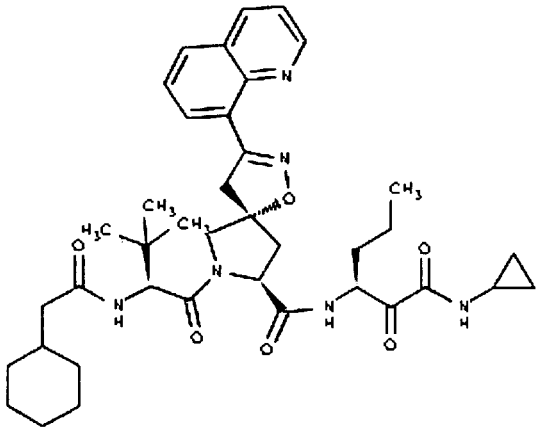
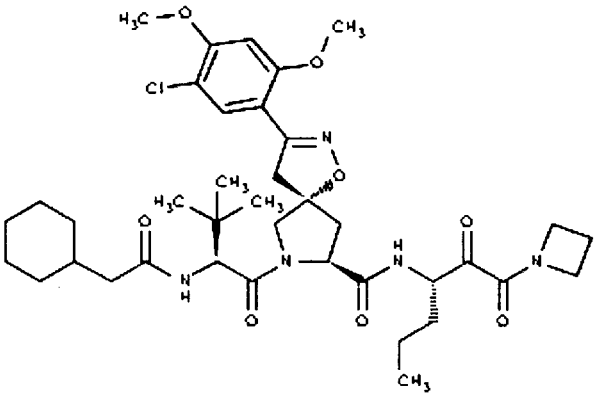
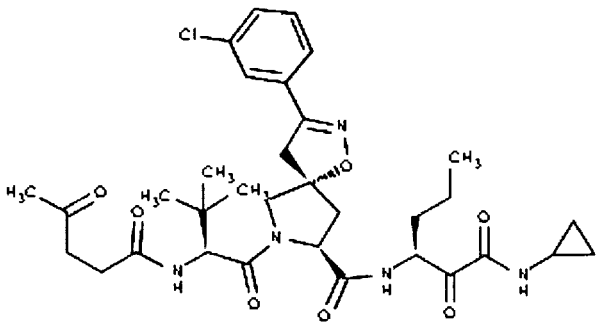
	27
	28
	29

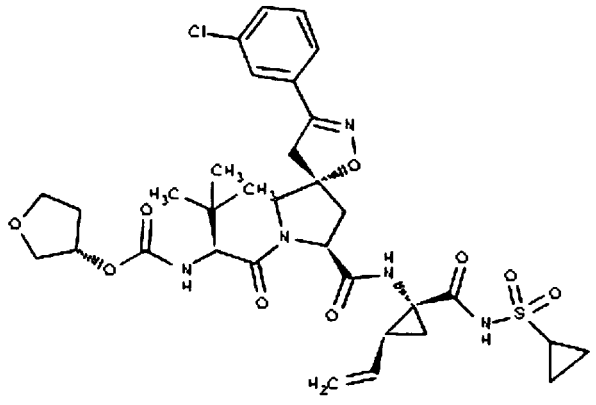
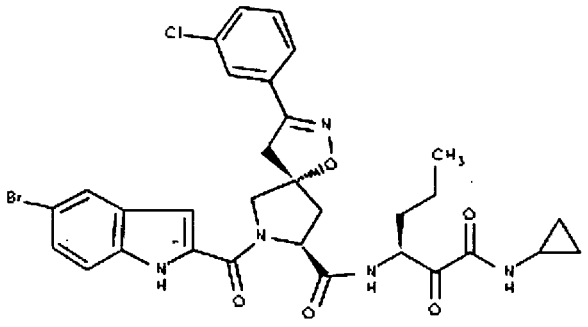
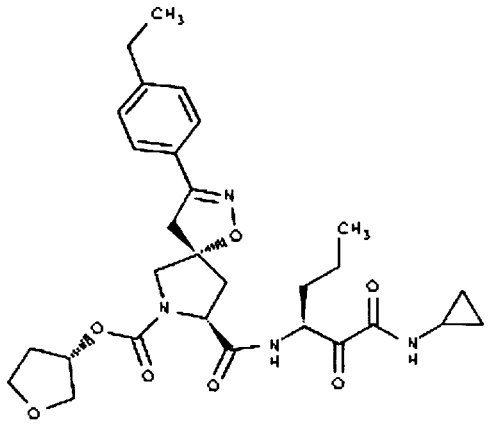
	30
	31
	32

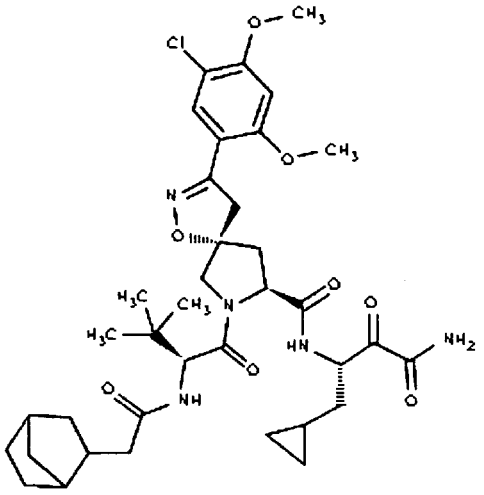
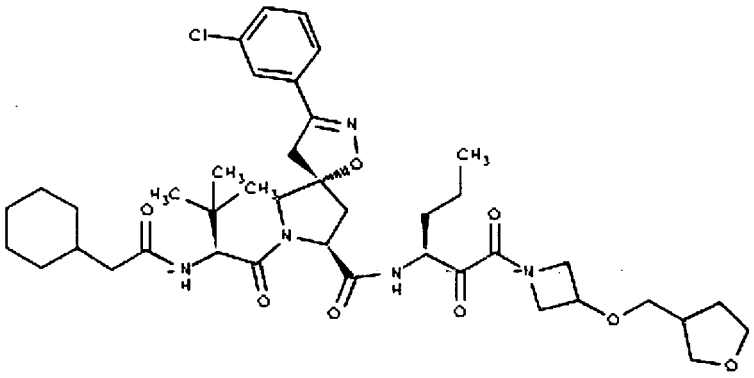
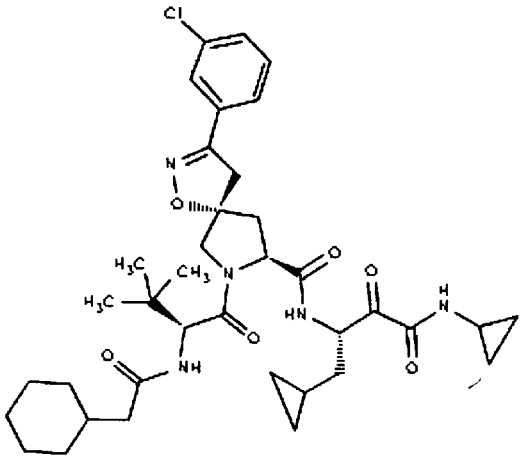
	33
	34
	35

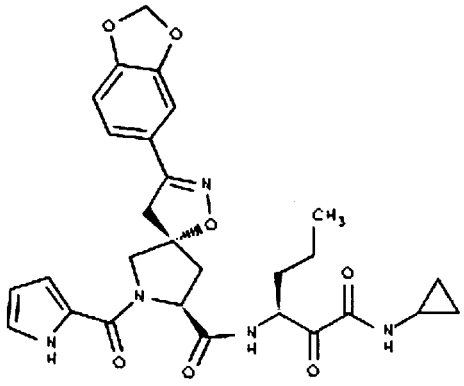
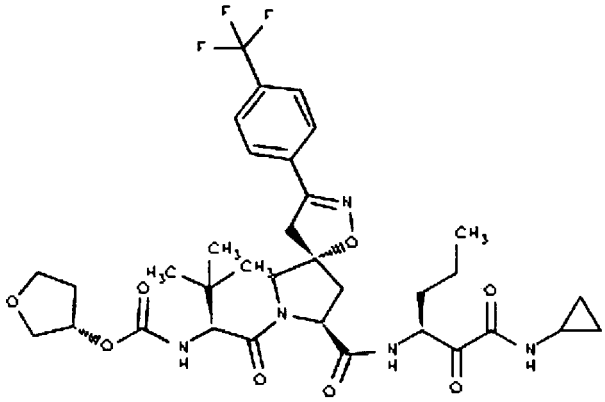
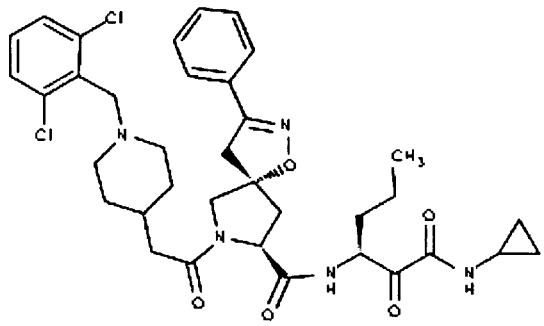
	36
	37
	38

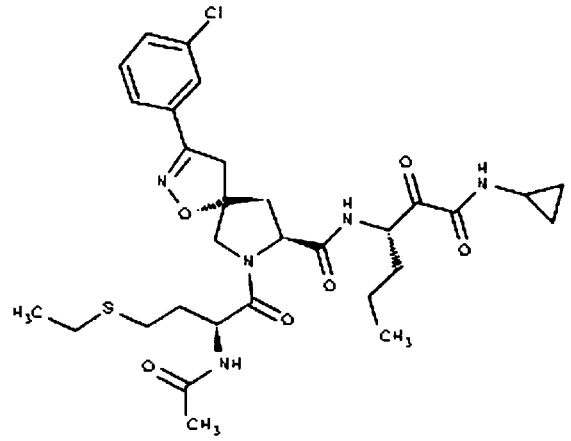
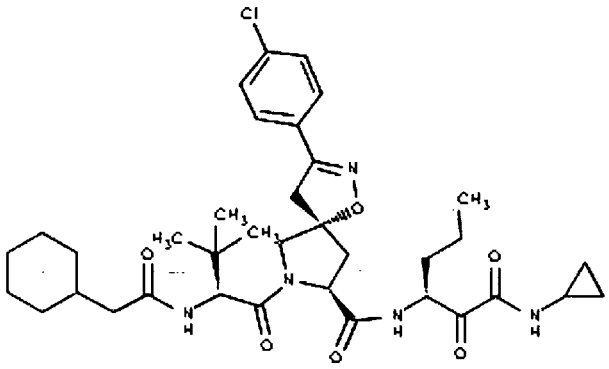
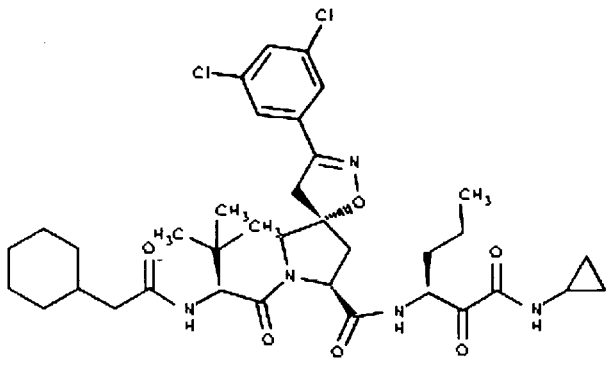
	39
	40
	41

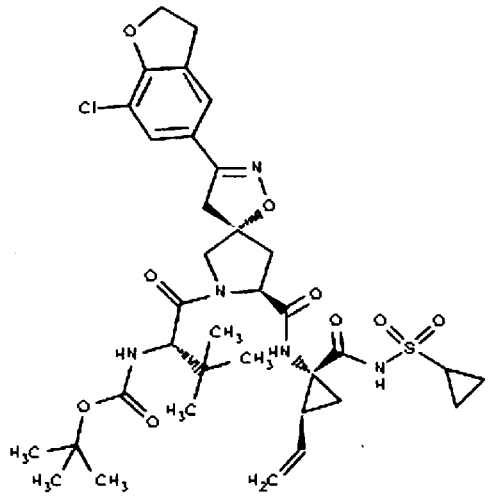
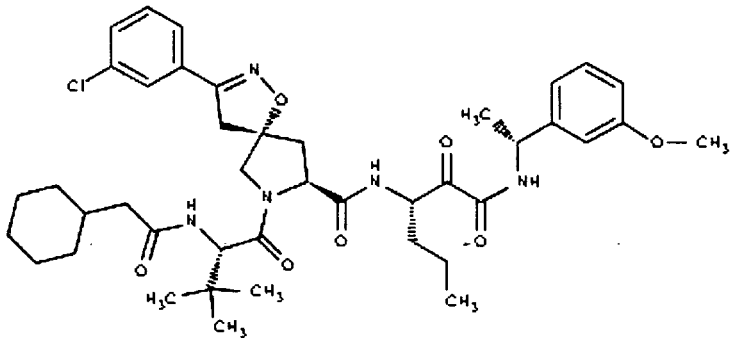
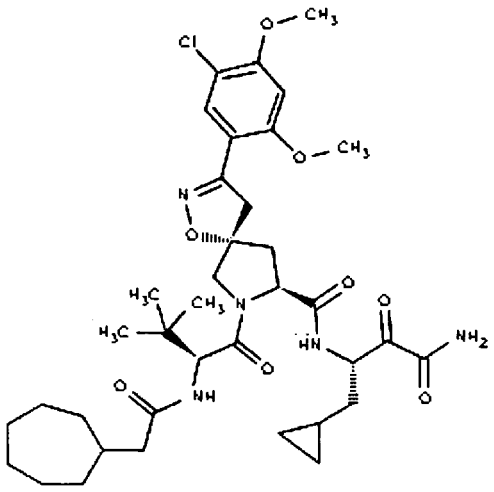
	42
	43
	44

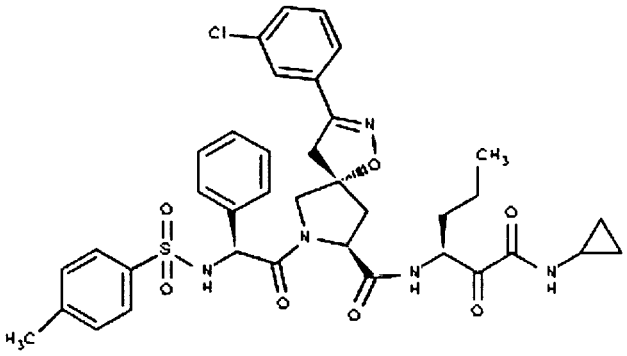
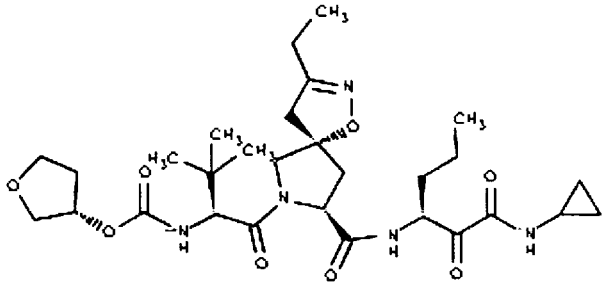
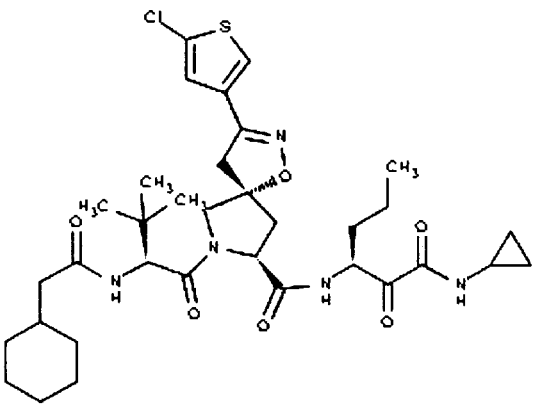
	45
	46
	47

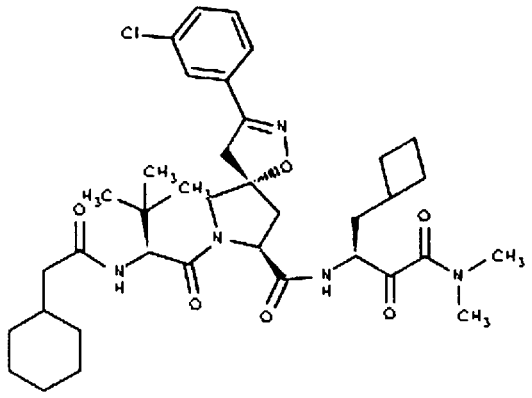
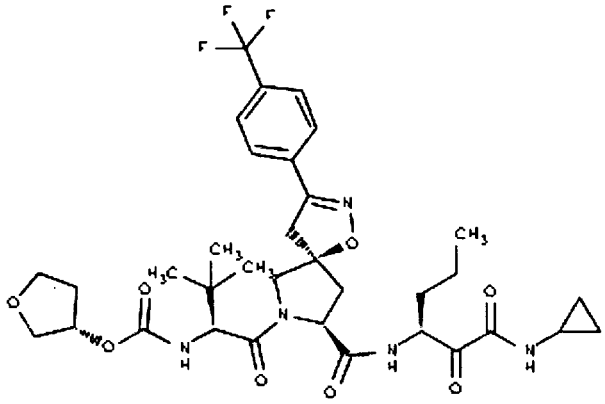
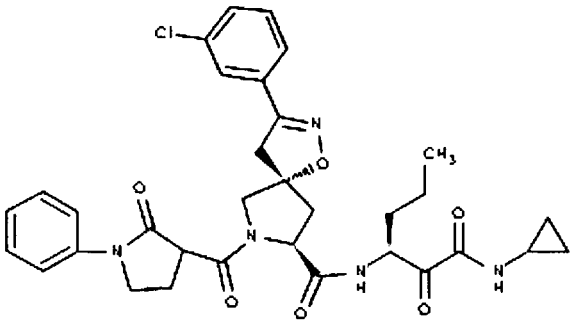
	48
	49
	50

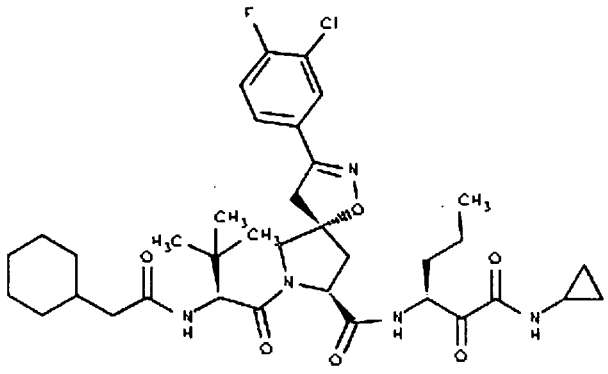
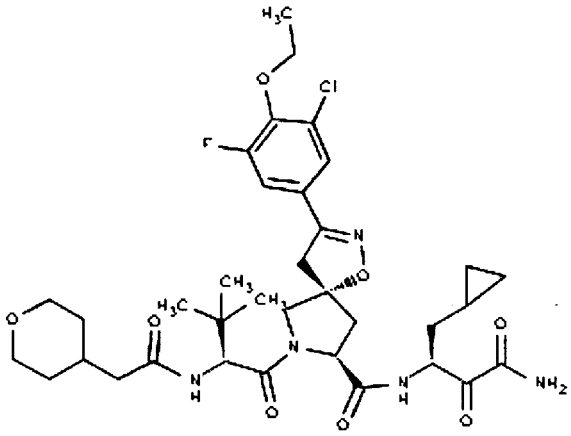
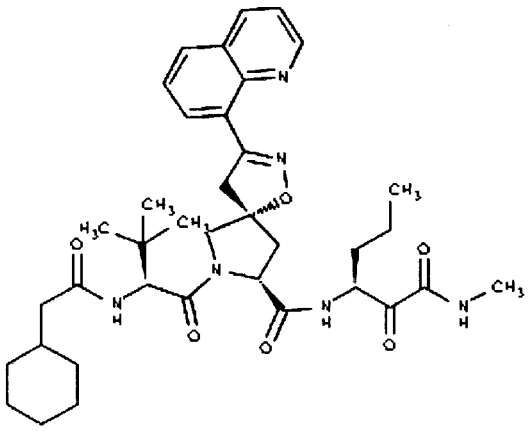
	51
	52
	53

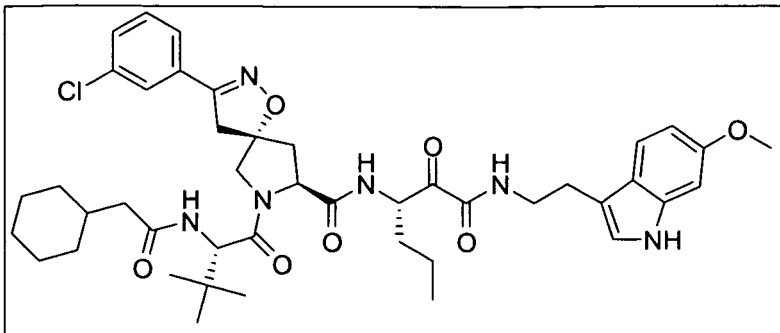
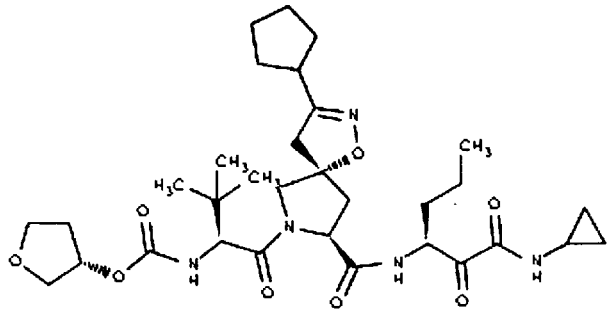
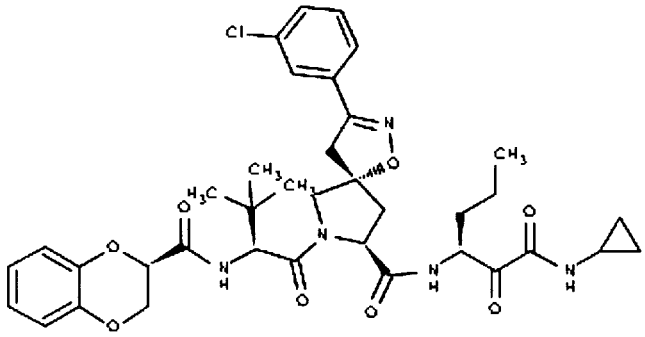
	54
	55
	56

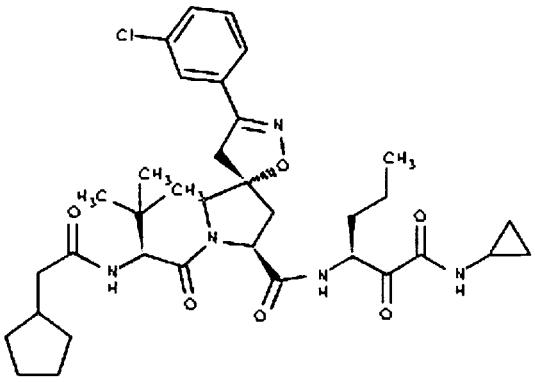
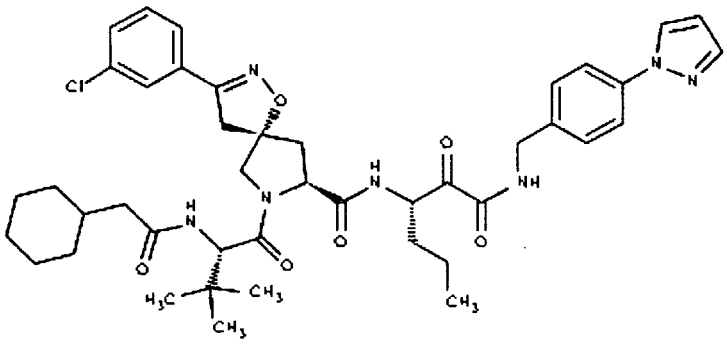
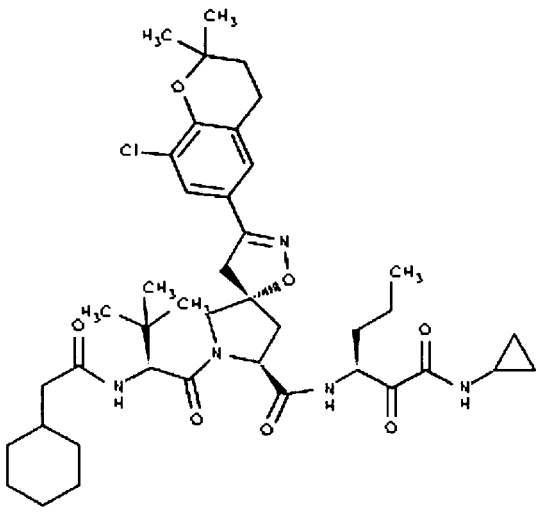
	57
	58
	59

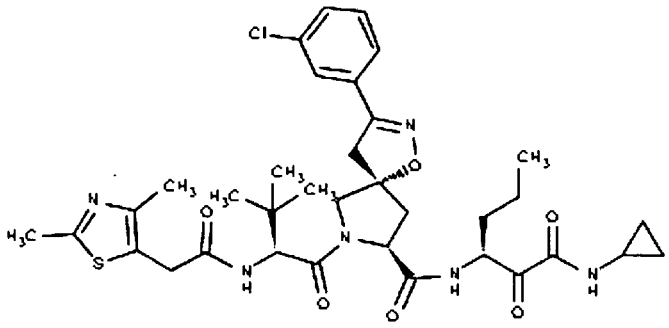
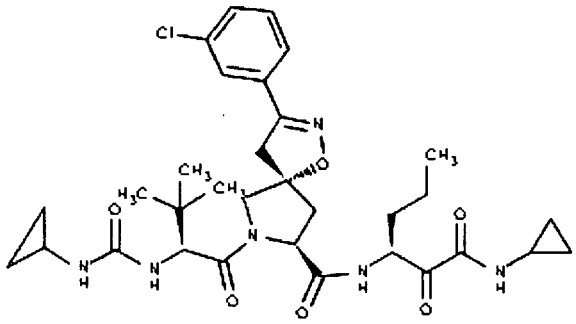
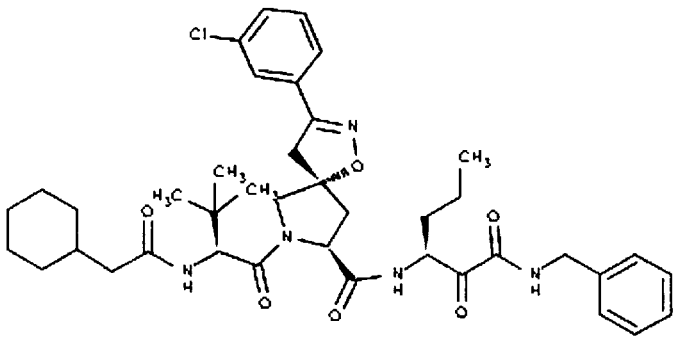
	60
	61
	62

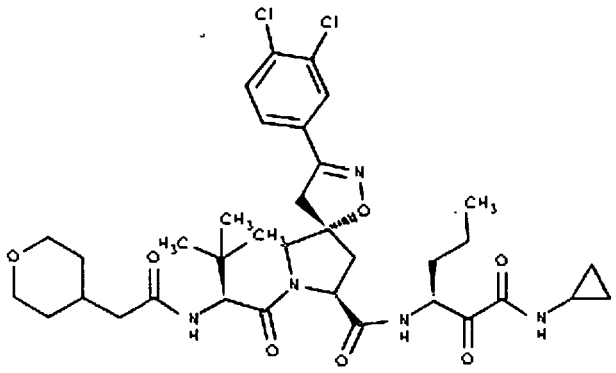
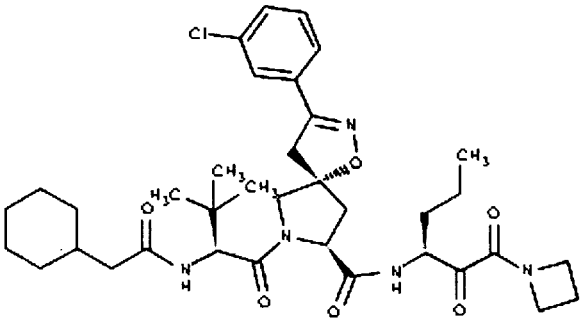
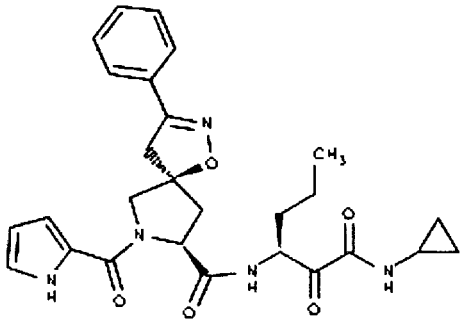
	63
	64
	65

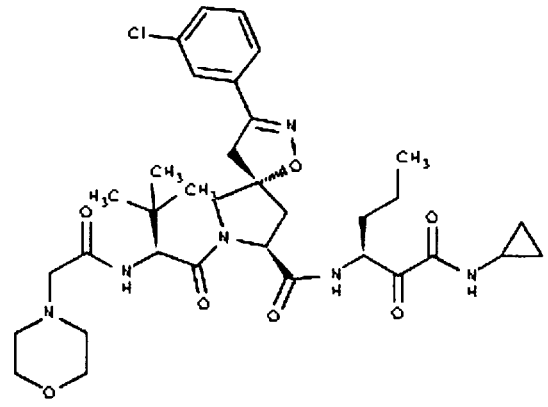
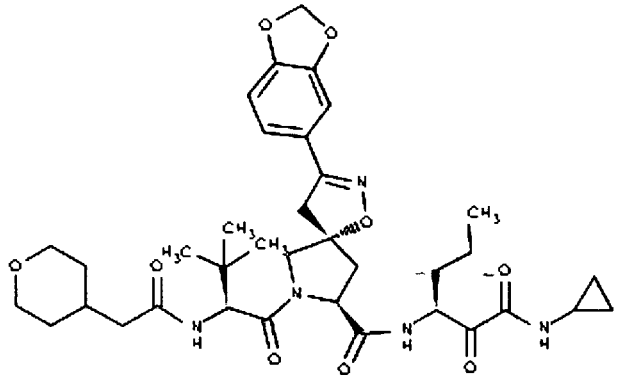
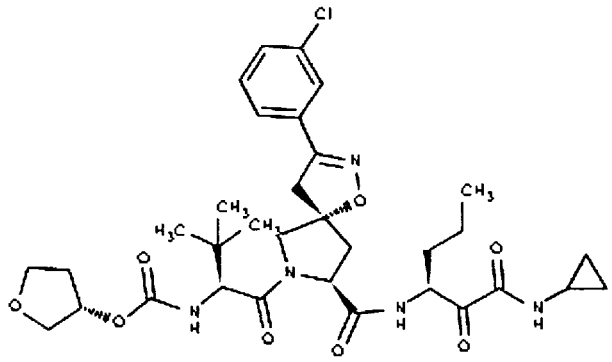
	66
	67
	68

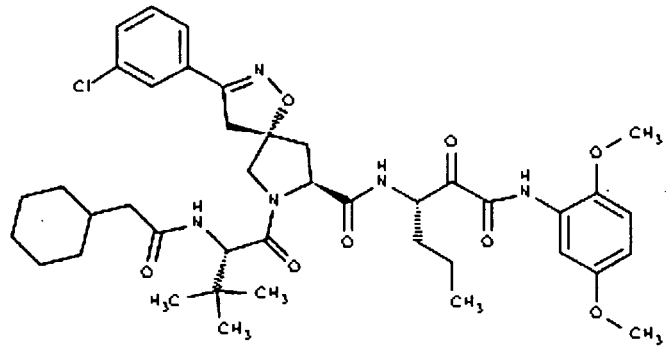
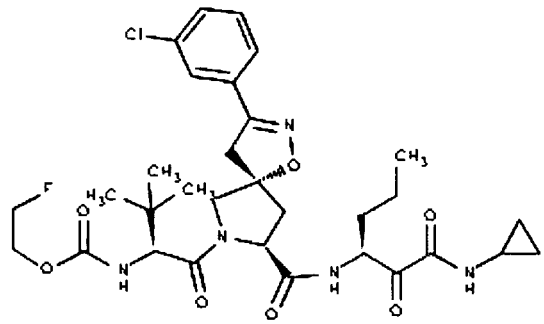
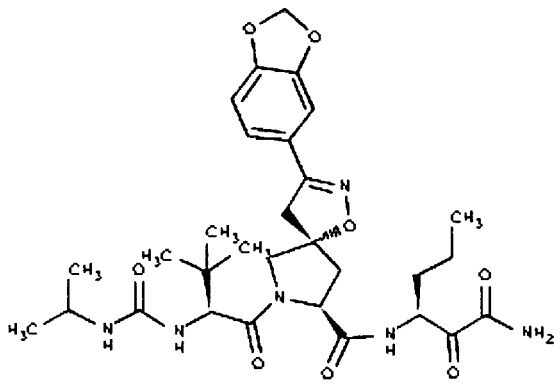
	69
	70
	71

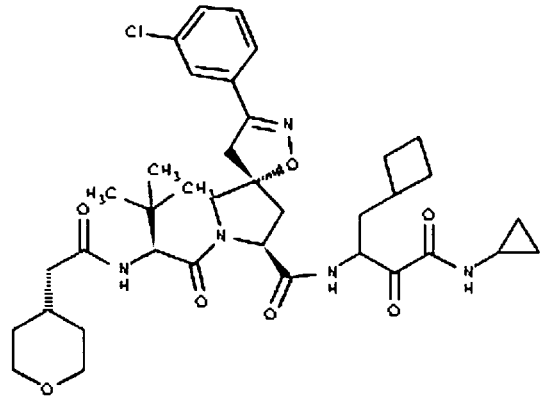
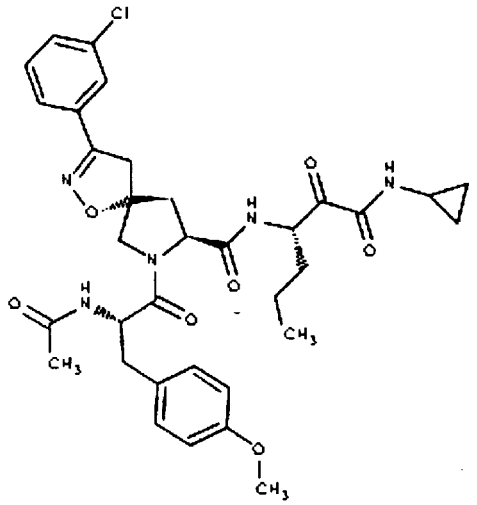
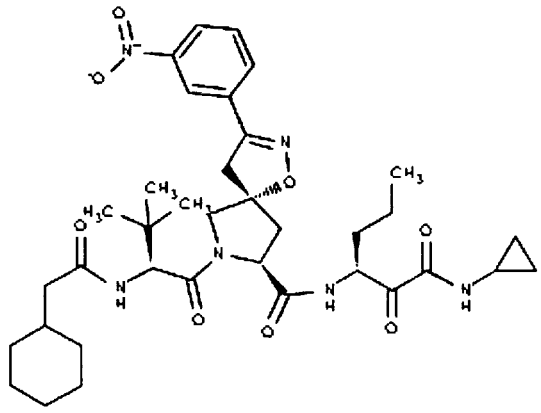
	72
	73
	74

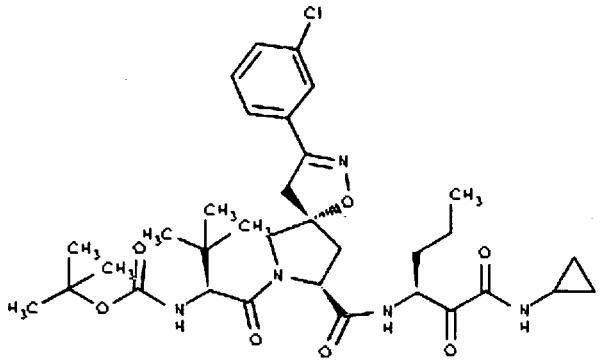
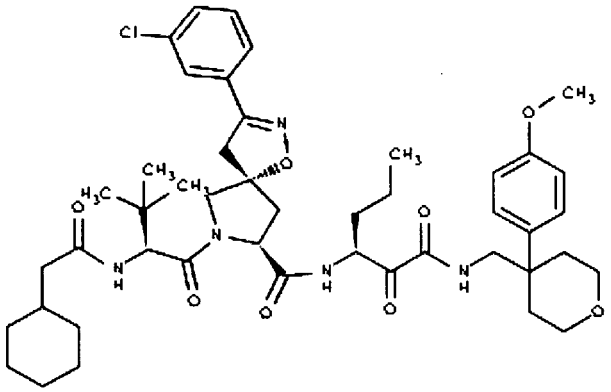
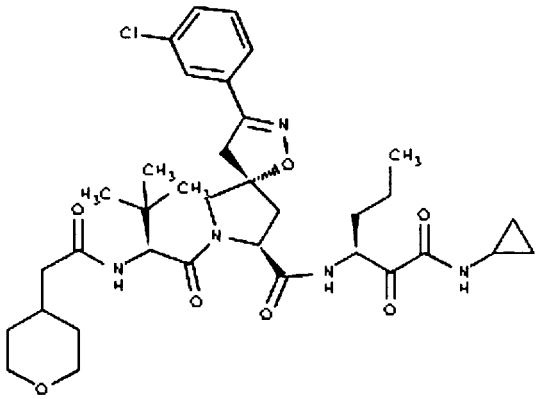
	75
	76
	77

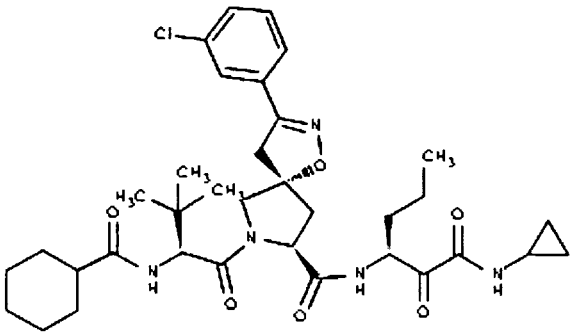
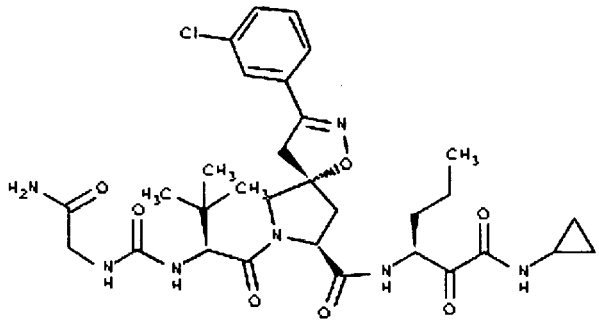
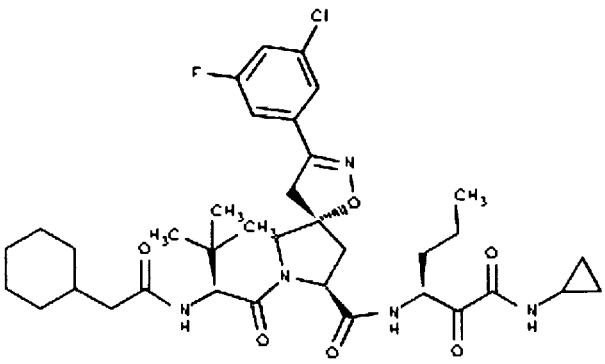
	78
	79
	80

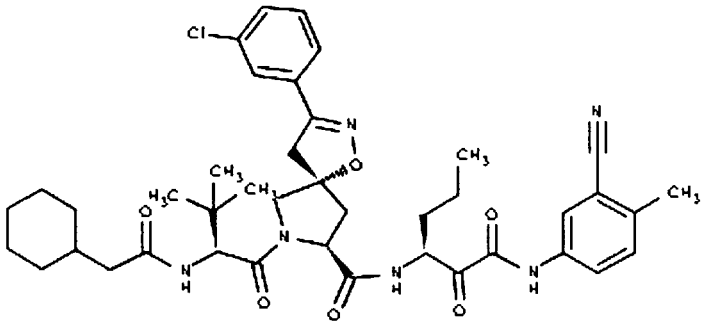
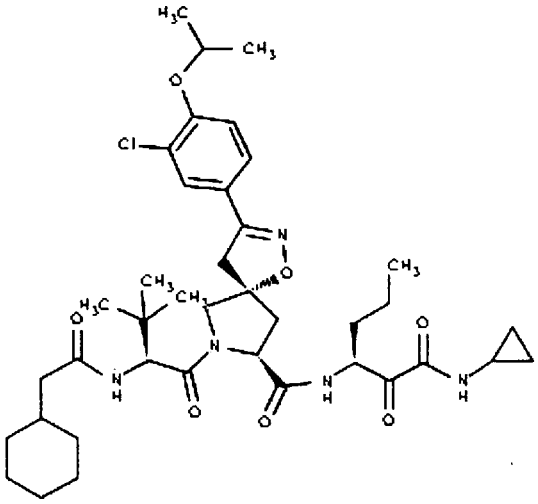
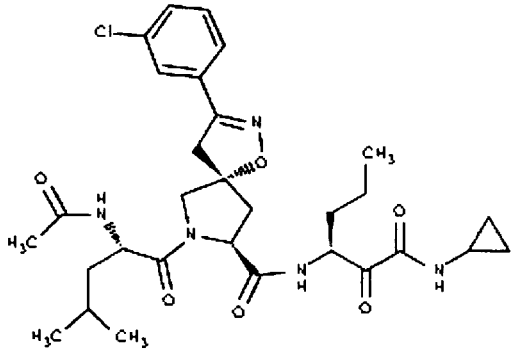
	81
	82
	83

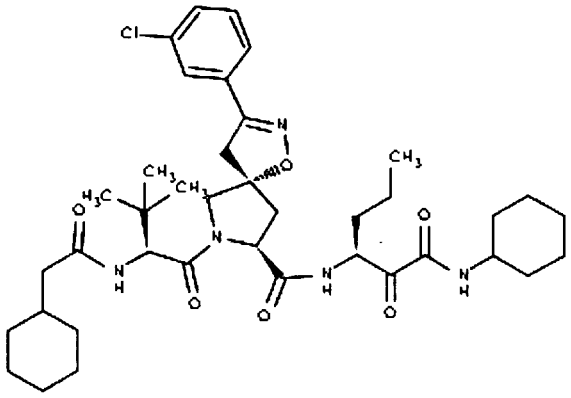
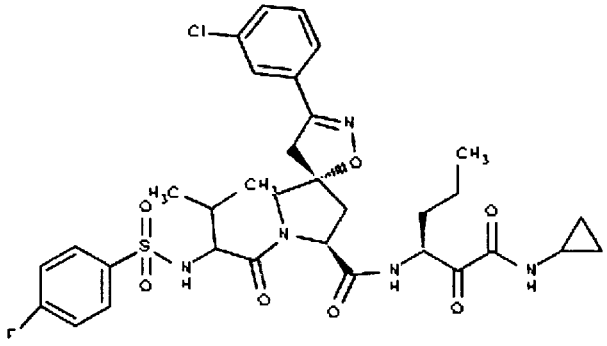
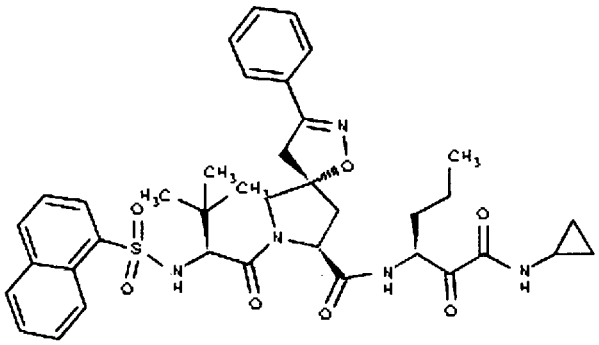
	84
	85
	86

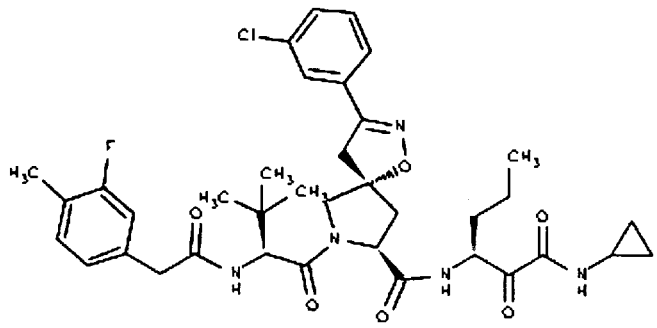
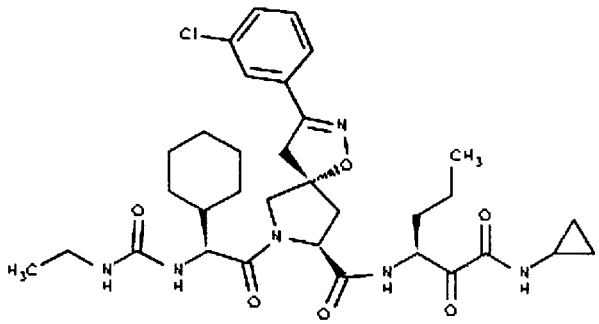
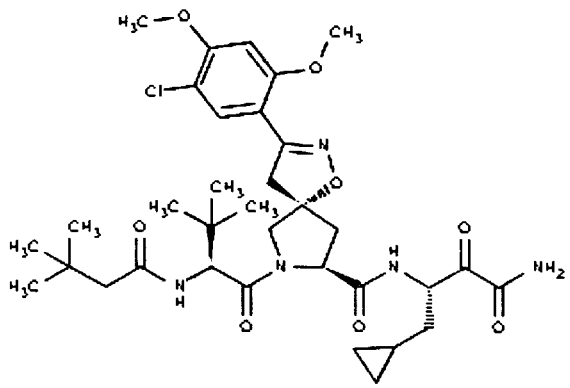
	87
	88
	89

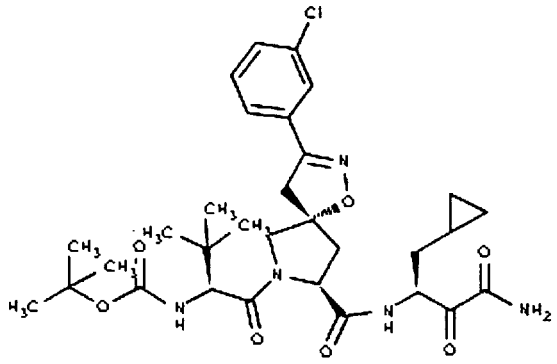
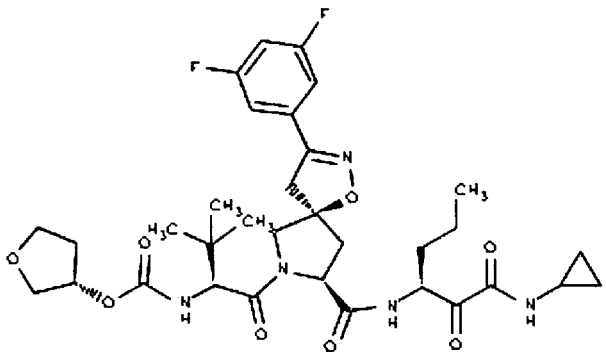
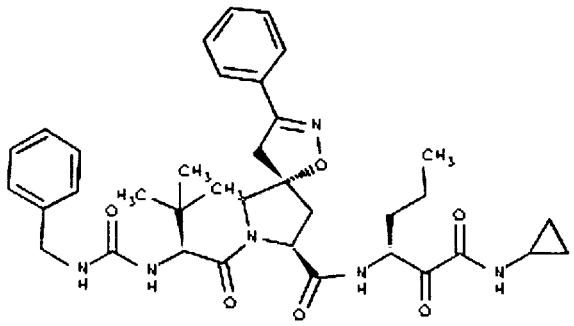
	90
	91
	92

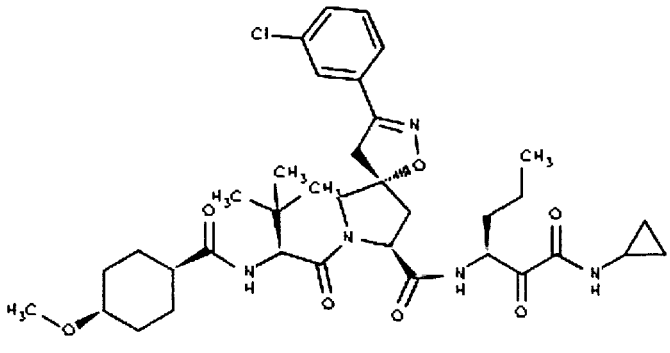
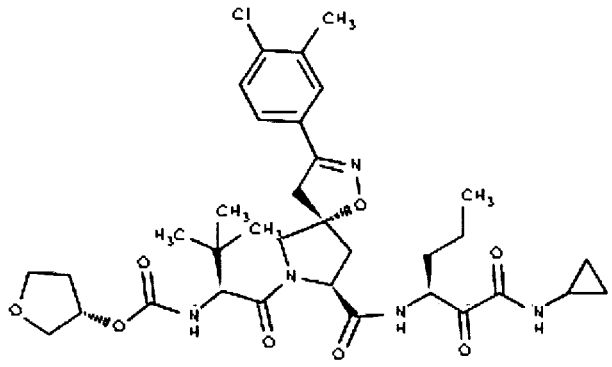
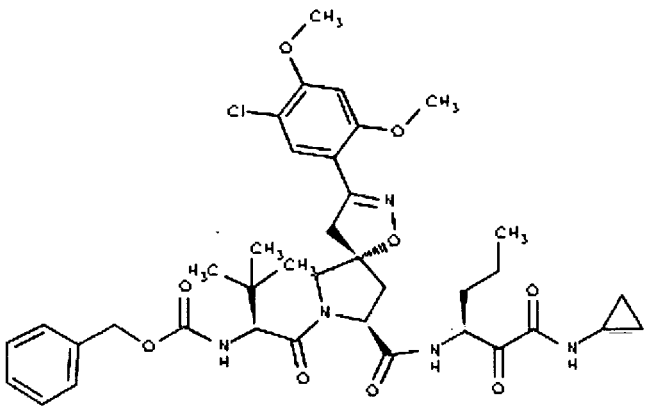
	93
	94
	95

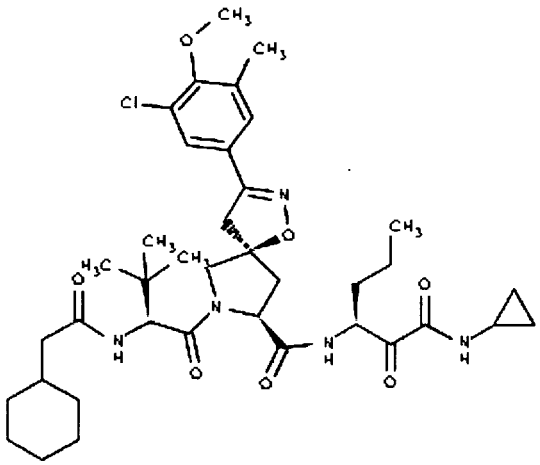
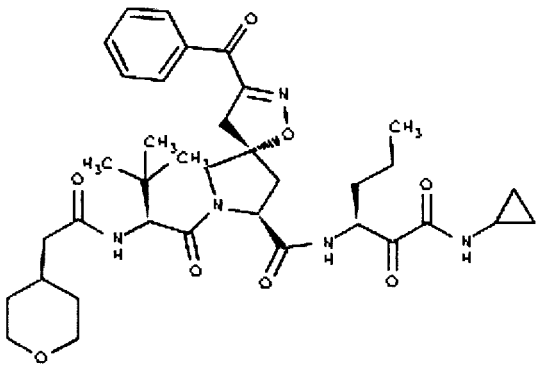
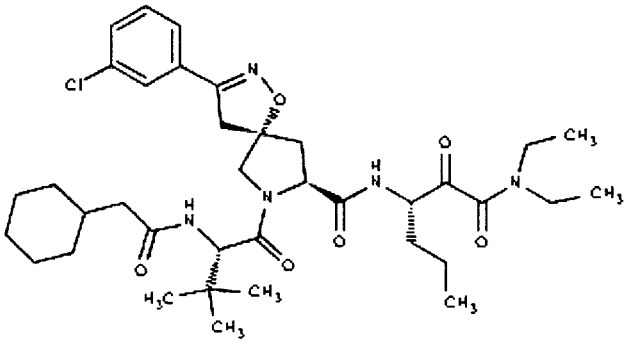
	96
	97
	98

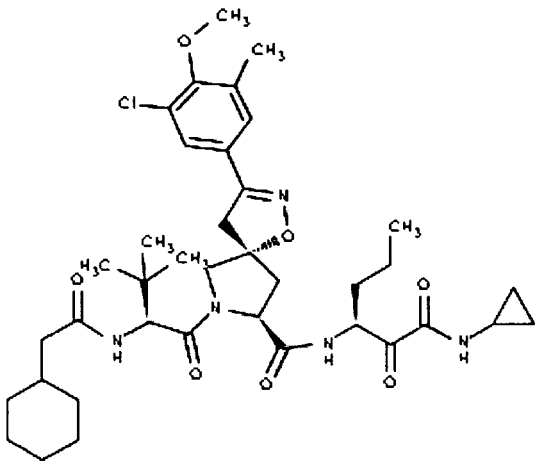
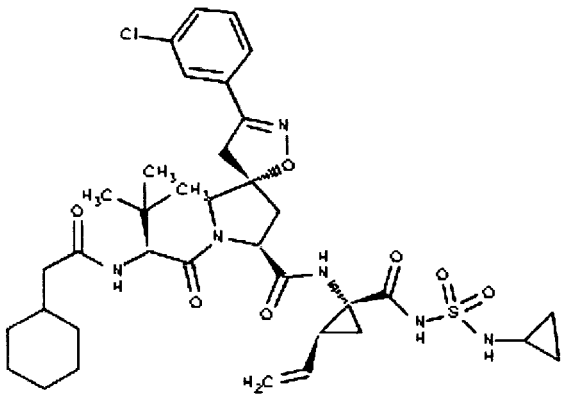
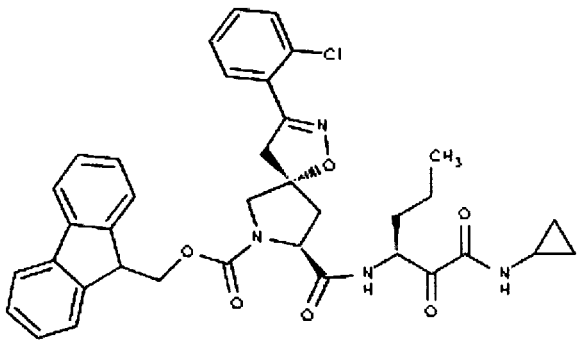
	99
	100
	101

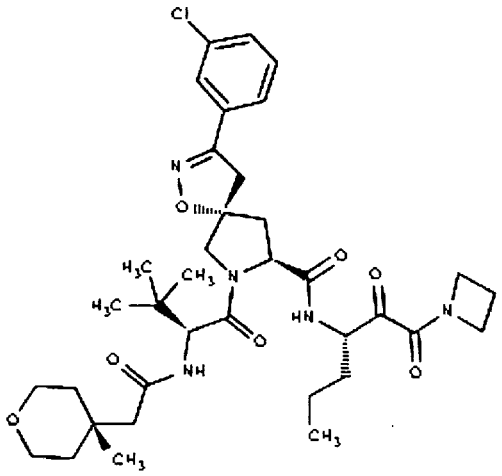
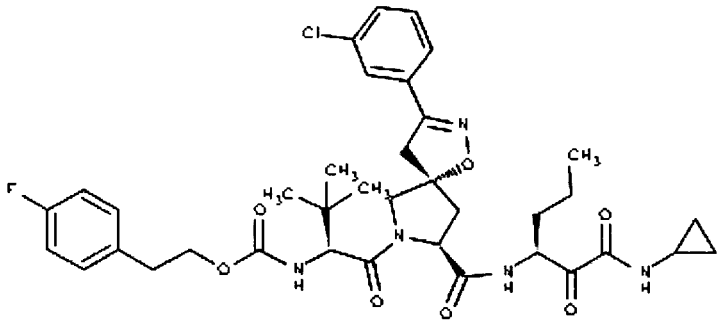
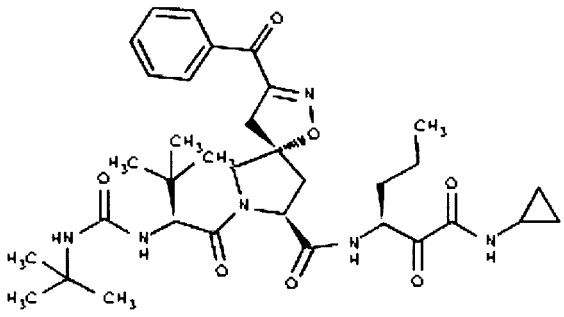
	102
	103
	104

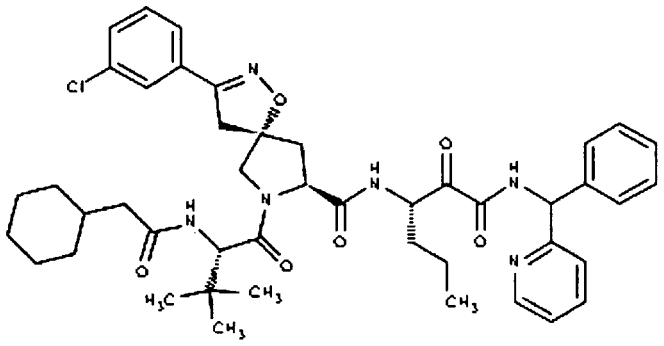
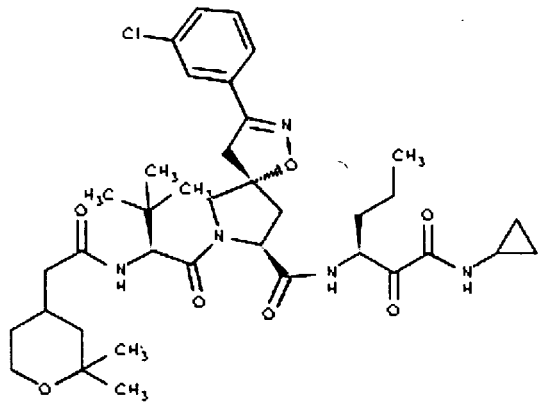
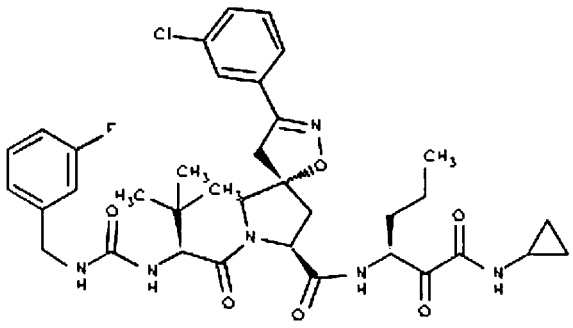
	105
	106
	107

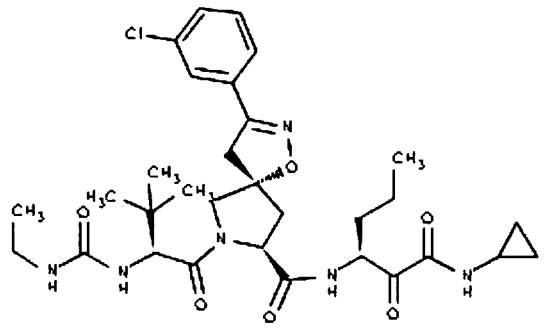
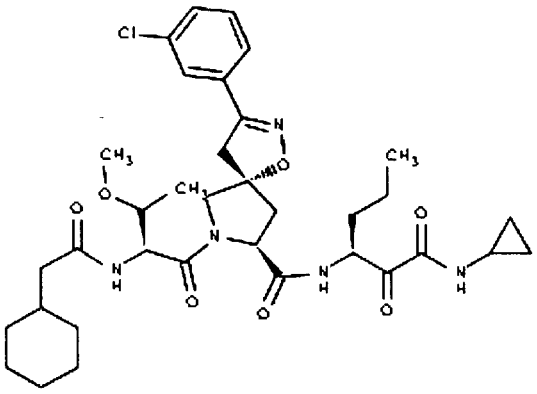
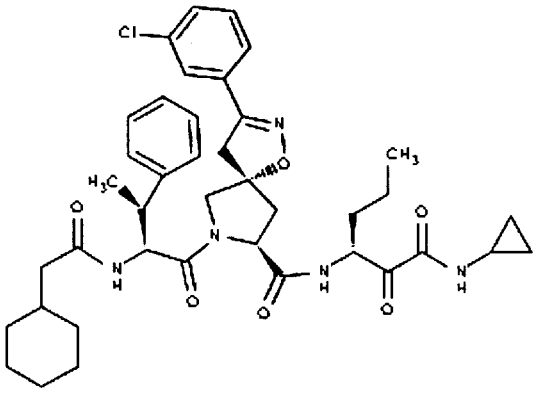
	108
	109
	110

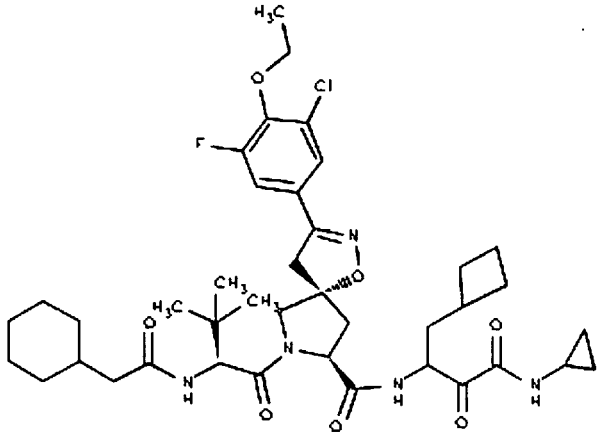
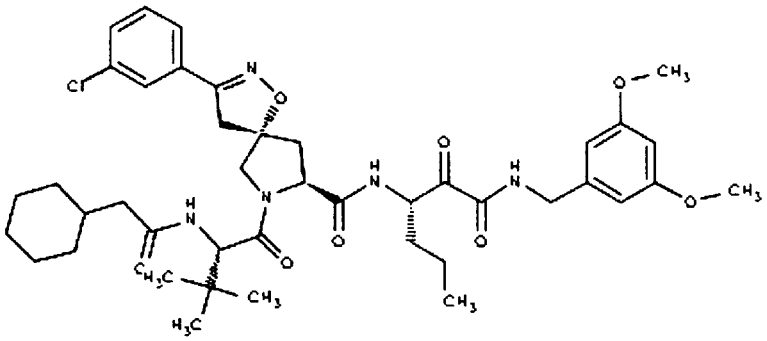
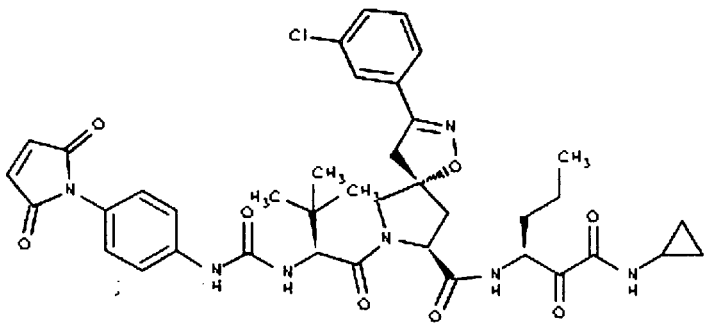
	111
	112
	113

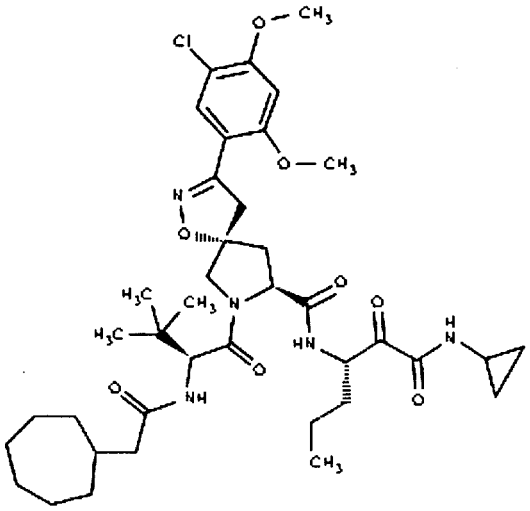
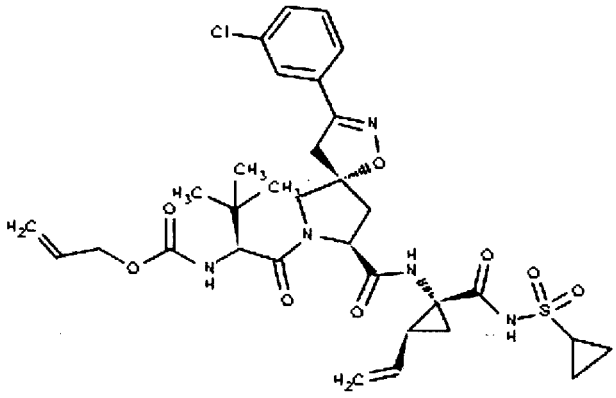
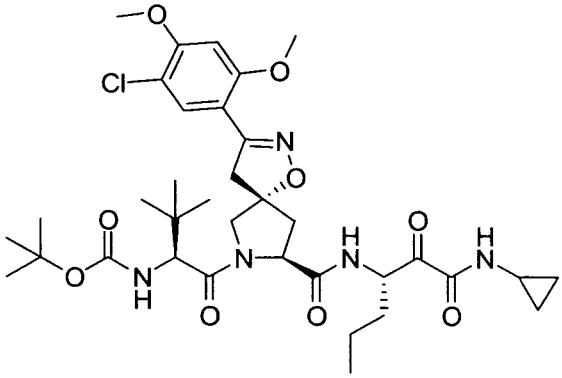
	114
	115
	116

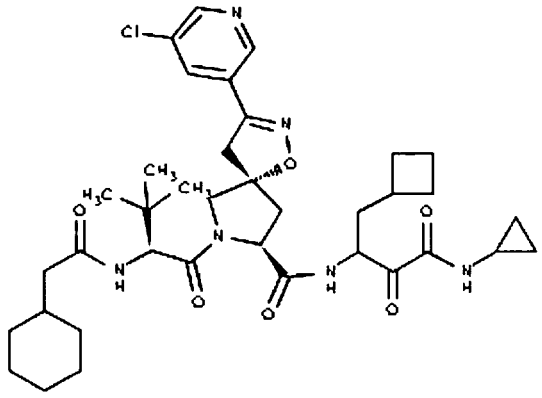
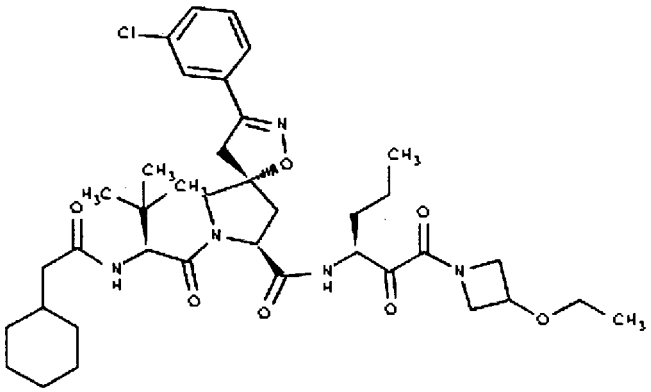
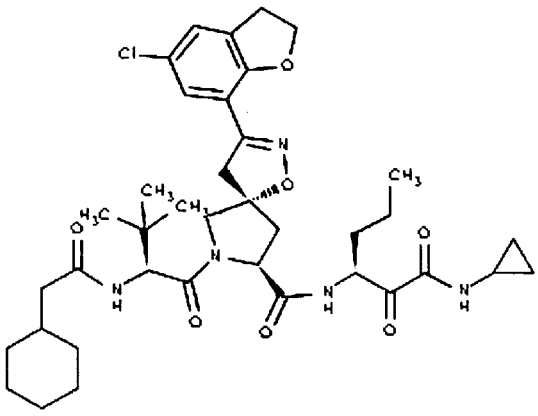
	117
	118
	119

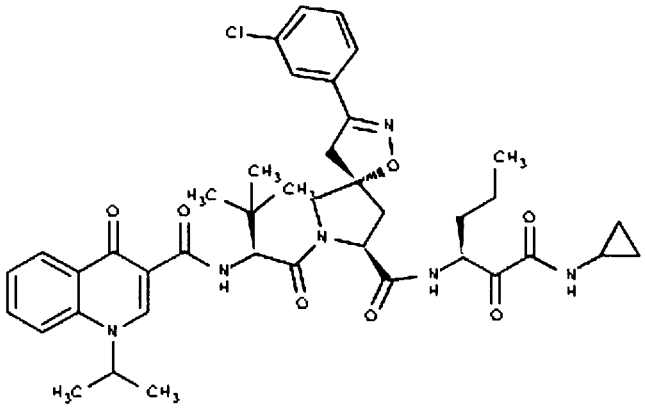
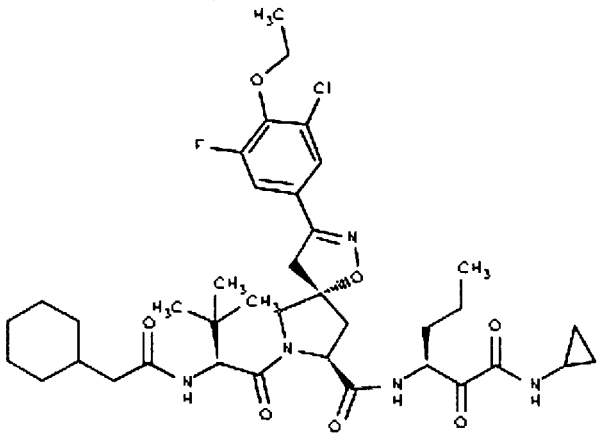
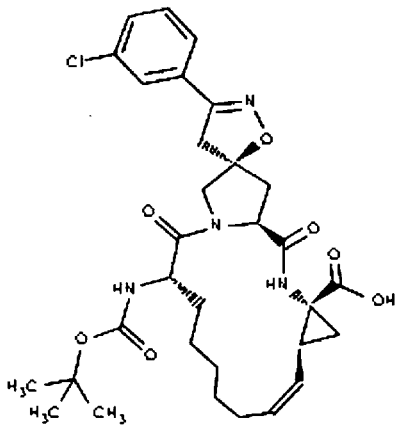
	120
	121
	122

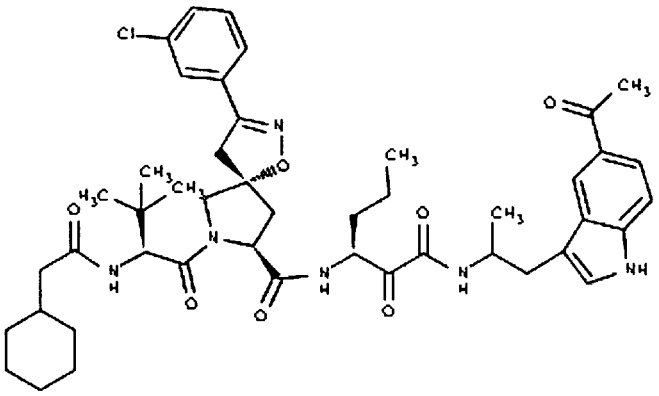
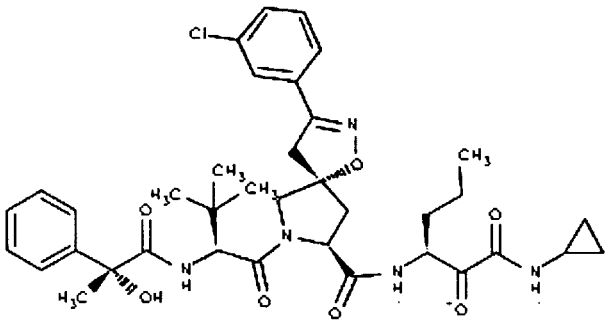
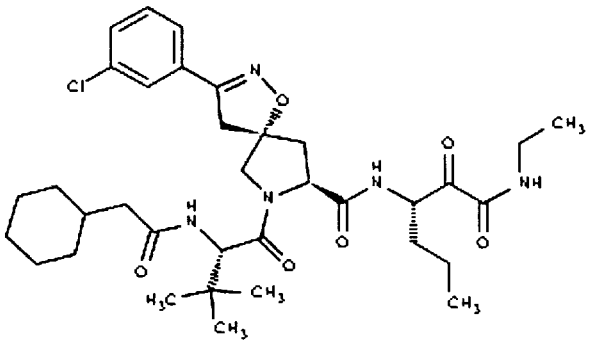
	123
	124
	125

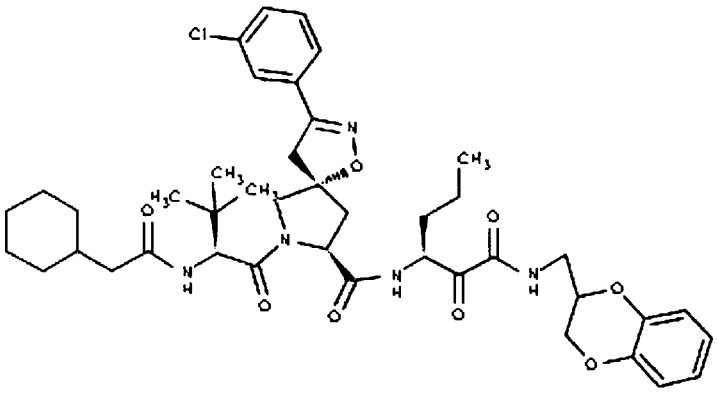
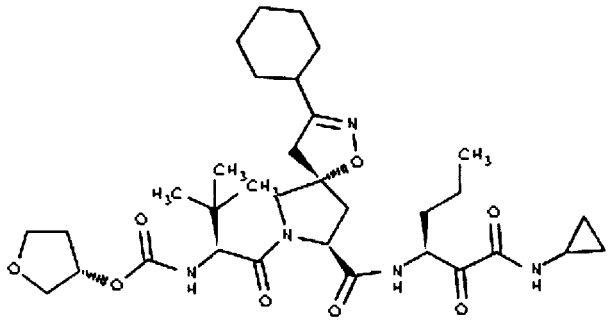
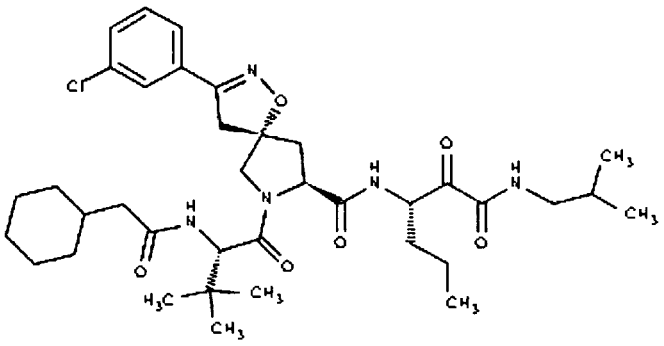
	126
	127
	128

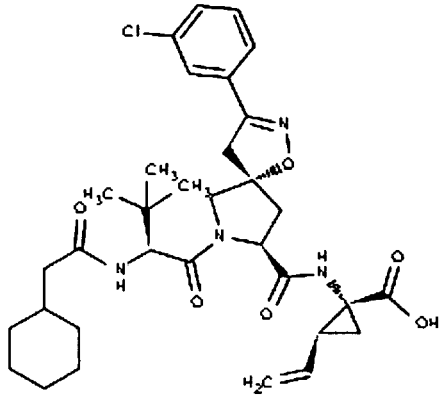
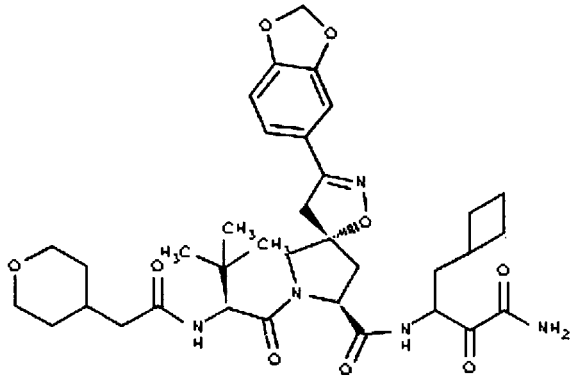
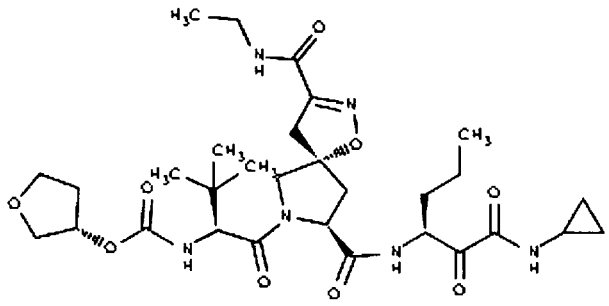
	129
	130
	131

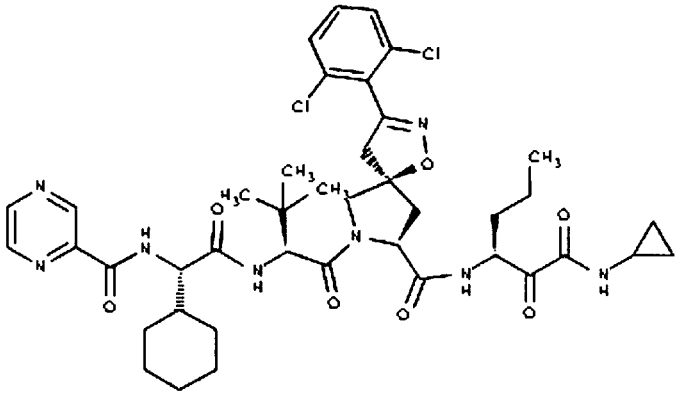
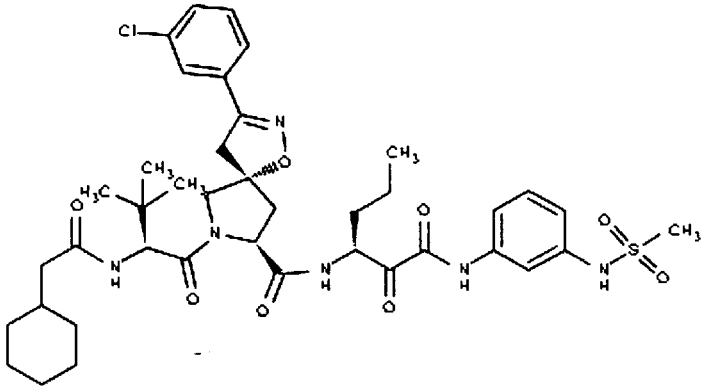
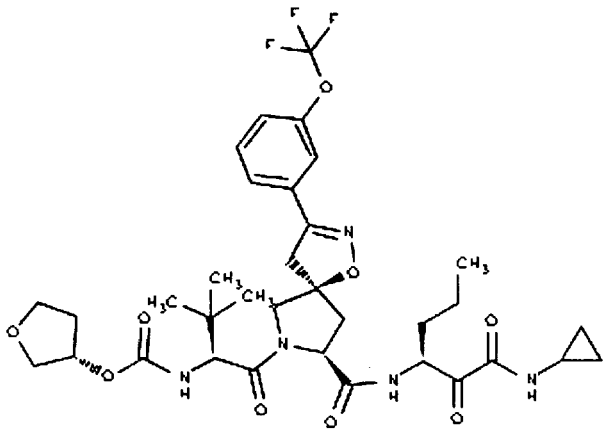
	132
	133
	134

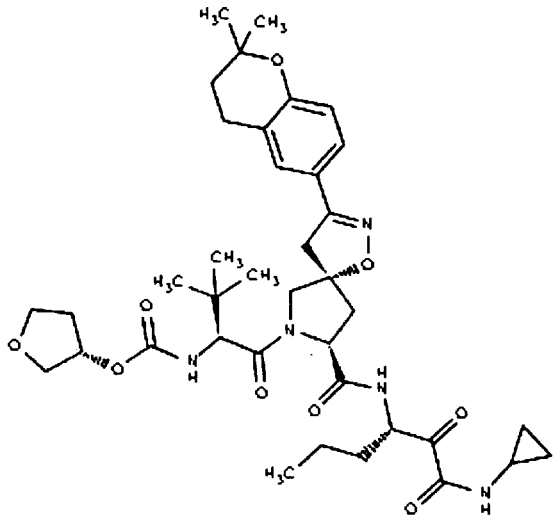
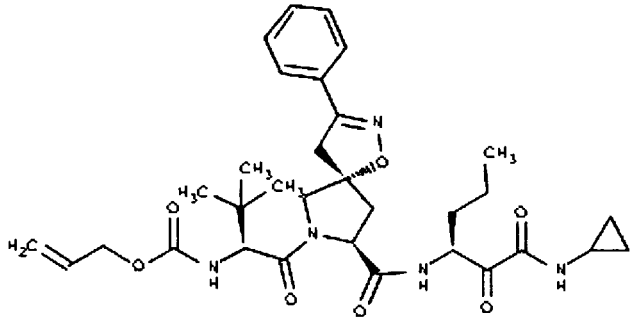
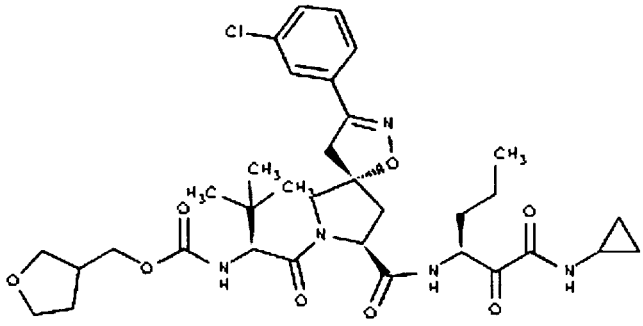
	135
	136
	137

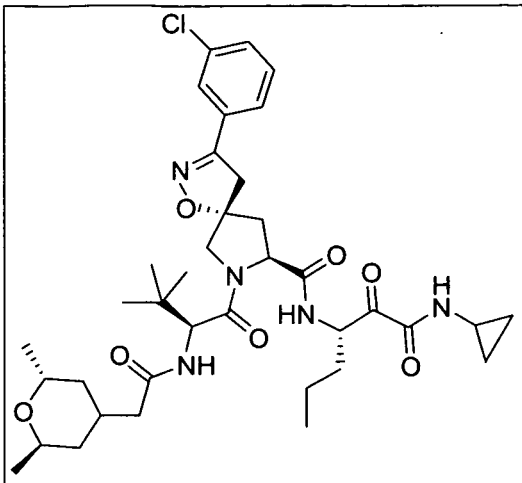
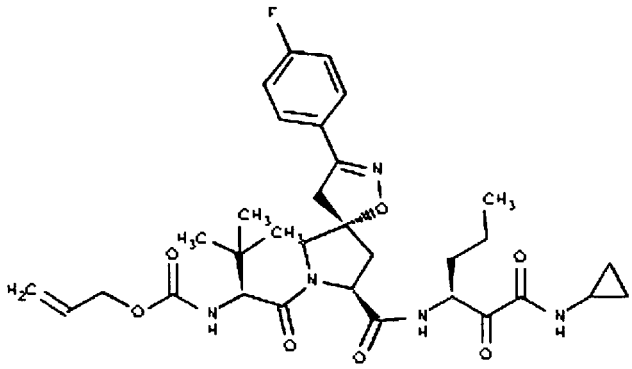
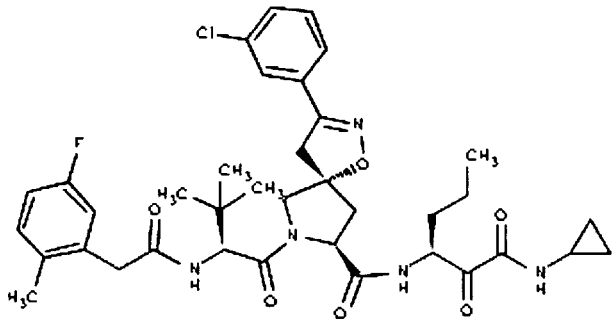
	138
	139
	140

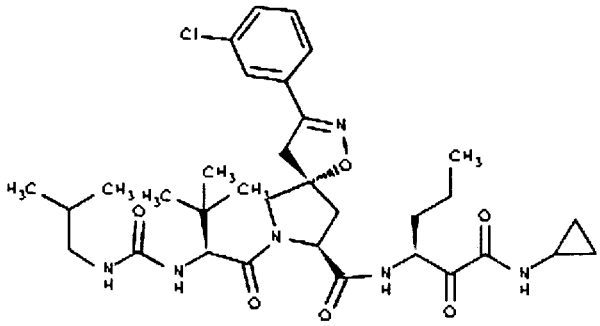
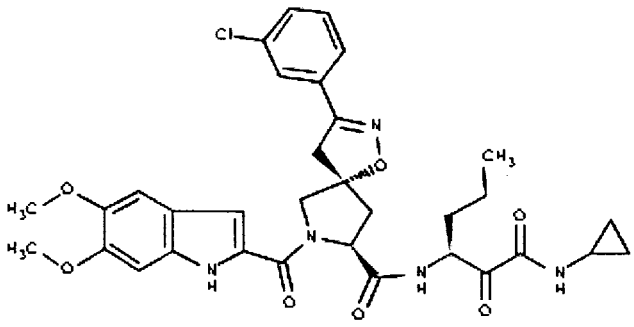
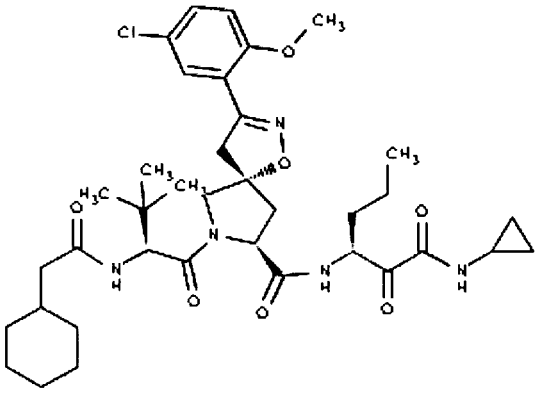
	141
	142
	143

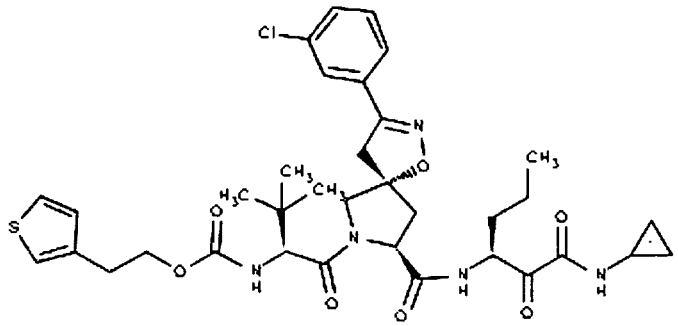
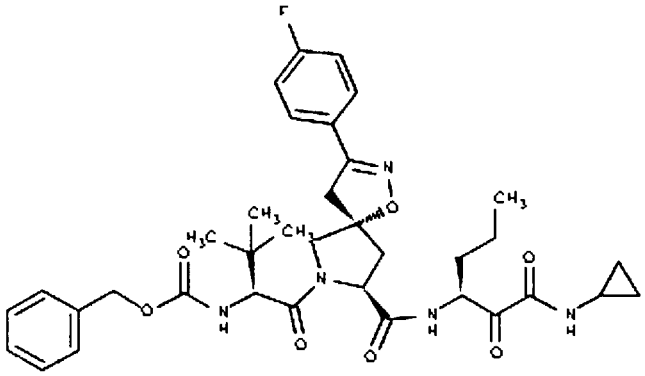
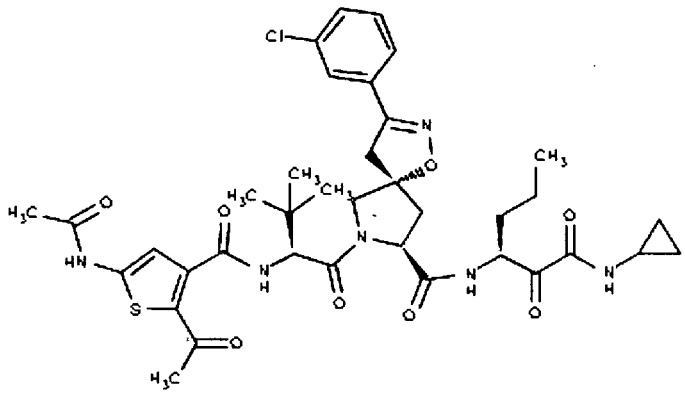
	144
	145
	146

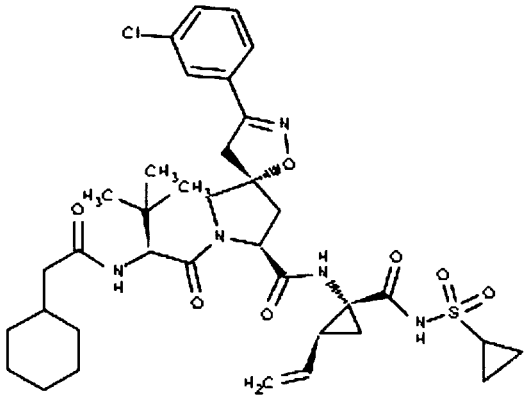
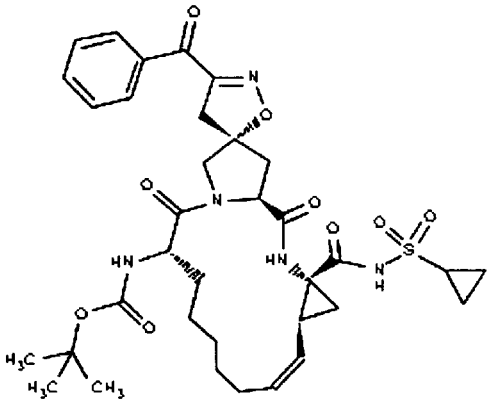
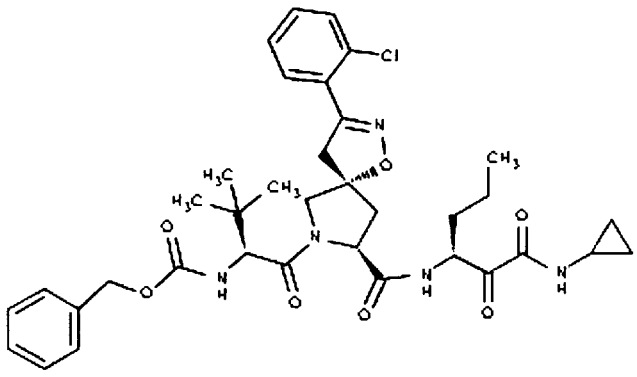
	147
	148
	149

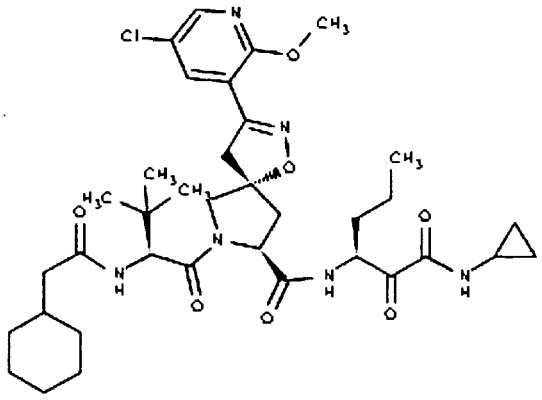
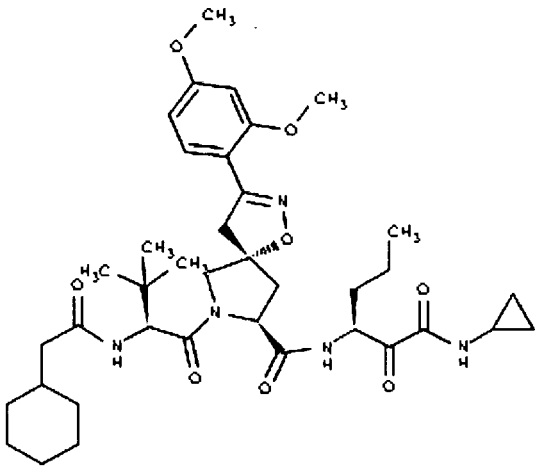
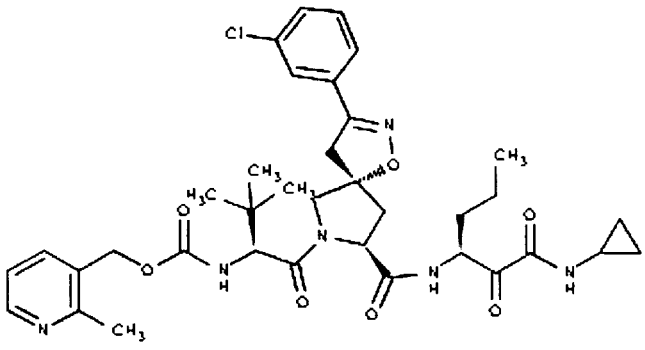
	150
	151
	152

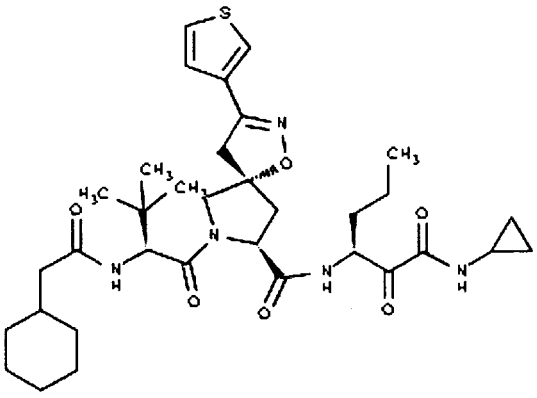
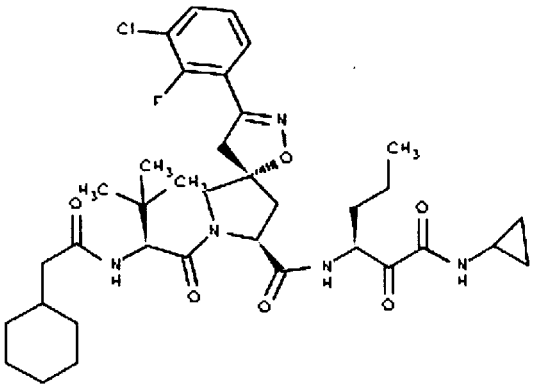
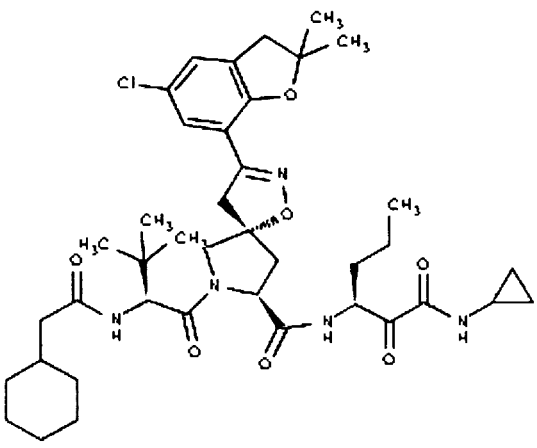
	153
	154
	155

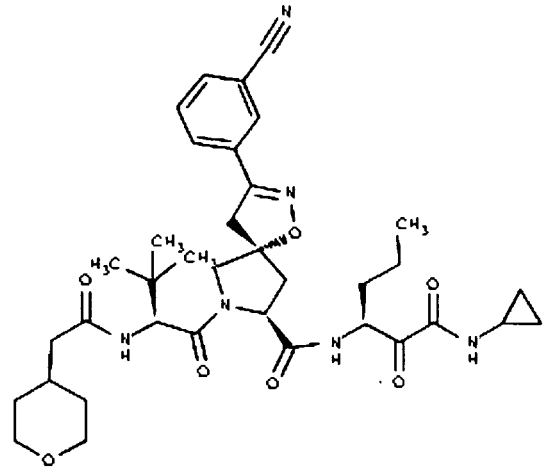
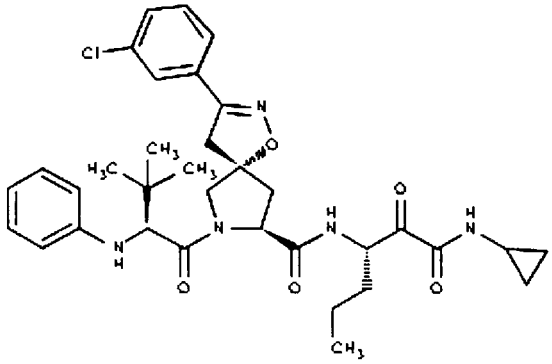
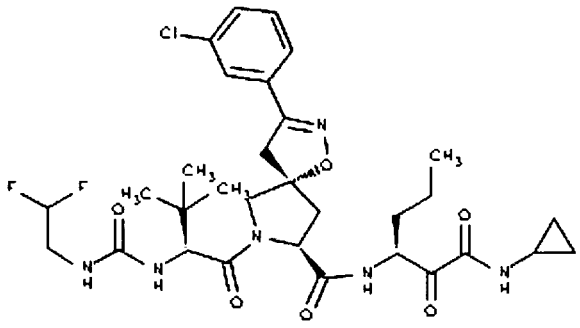
	156
	157
	158

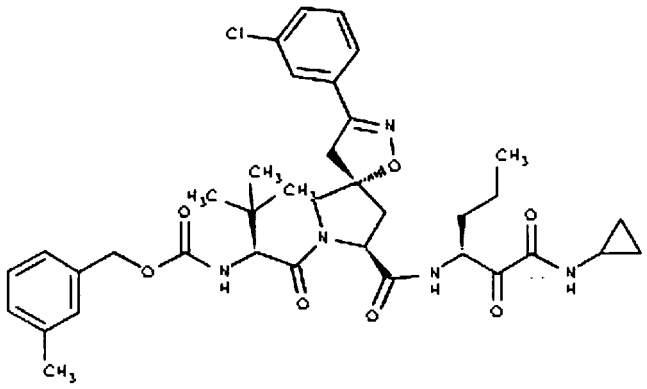
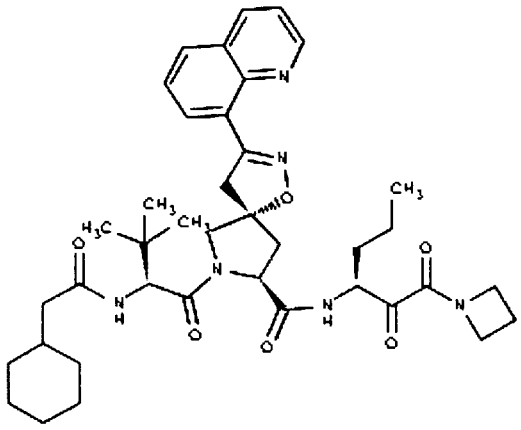
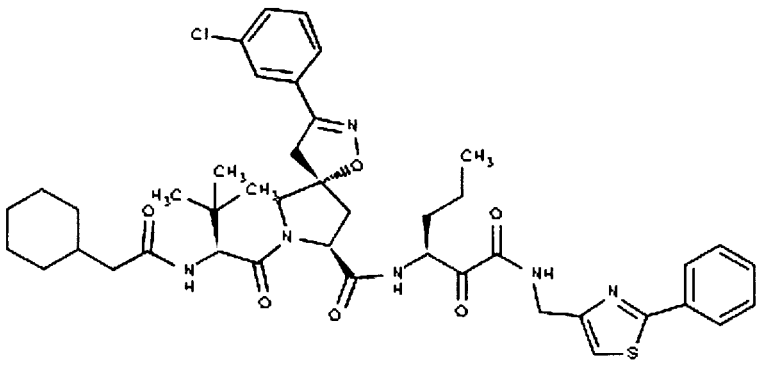
	159
	160
	161

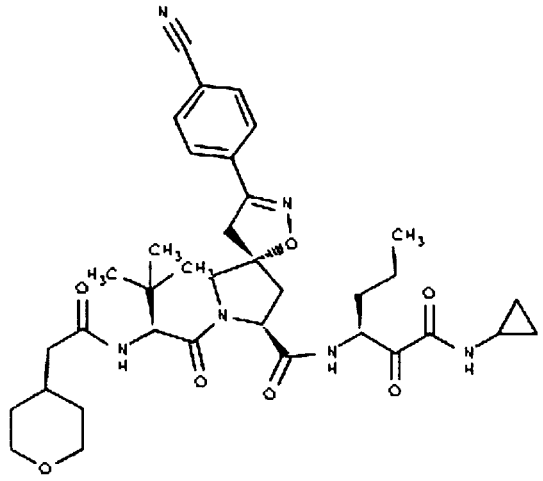
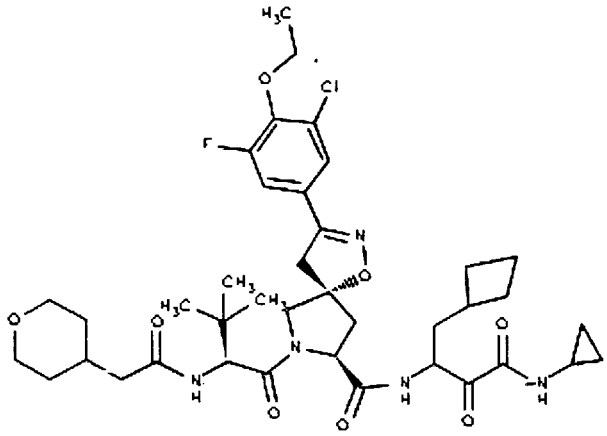
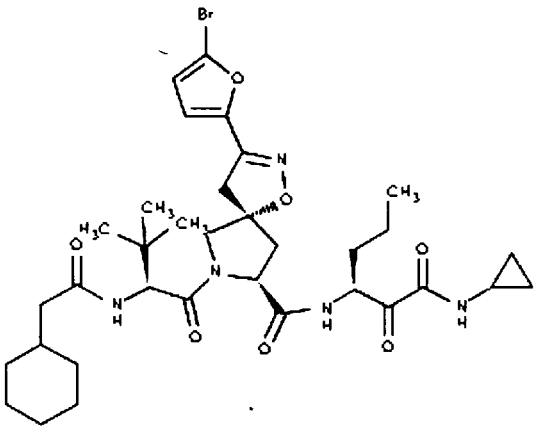
	162
	163
	164

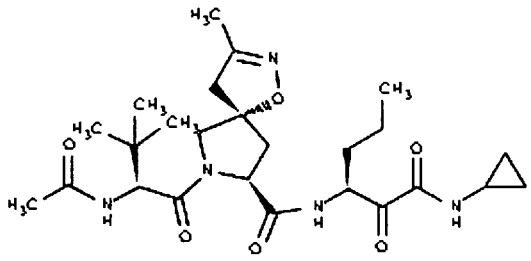
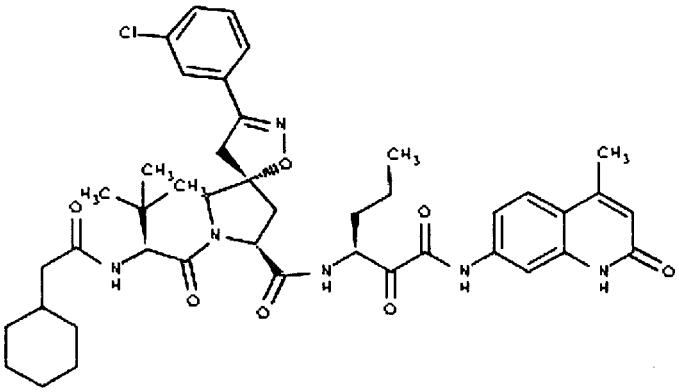
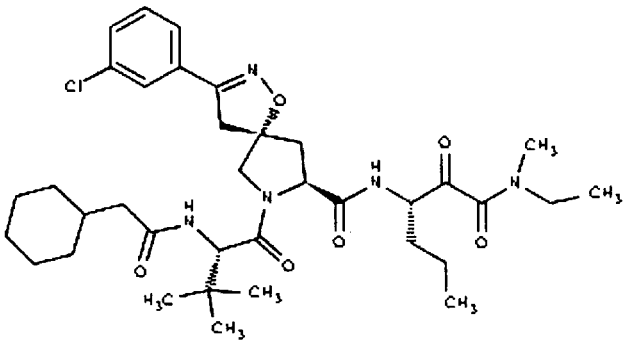
	165
	166
	167

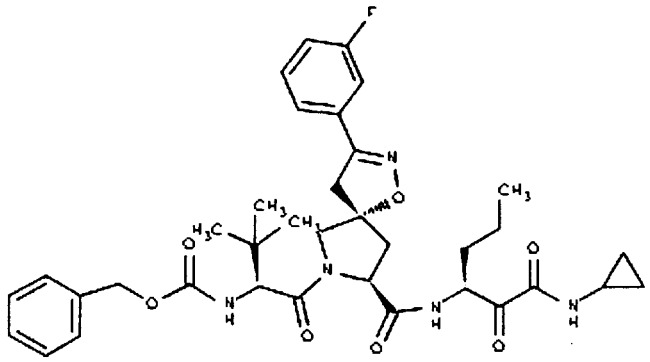
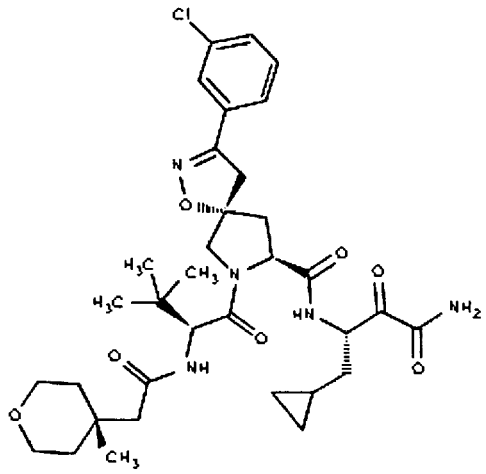
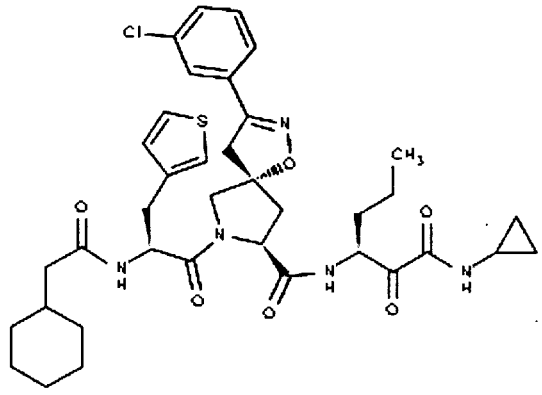
	168
	169
	170

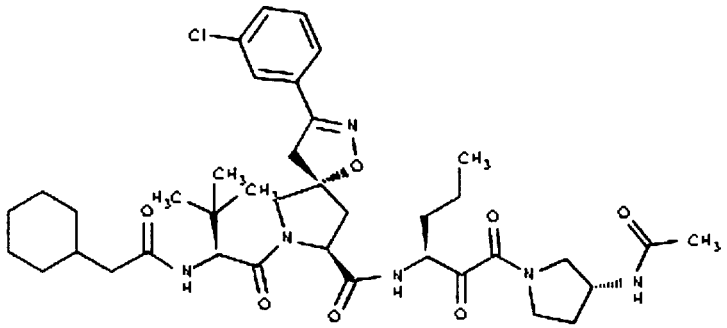
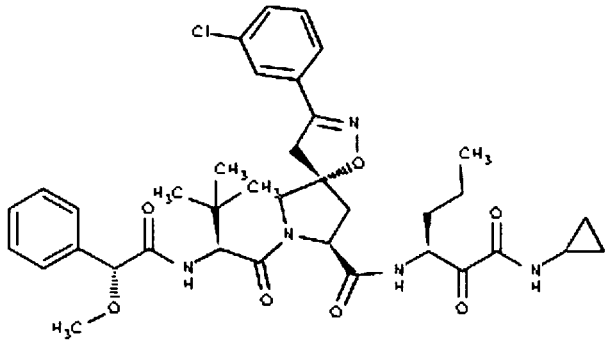
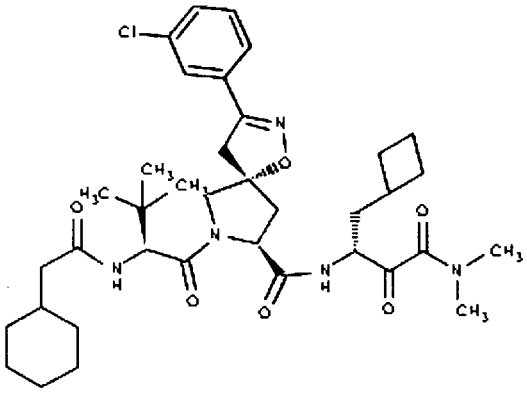
	171
	172
	173

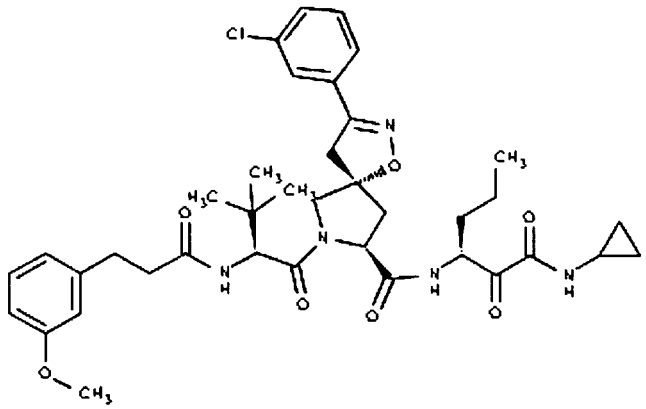
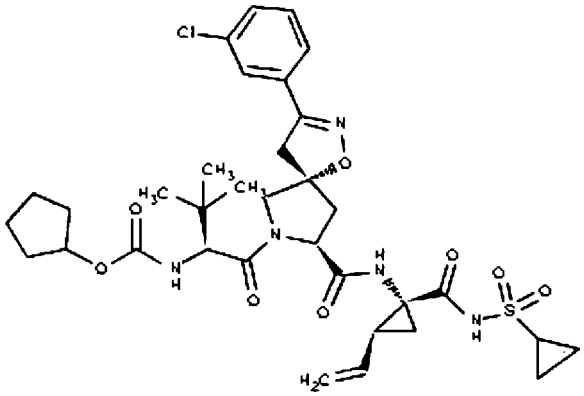
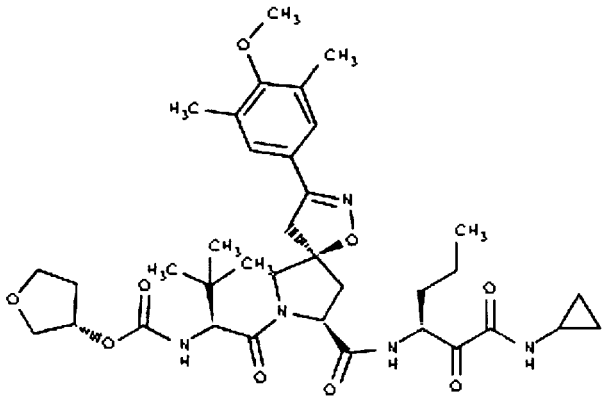
	174
	175
	176

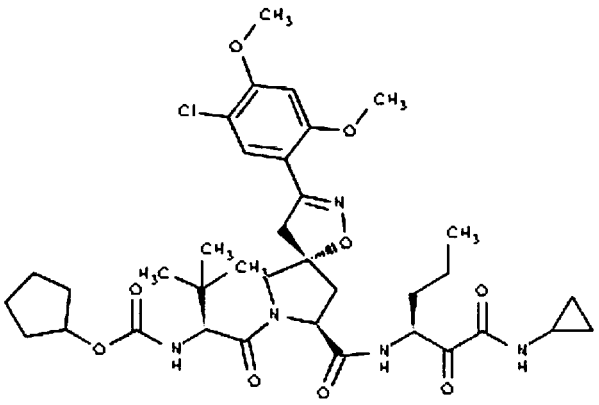
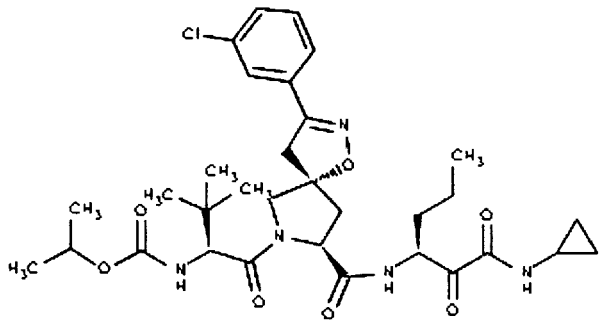
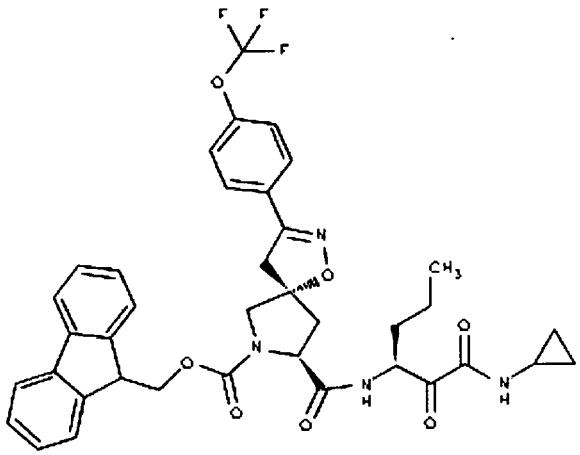
	177
	178
	179

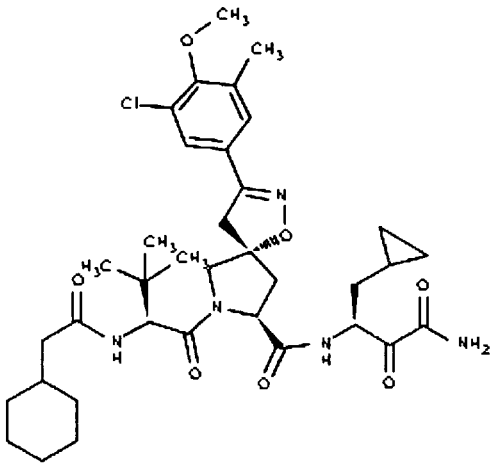
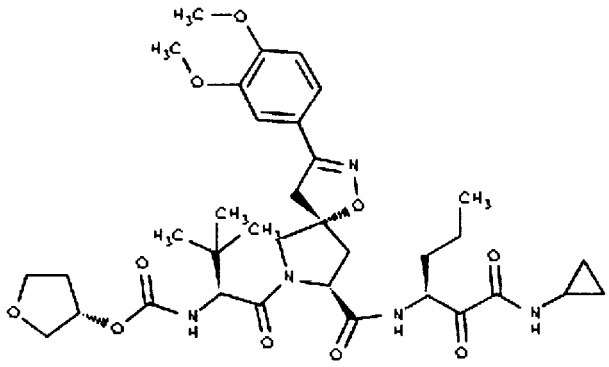
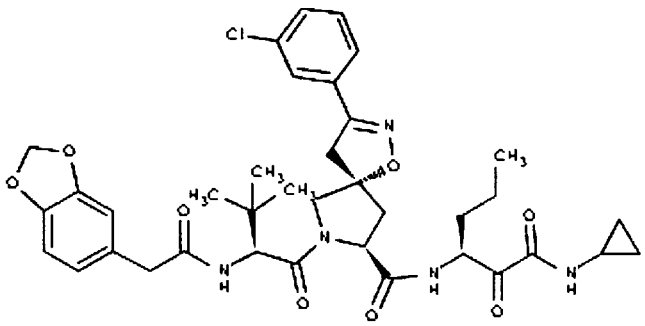
	180
	181
	182

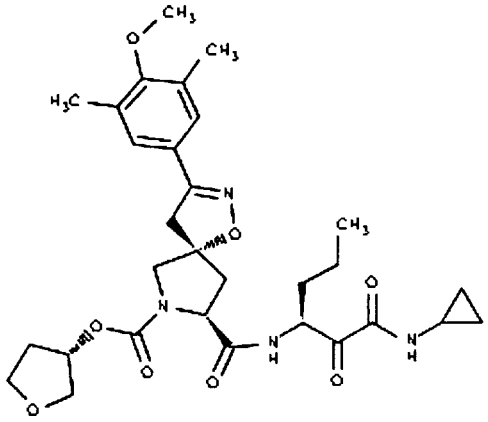
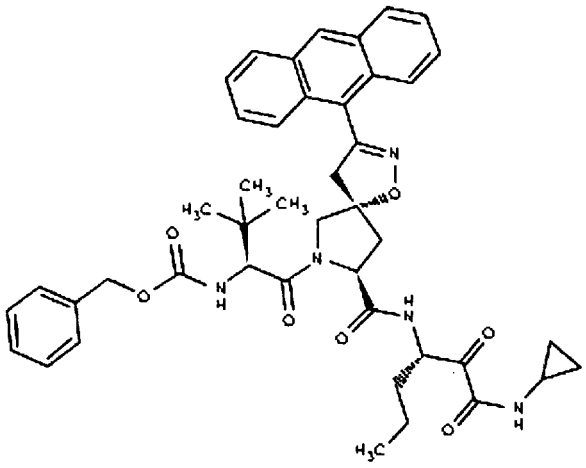
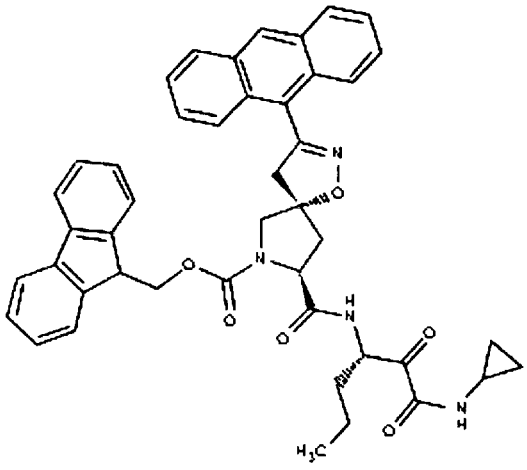
	183
	184
	185

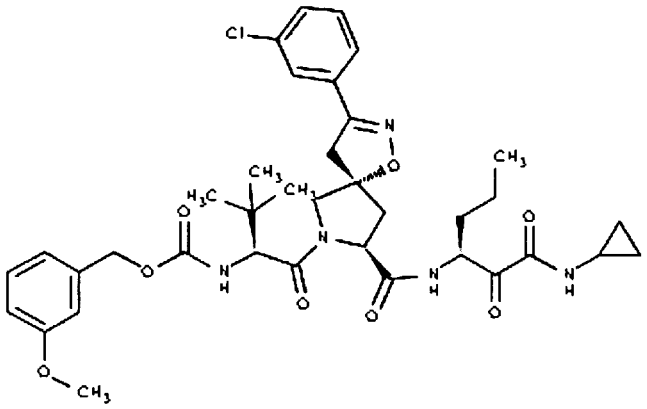
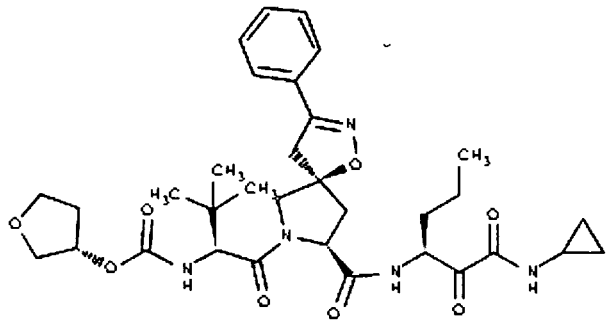
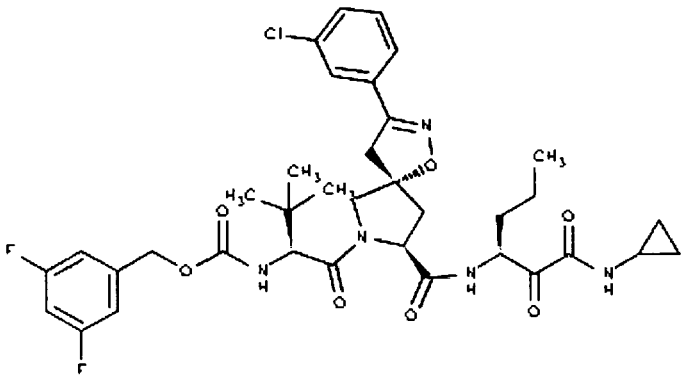
	186
	187
	188

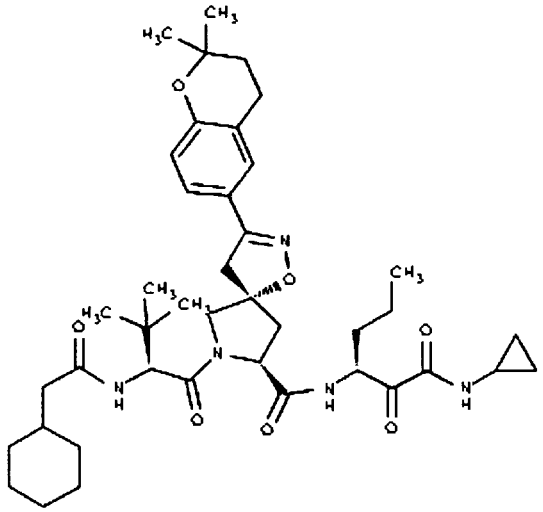
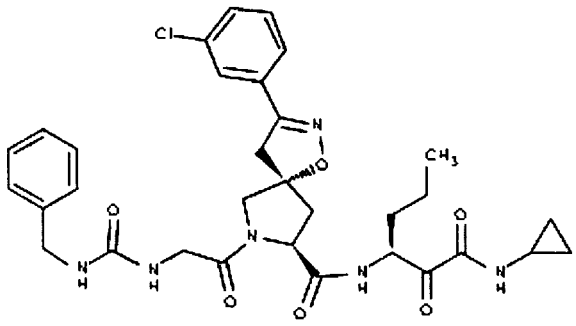
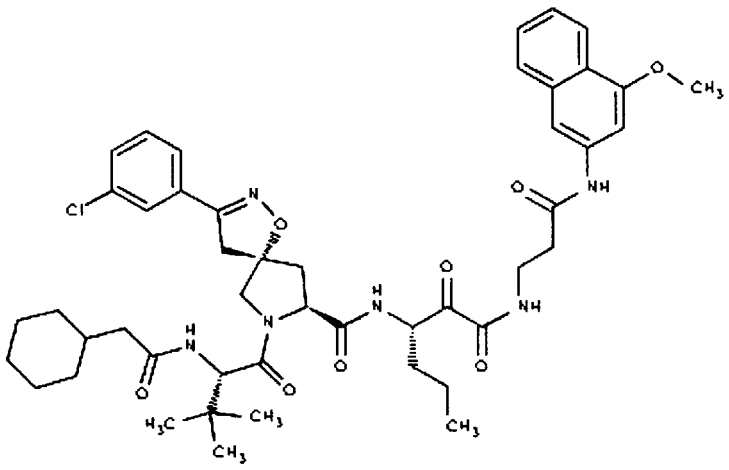
	189
	190
	191

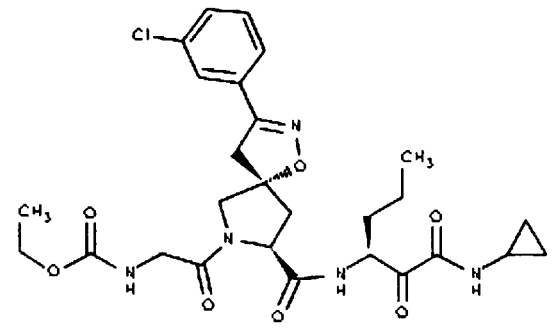
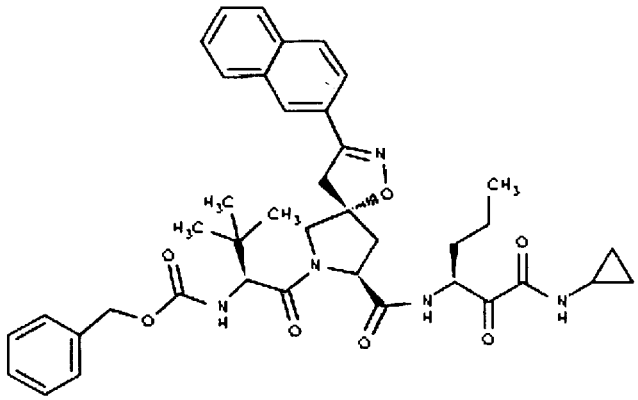
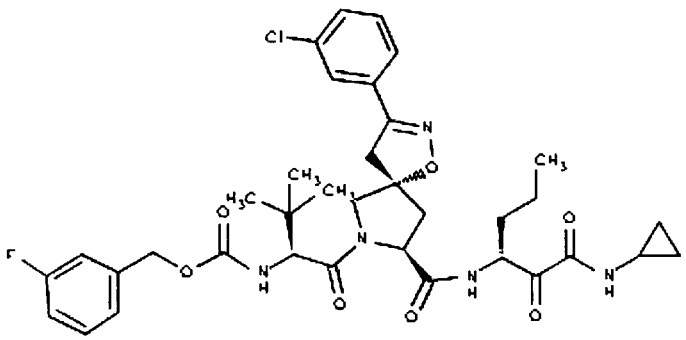
	192
	193
	194

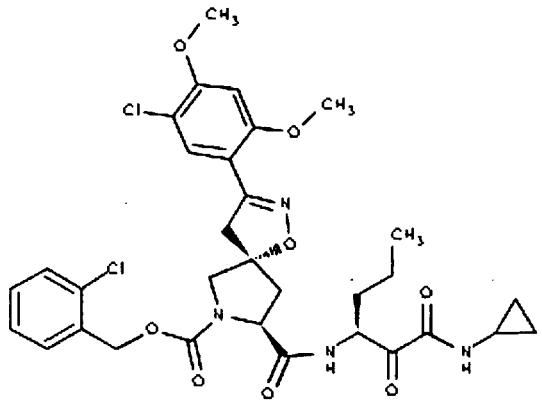
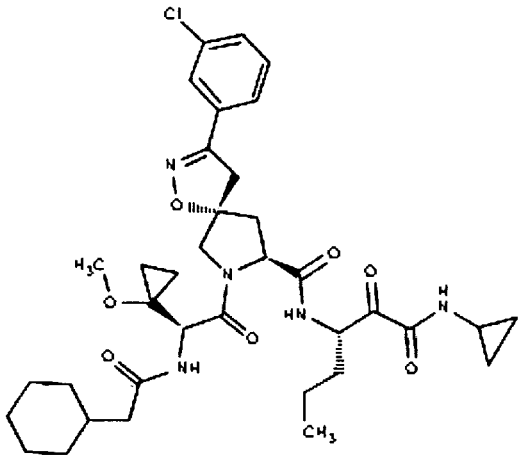
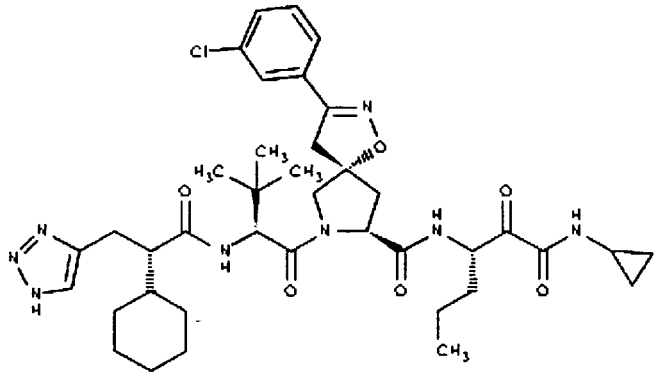
	195
	196
	197

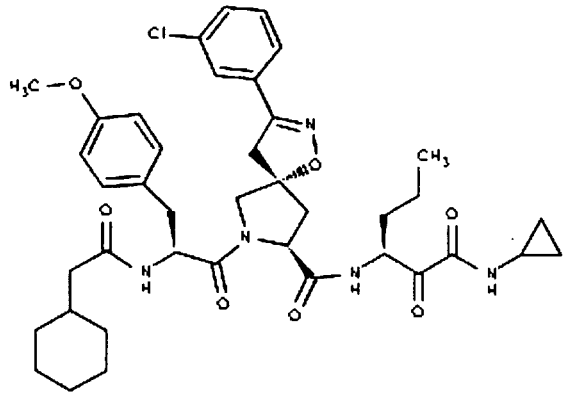
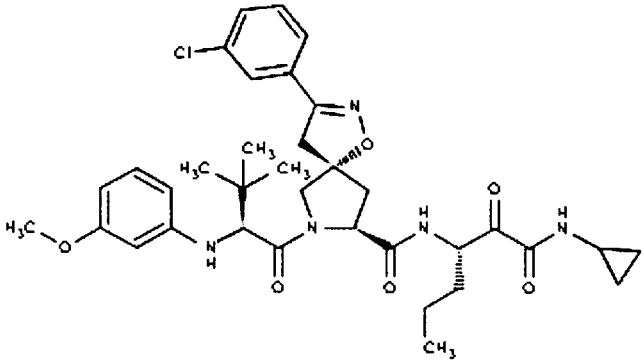
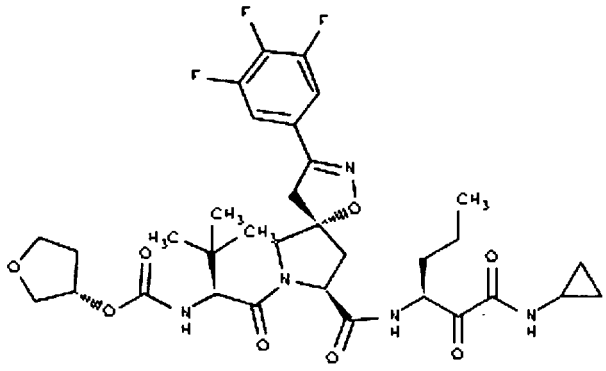
	198
	199
	200

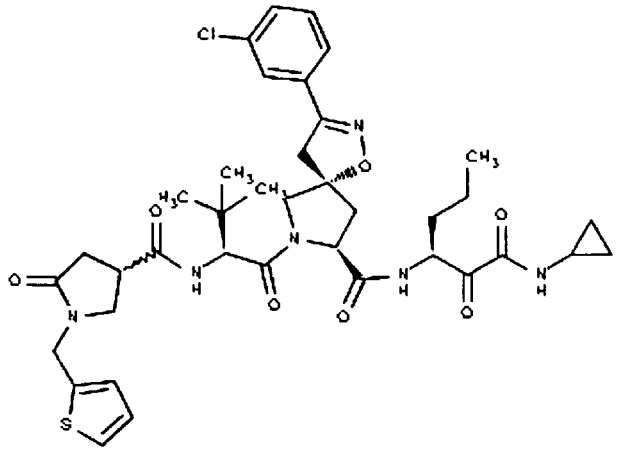
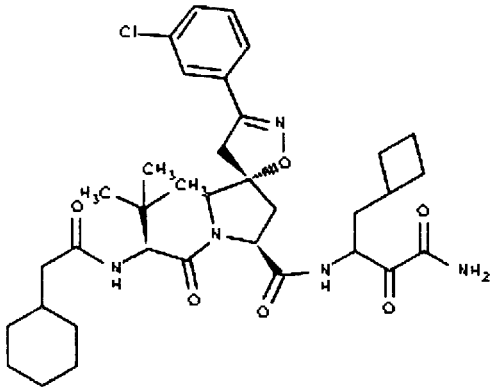
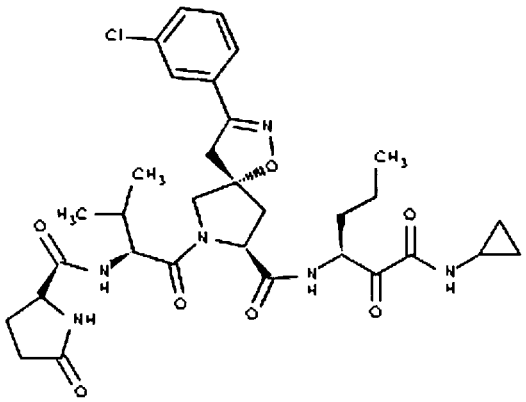
	201
	202
	203

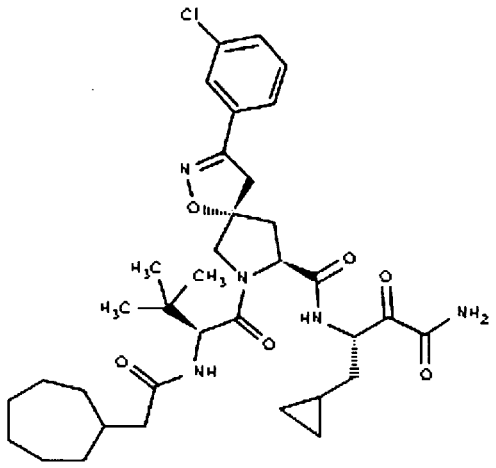
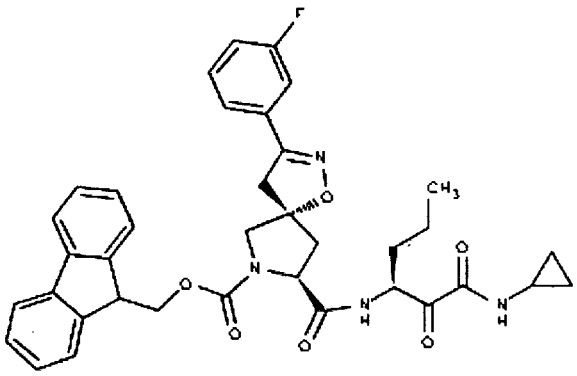
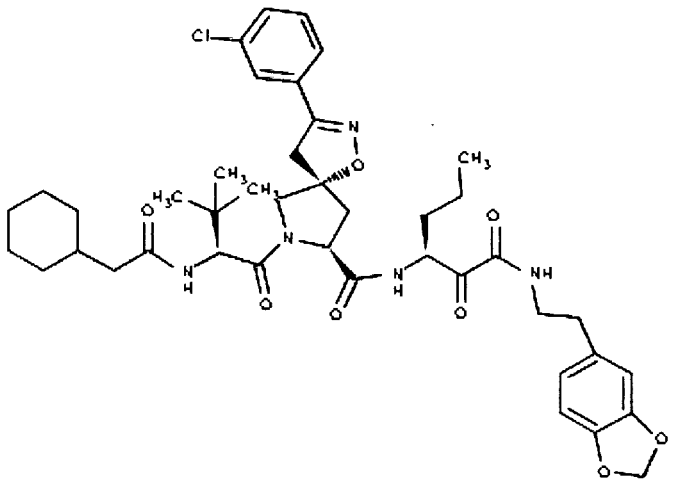
	204
	205
	206

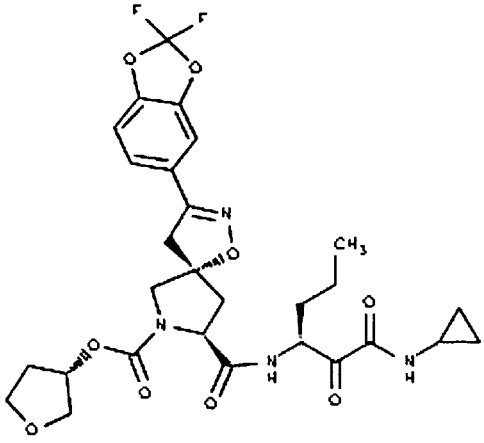
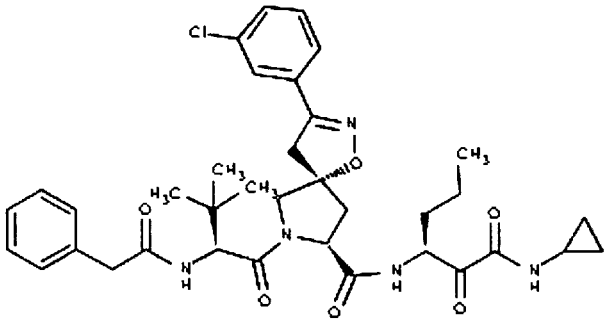
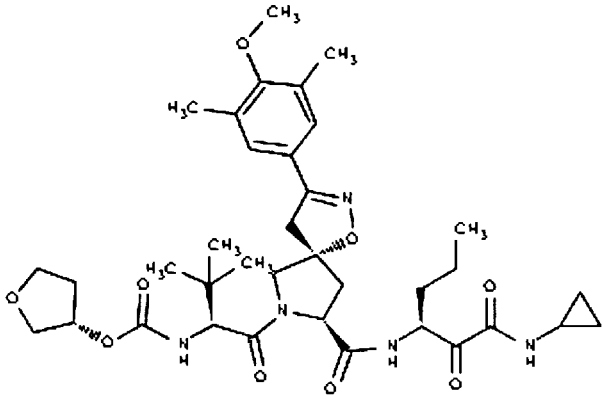
	207
	208
	209

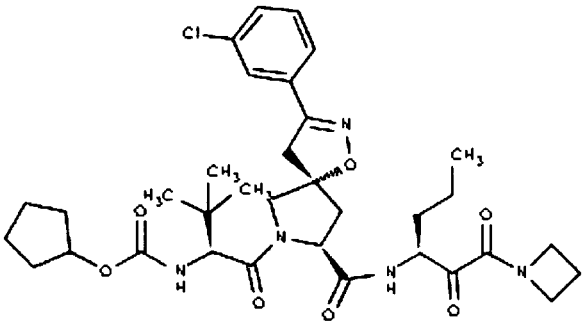
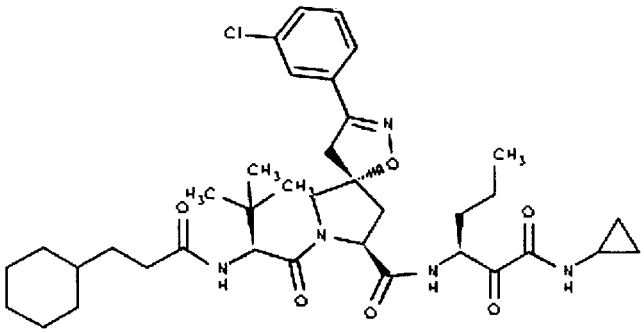
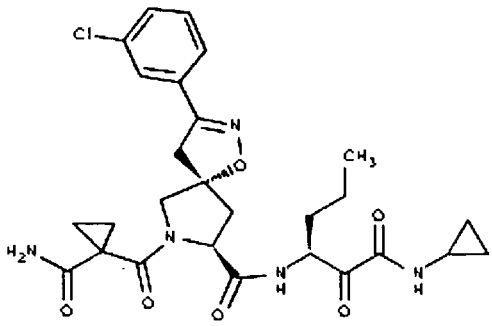
	210
	211
	212

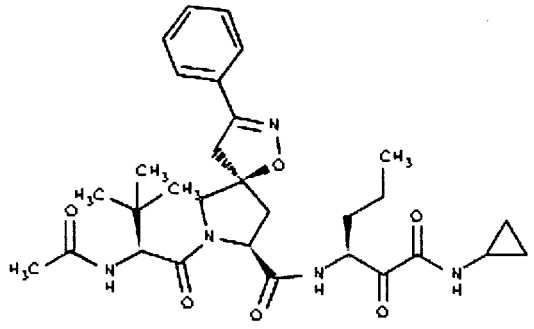
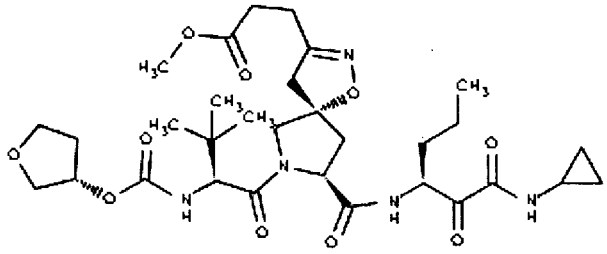
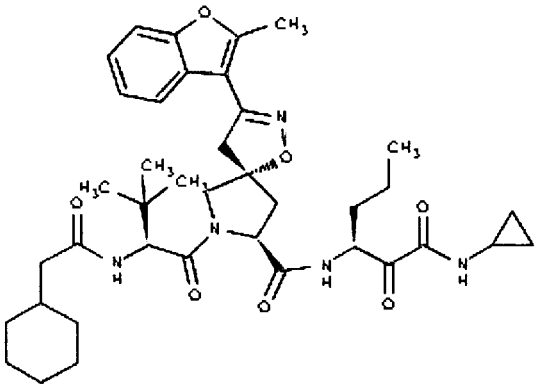
	213
	214
	215

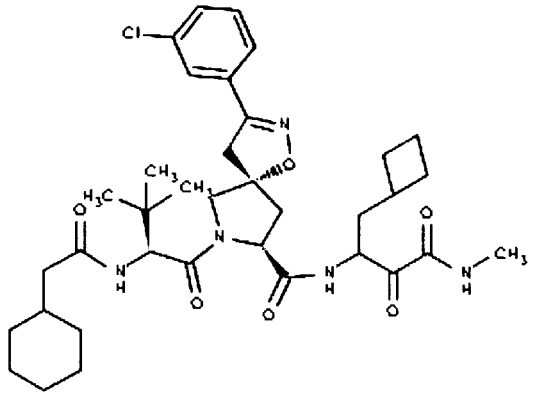
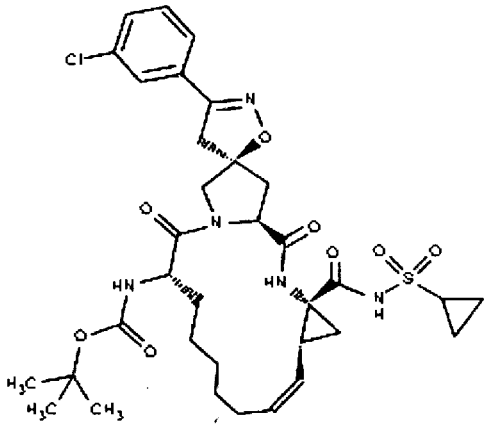
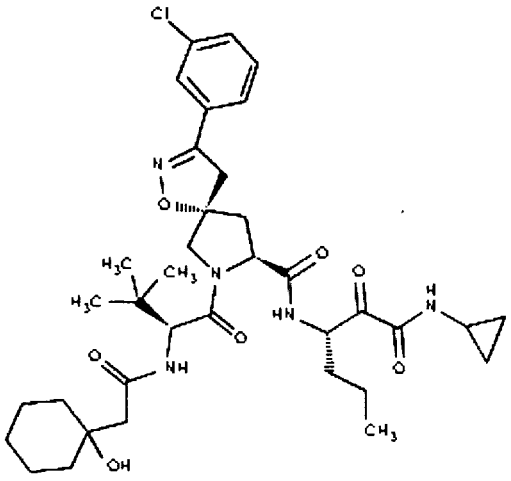
	216
	217
	218

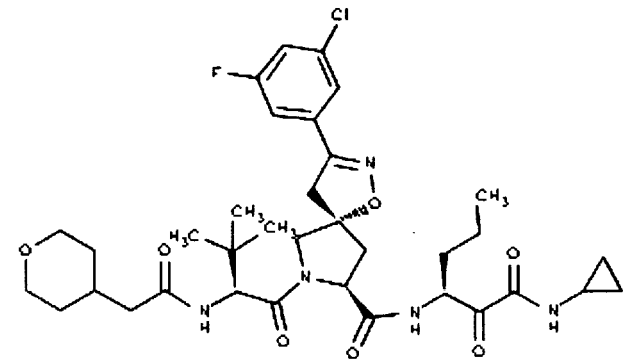
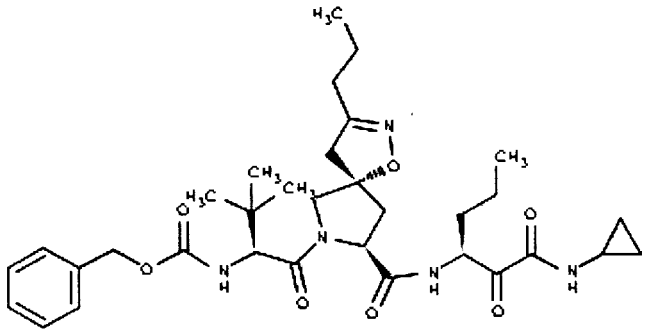
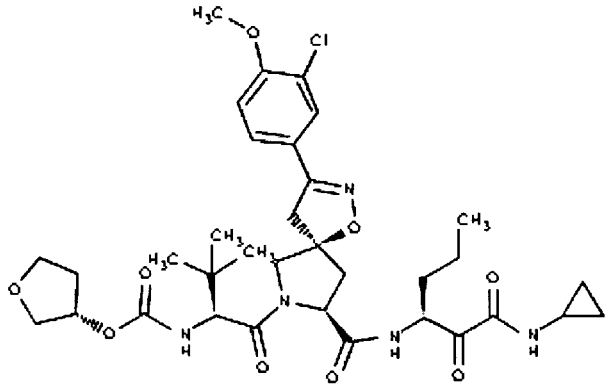
	219
	220
	221

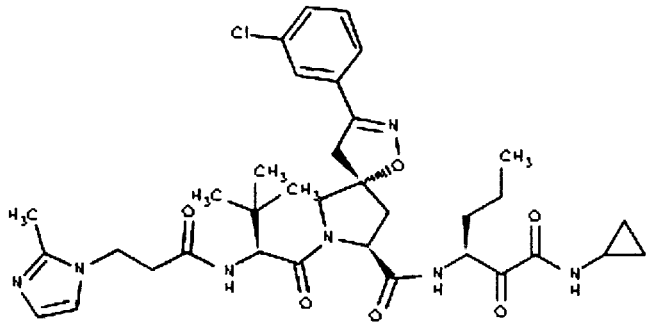
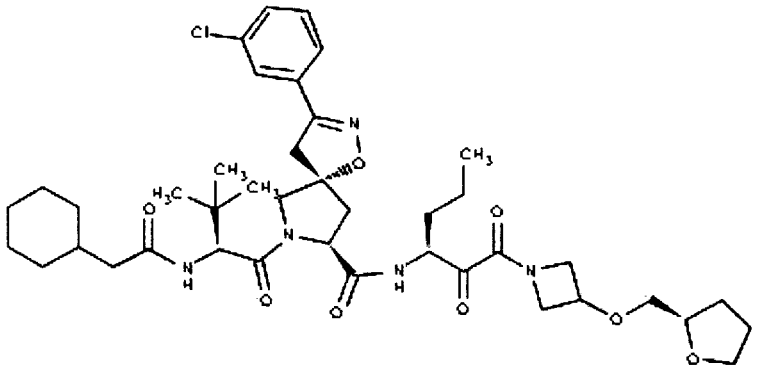
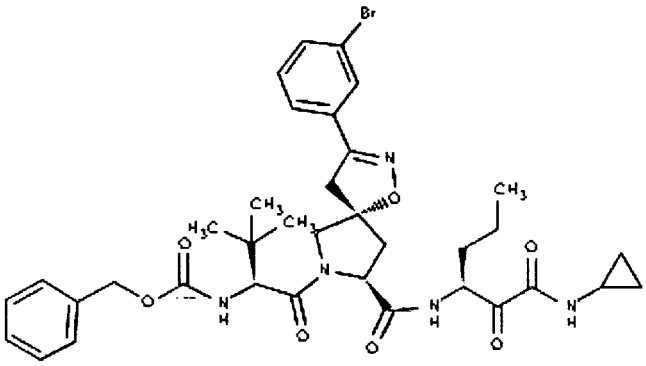
	222
	223
	224

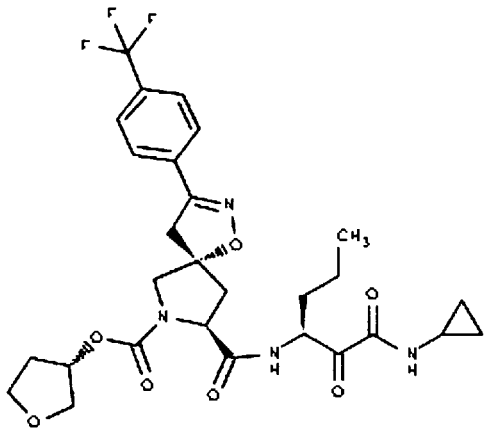
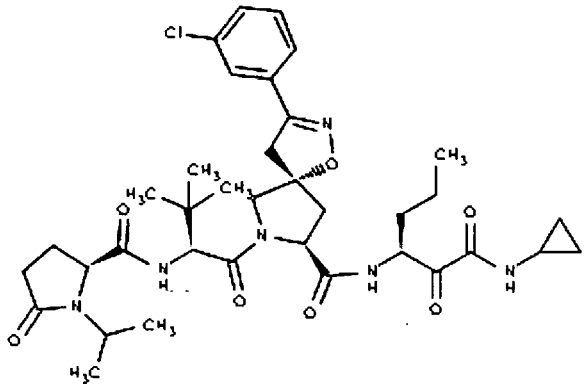
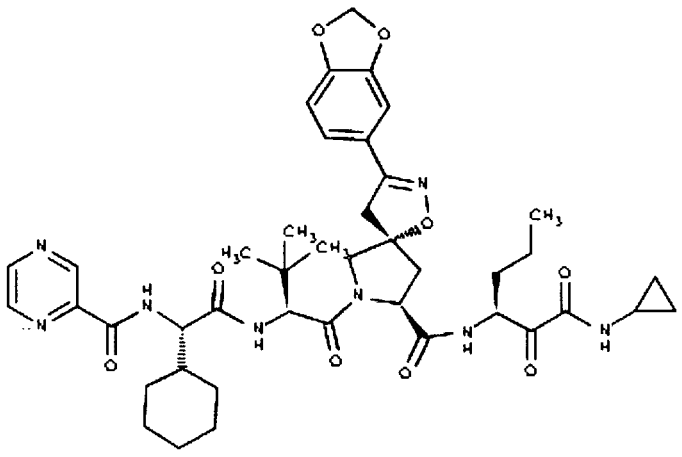
	225
	226
	227

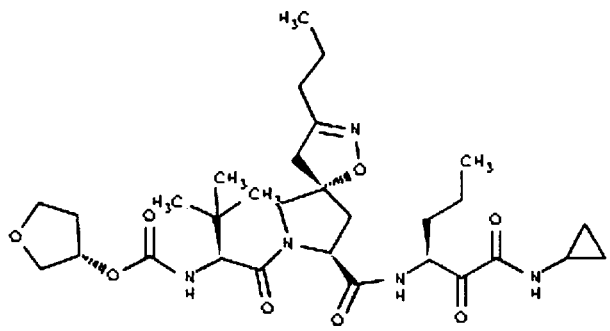
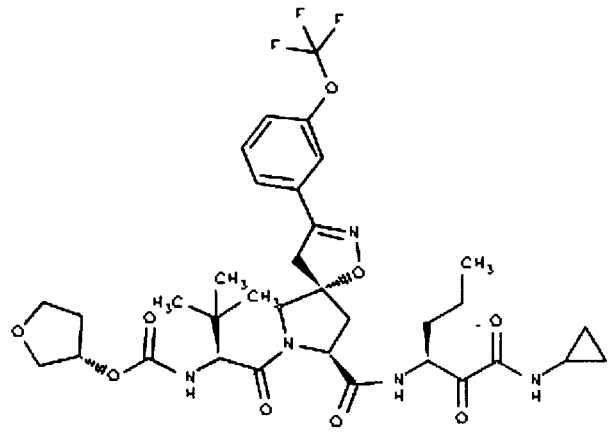
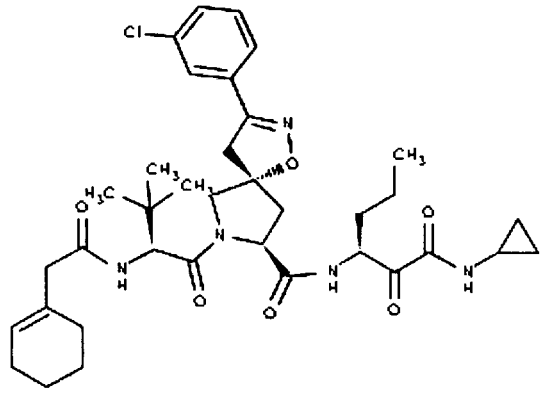
	228
	229
	230

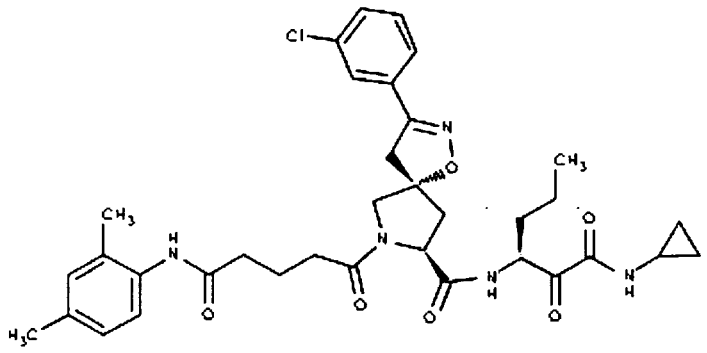
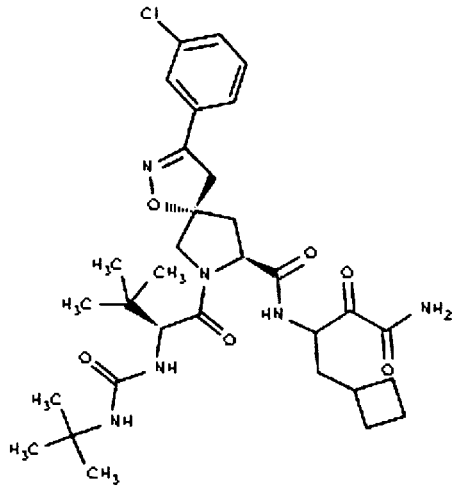
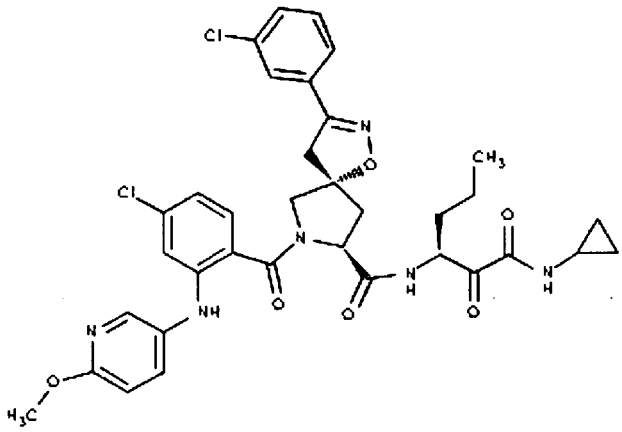
	231
	232
	233

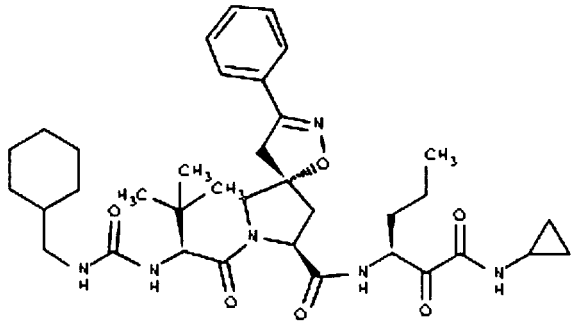
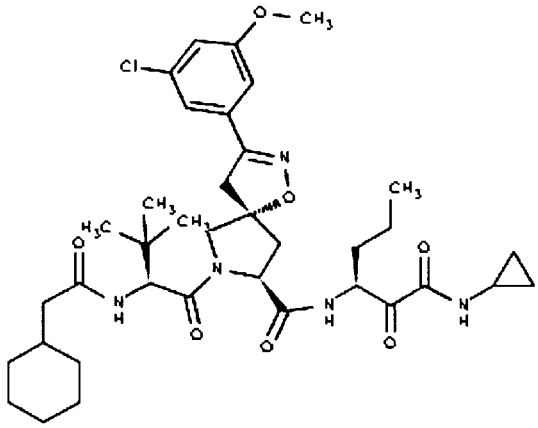
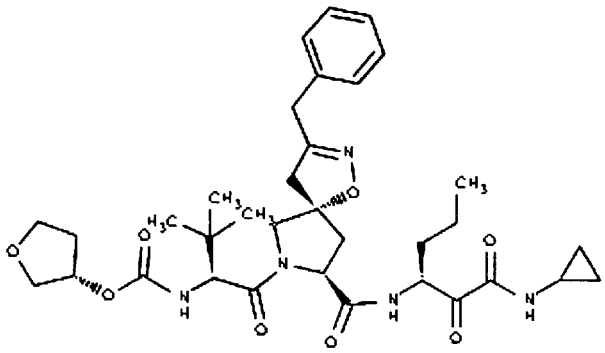
	234
	235
	236

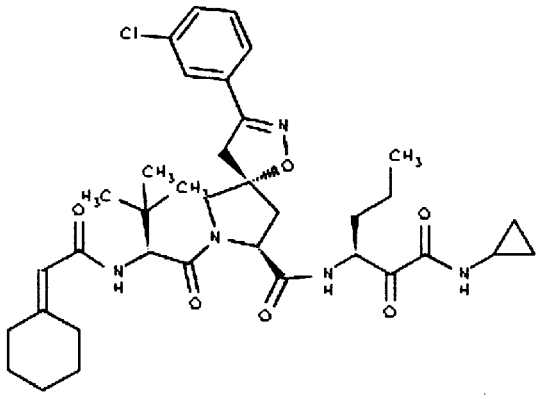
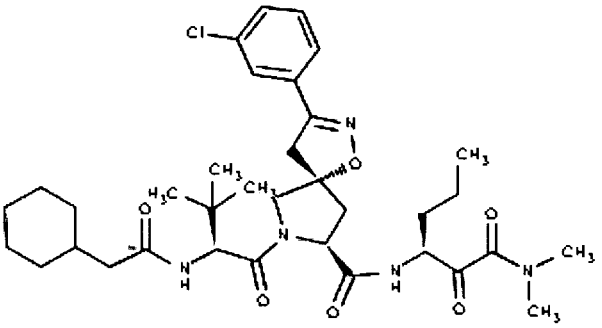
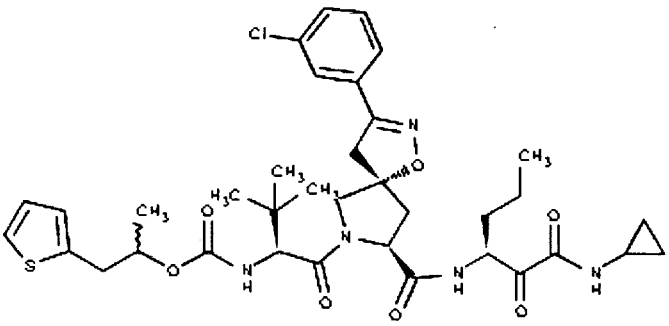
	237
	238
	239

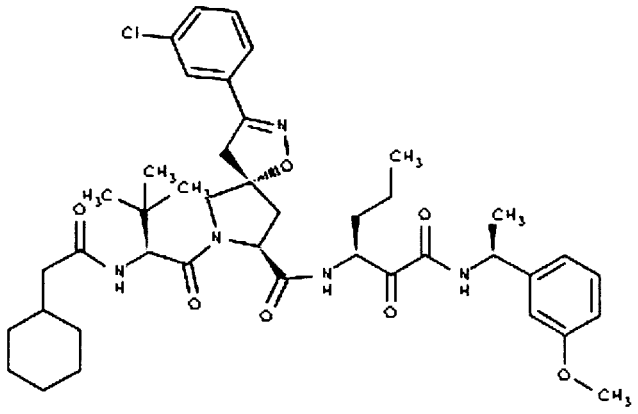
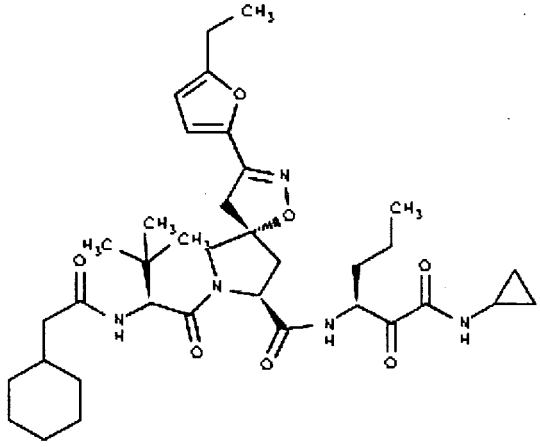
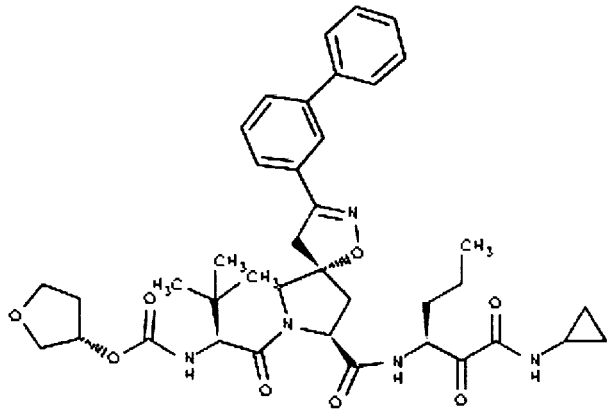
	240
	241
	242

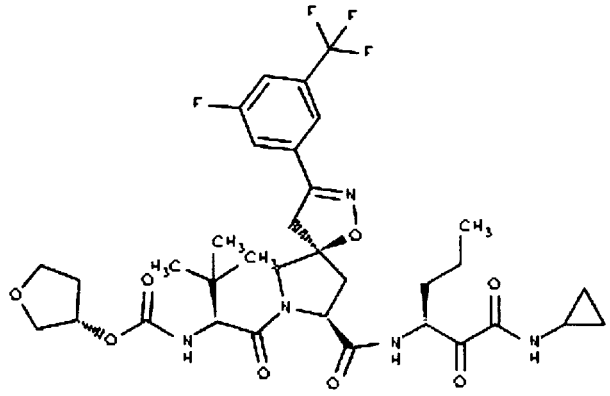
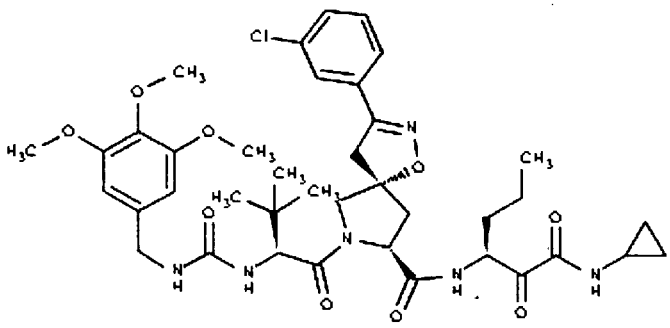
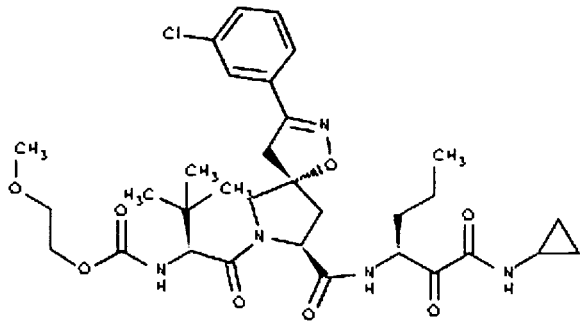
	243
	244
	245

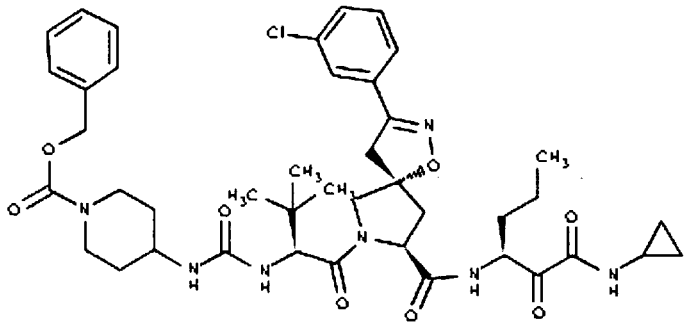
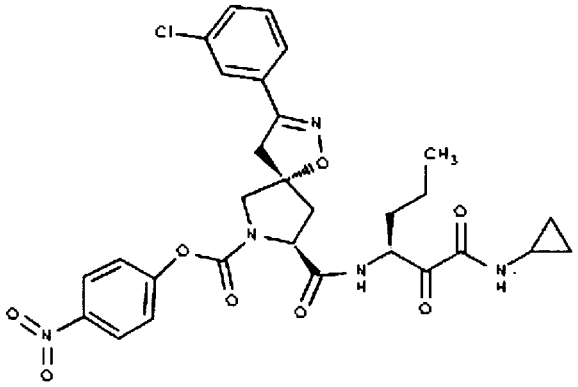
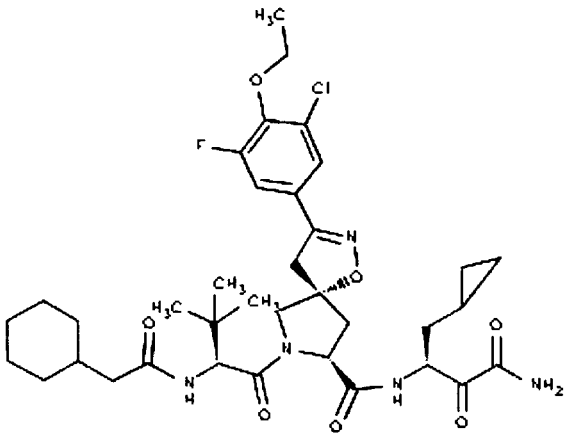
	246
	247
	248

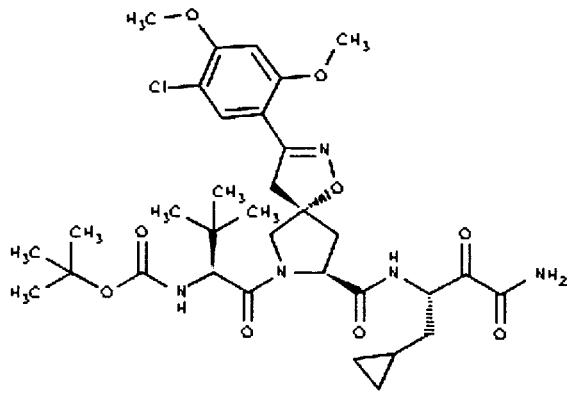
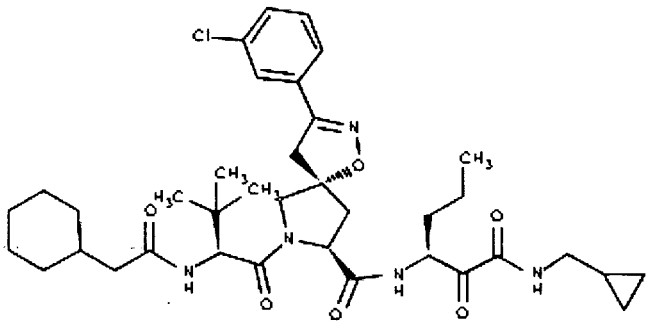
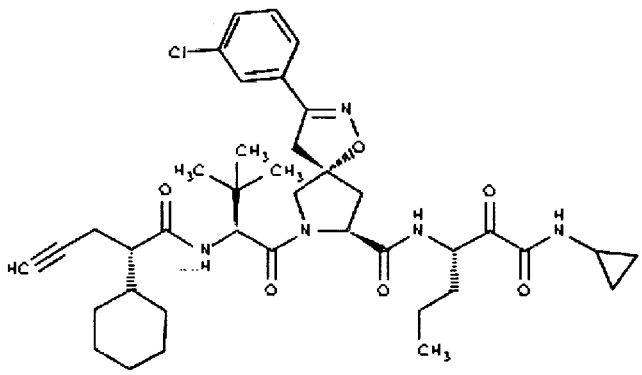
	249
	250
	251

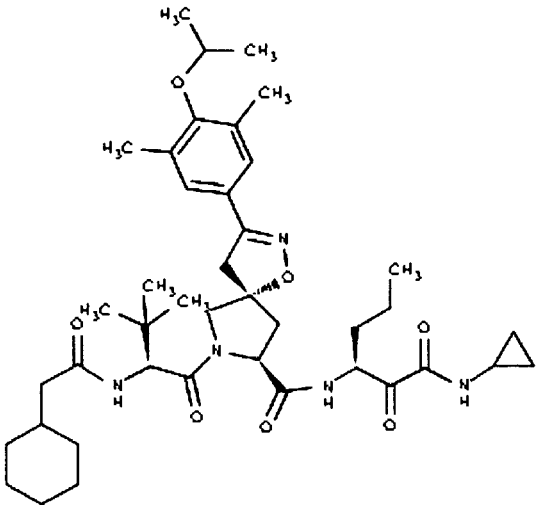
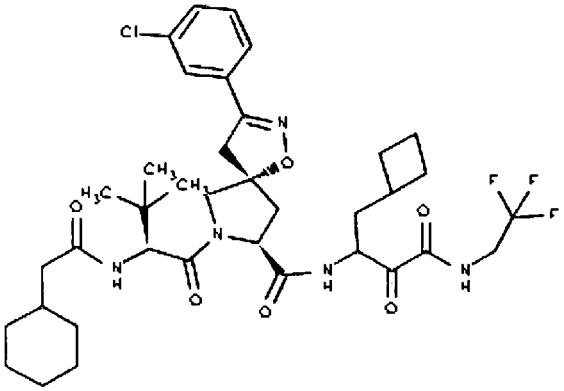
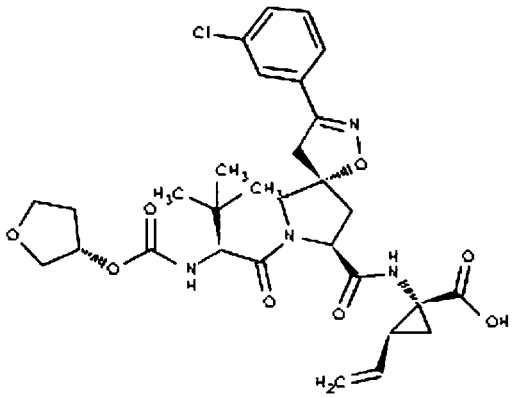
	252
	253
	254

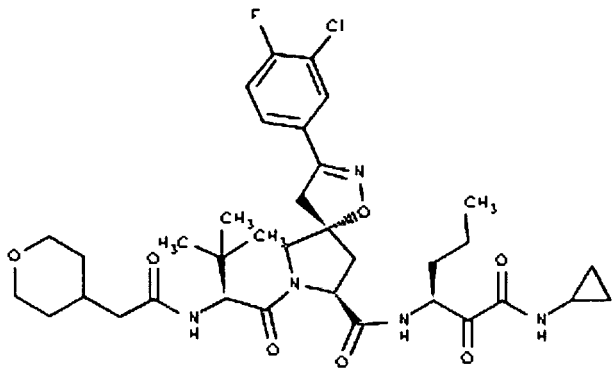
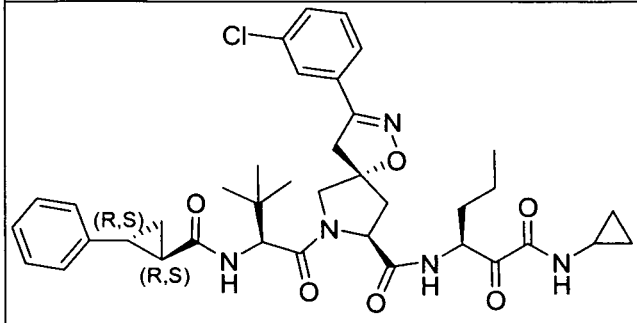
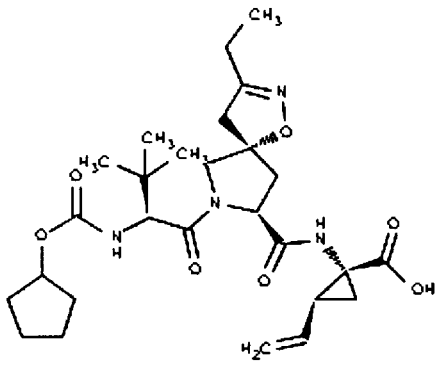
	255
	256
	257

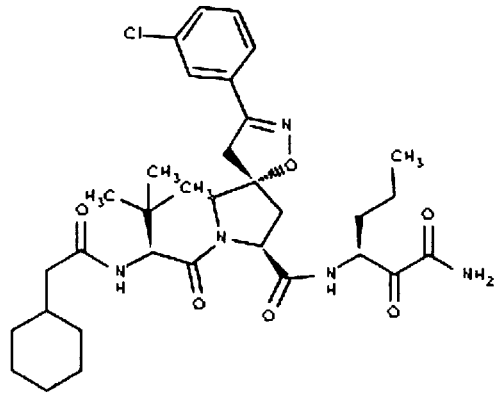
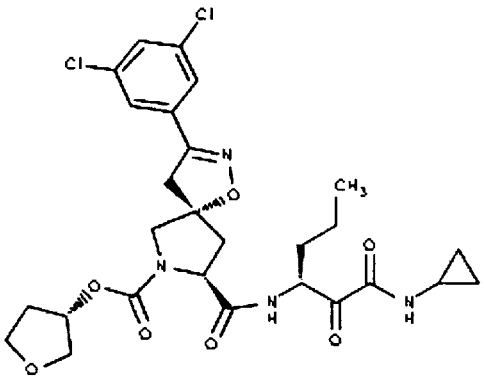
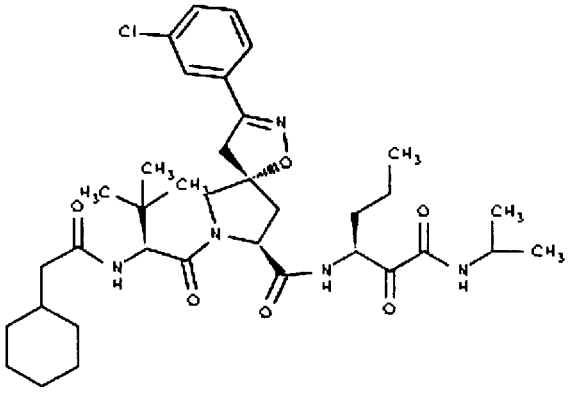
	258
	259
	260

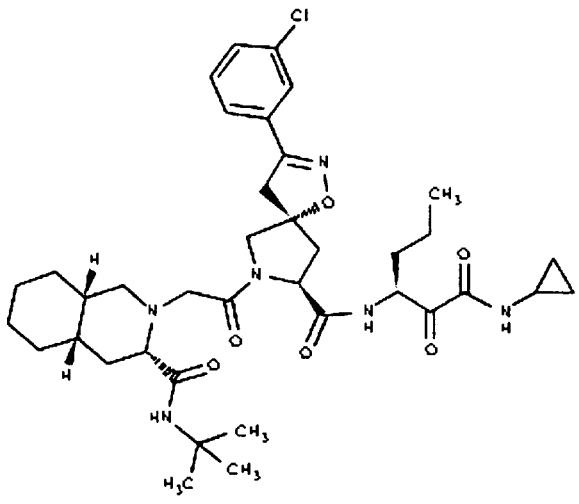
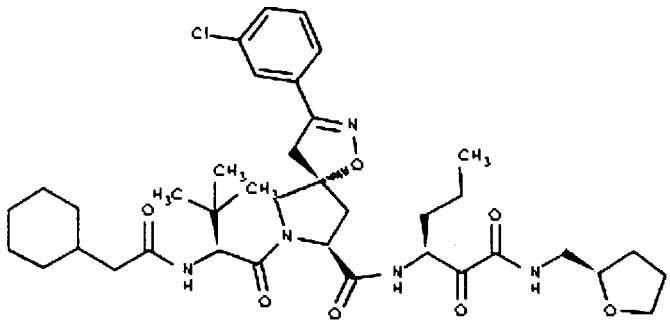
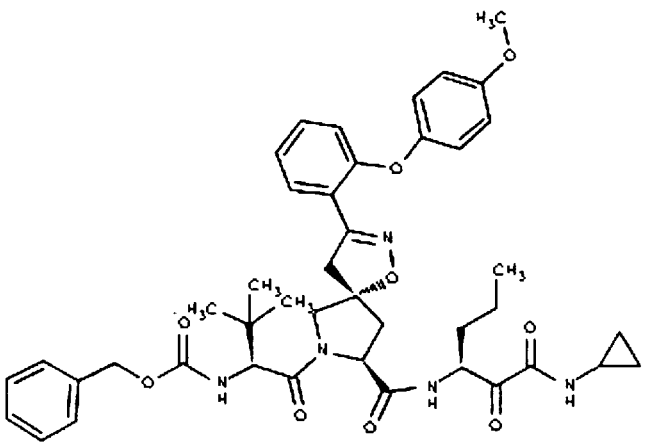
	261
	262
	263

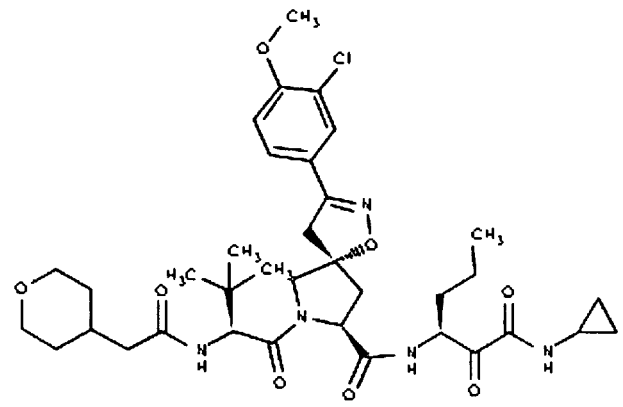
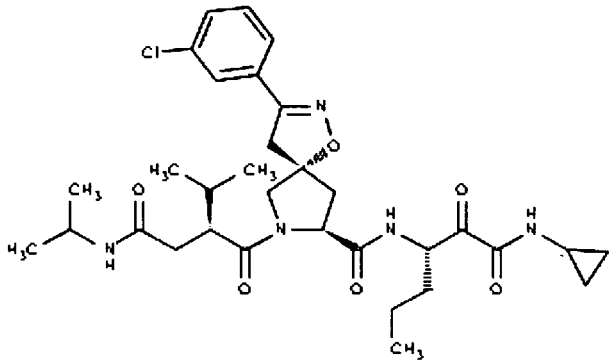
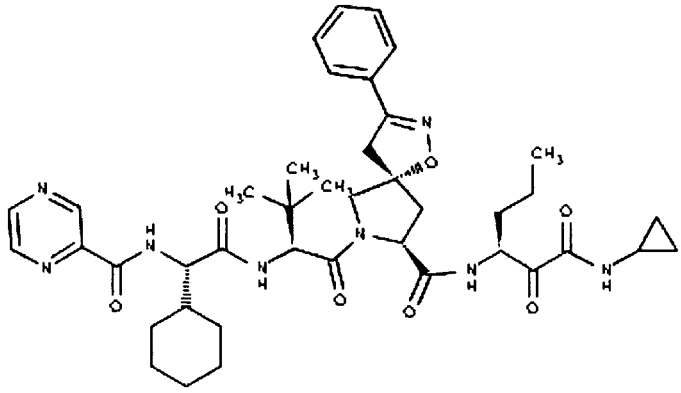
	264
	265
	266

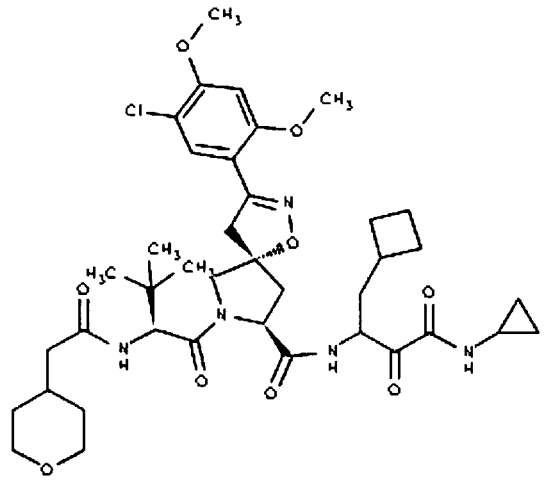
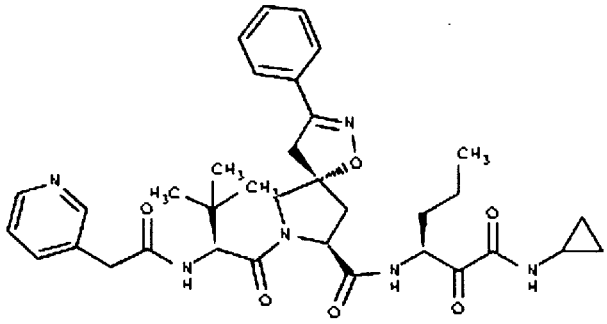
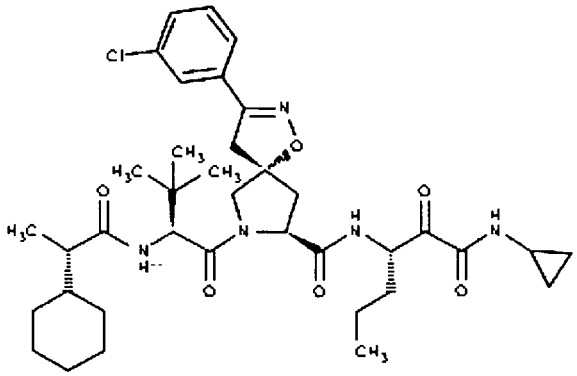
	267
	268
	269

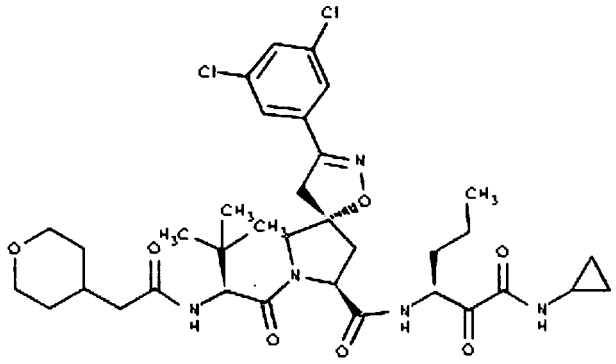
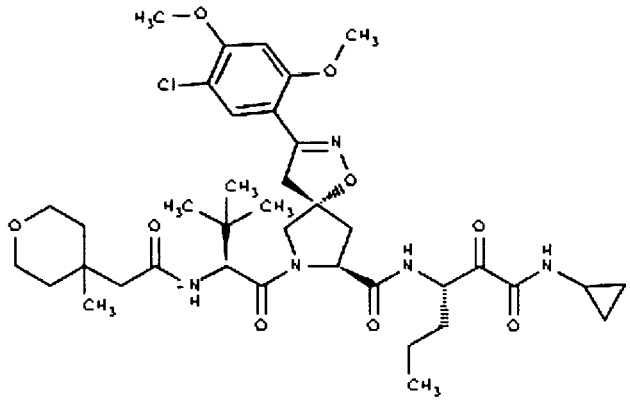
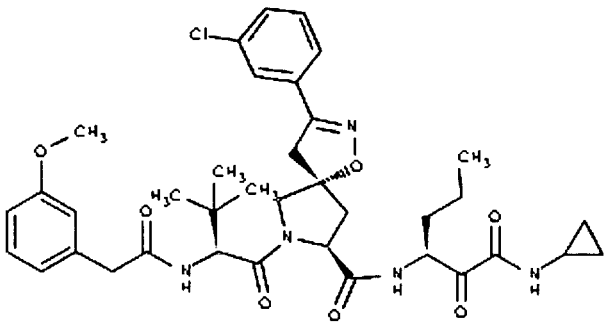
	270
	271
	272

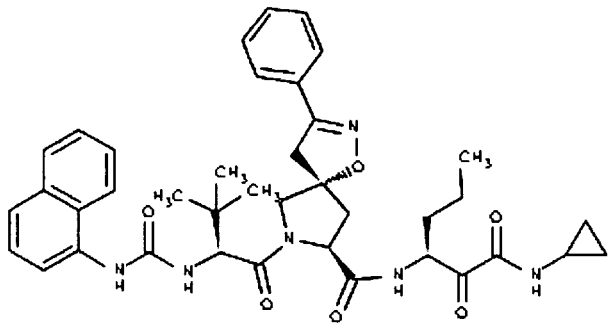
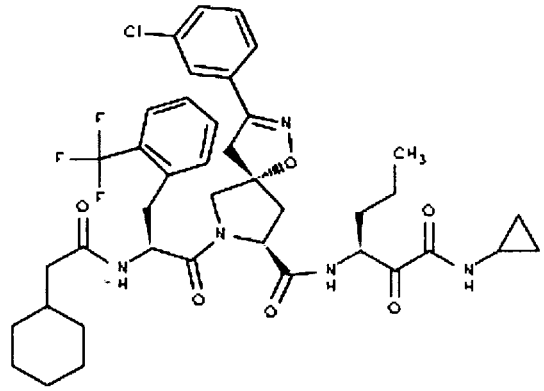
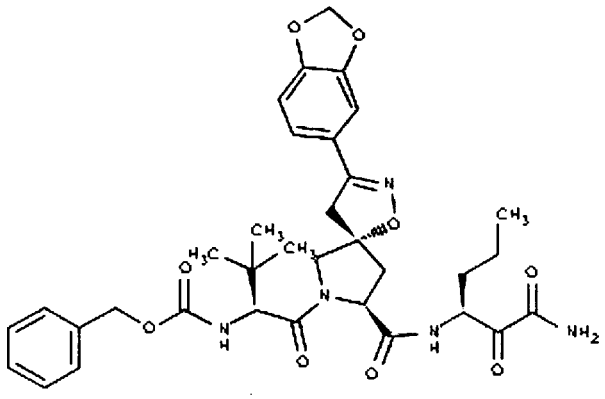
	273
	274
	275

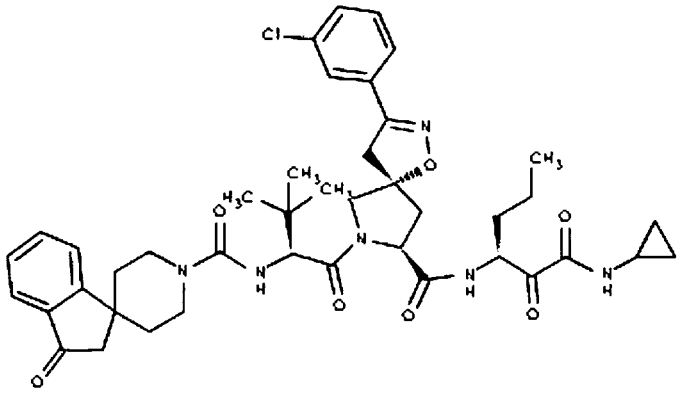
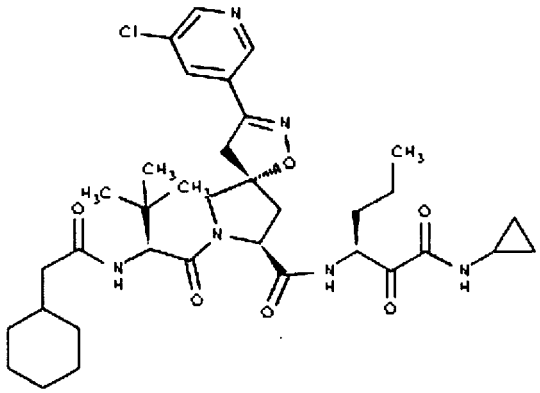
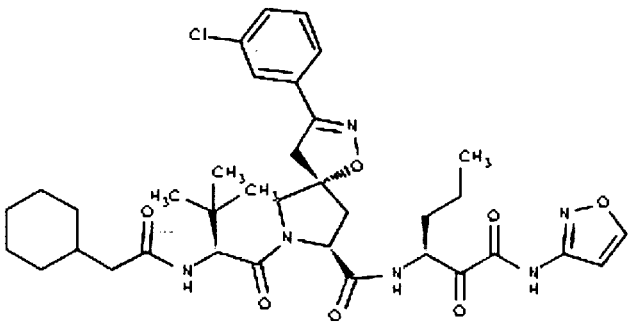
	276
	277
	278

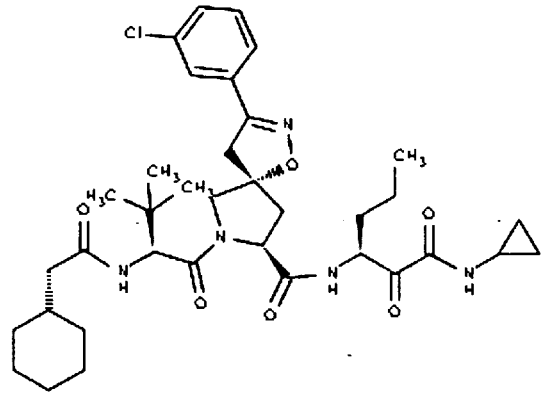
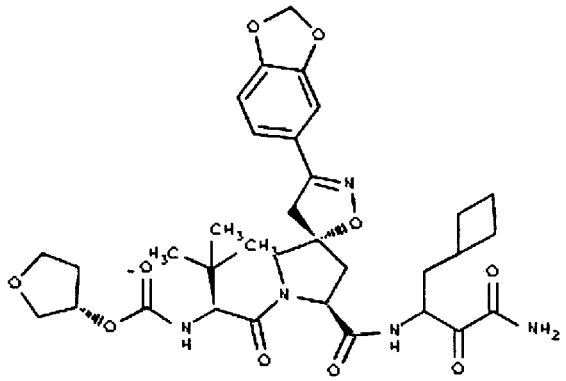
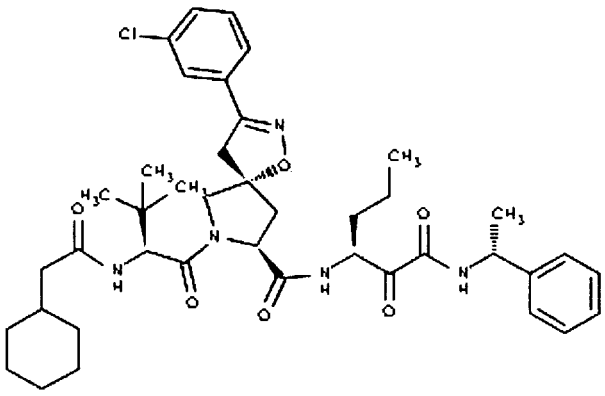
	279
	280
	281

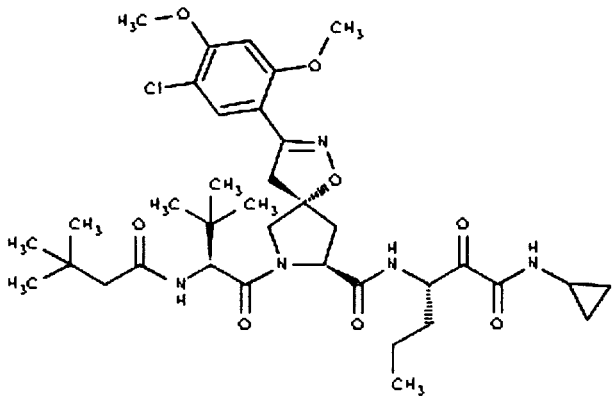
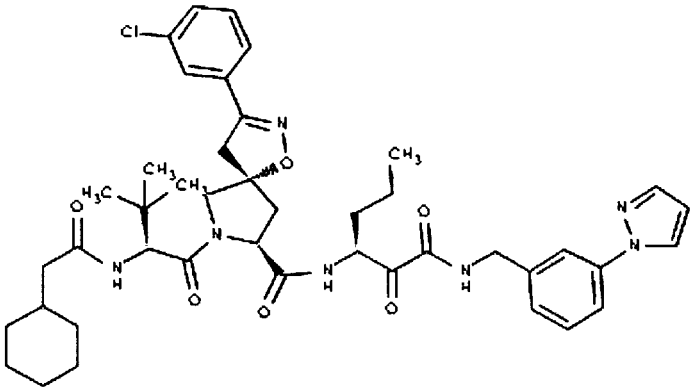
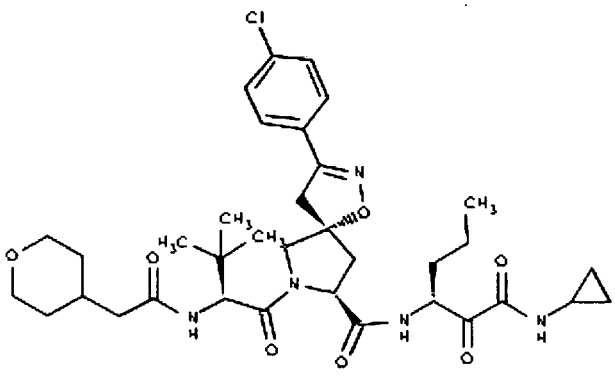
	282
	283
	284

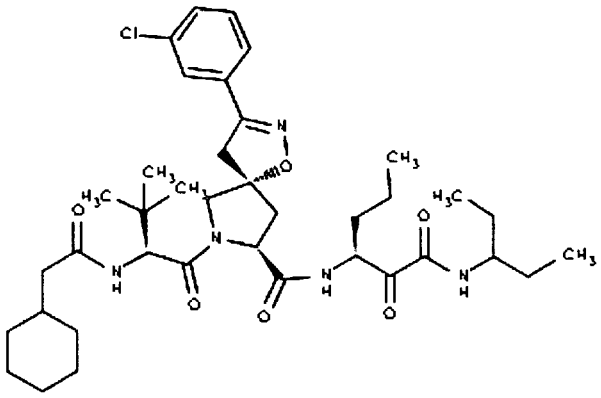
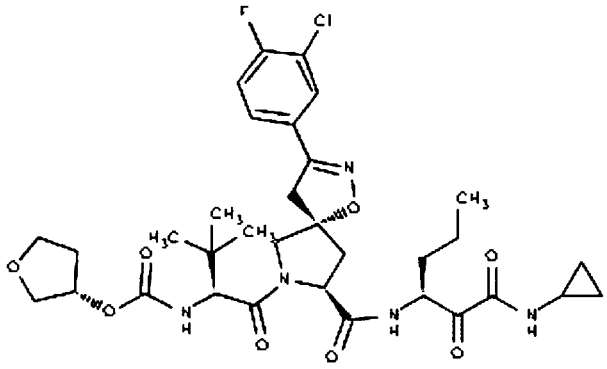
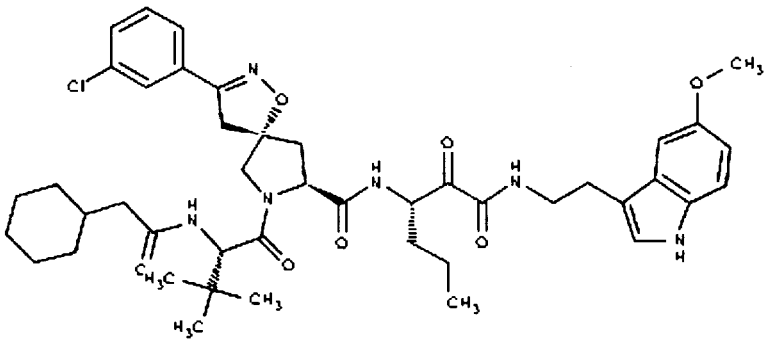
	285
	286
	287

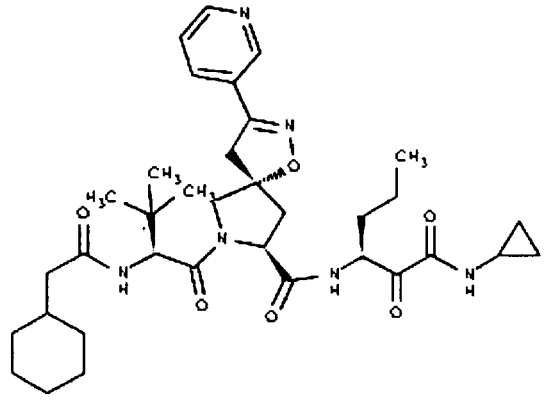
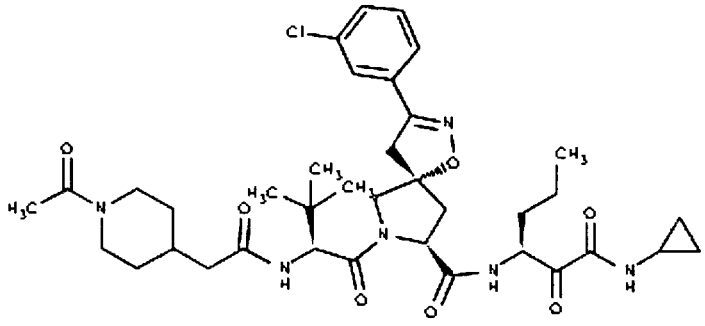
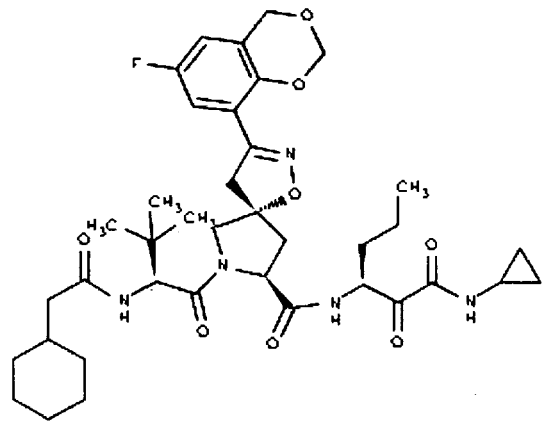
	288
	289
	290

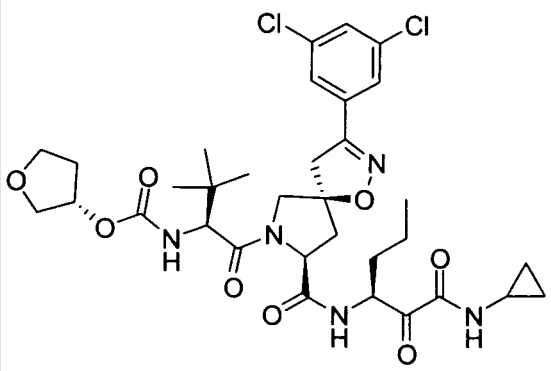
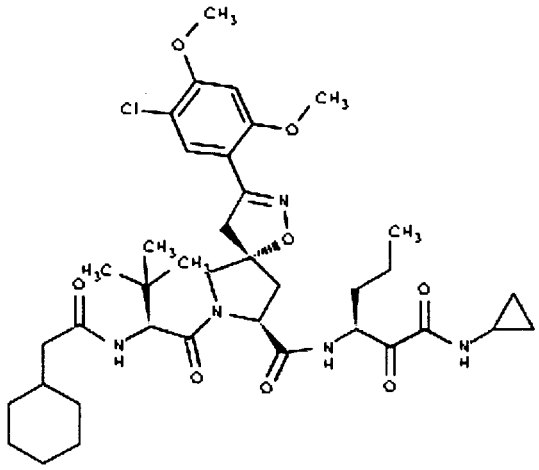
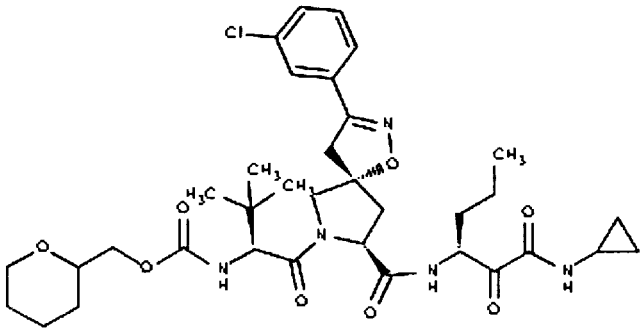
	291
	292
	293

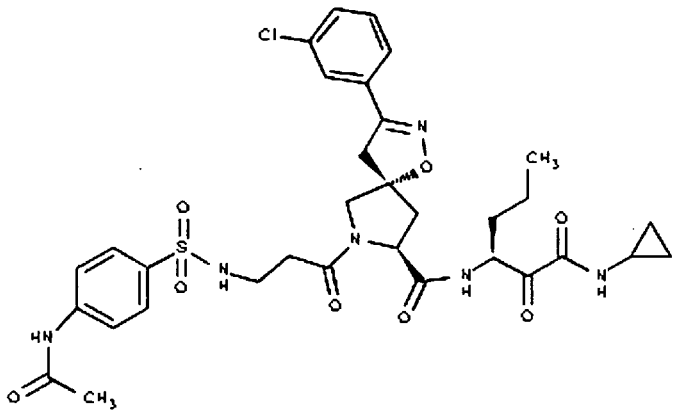
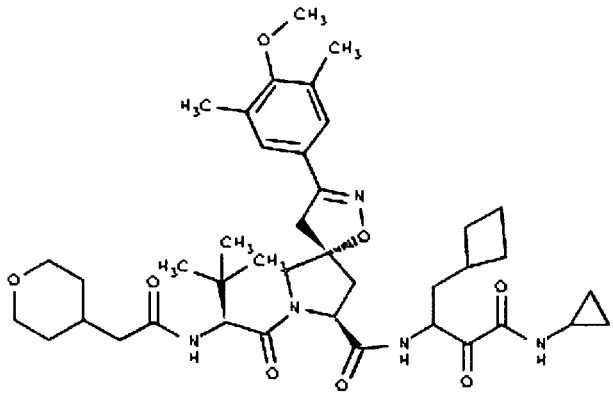
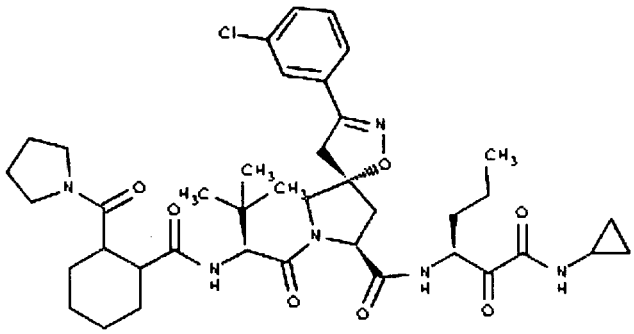
	294
	295
	296

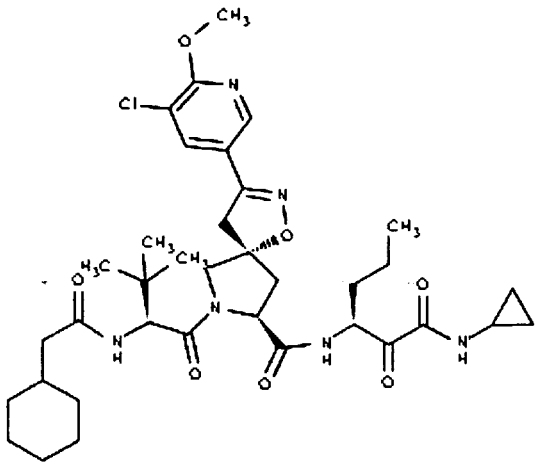
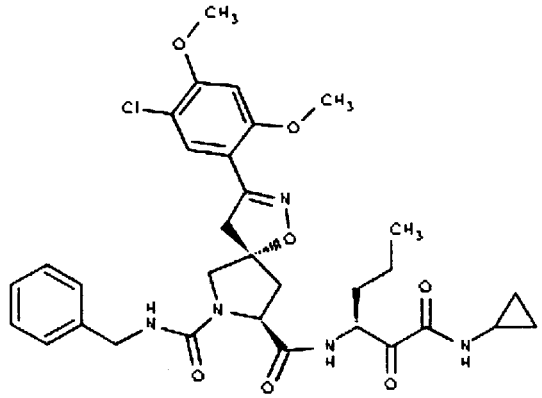
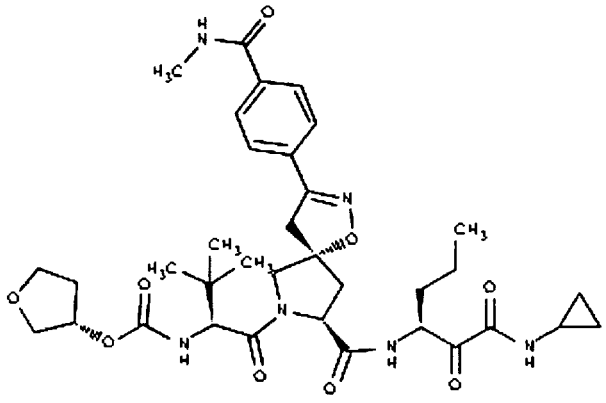
	297
	298
	299

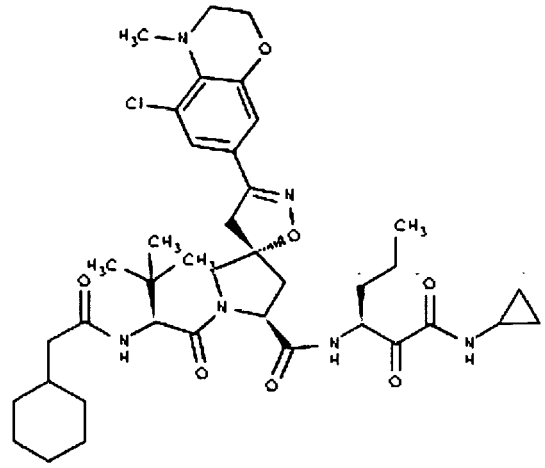
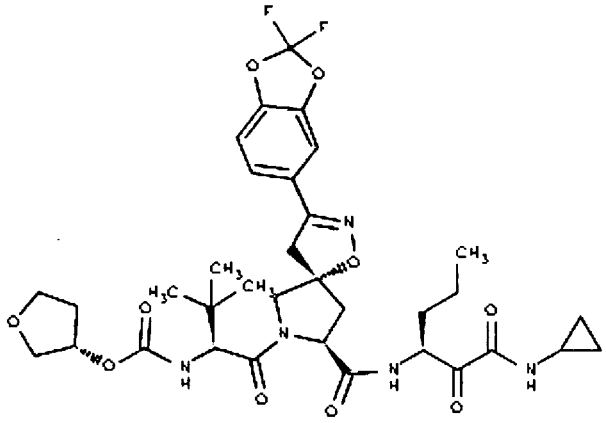
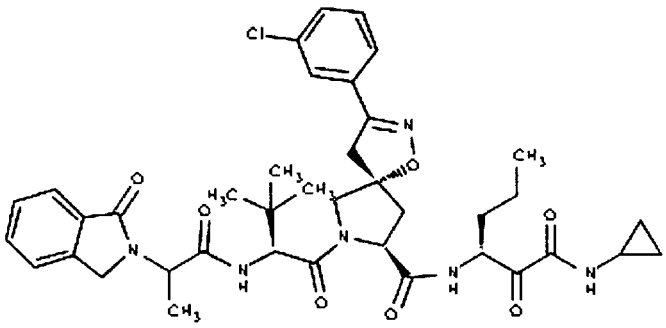
	300
	301
	302

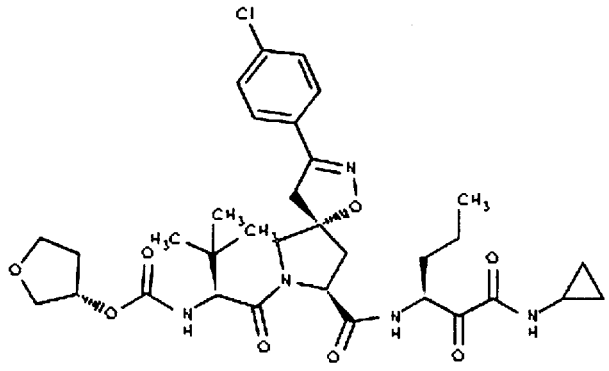
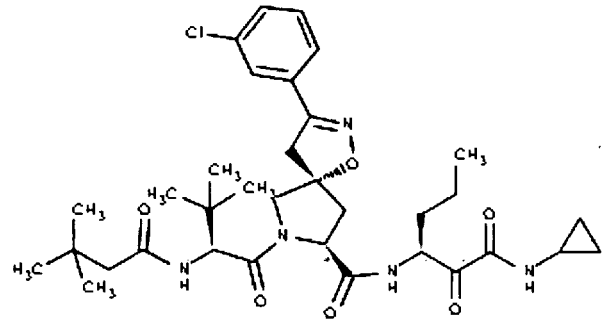
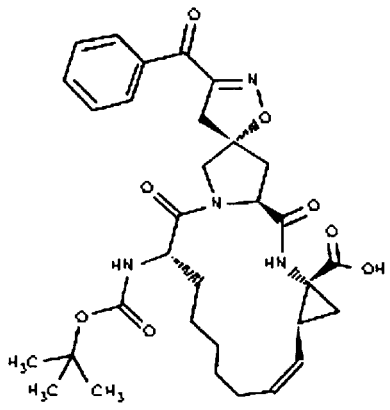
	303
	304
	305

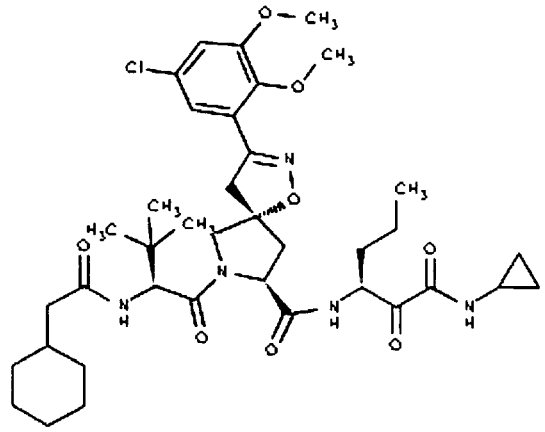
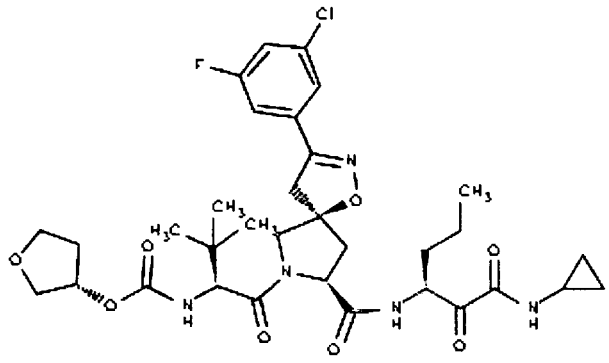
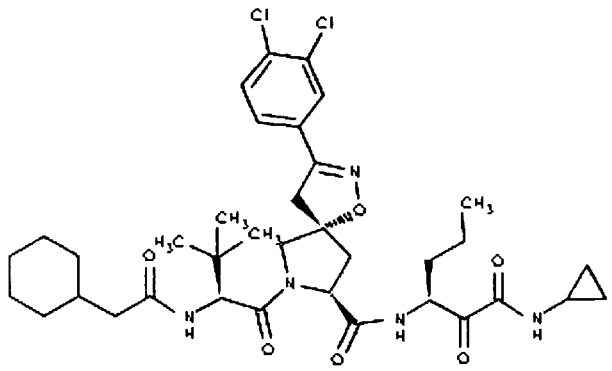
	306
	307
	308

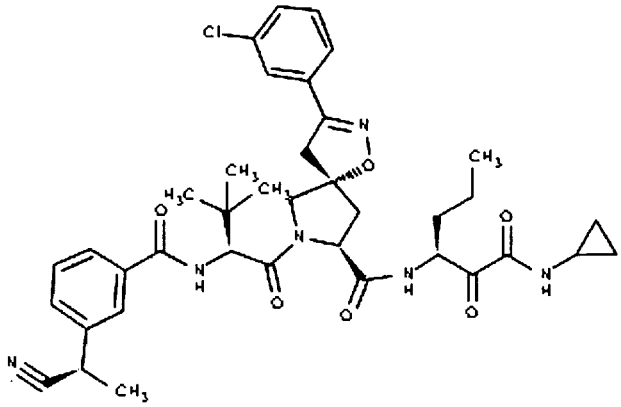
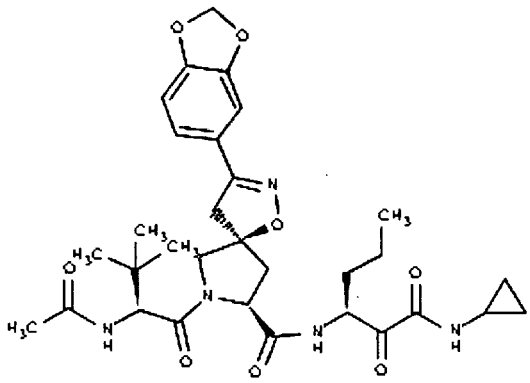
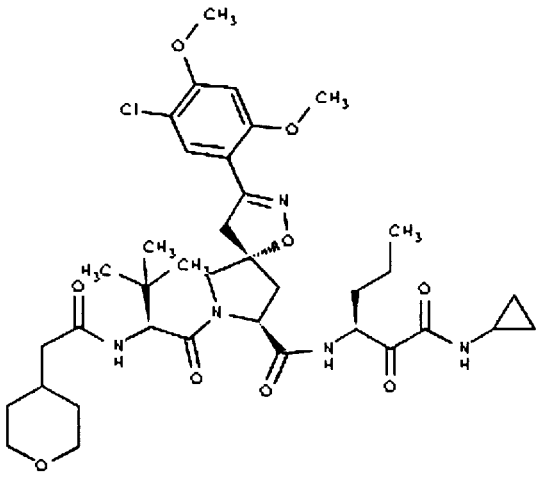
	309
	310
	311

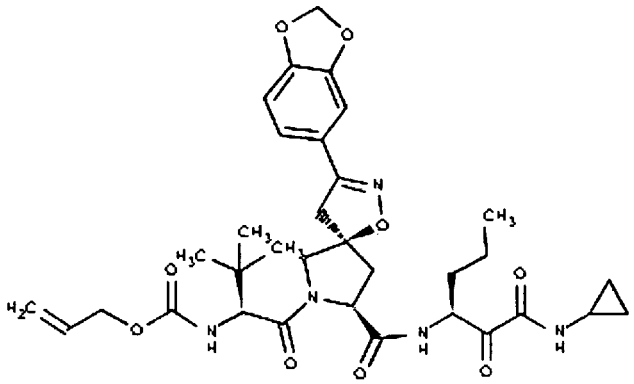
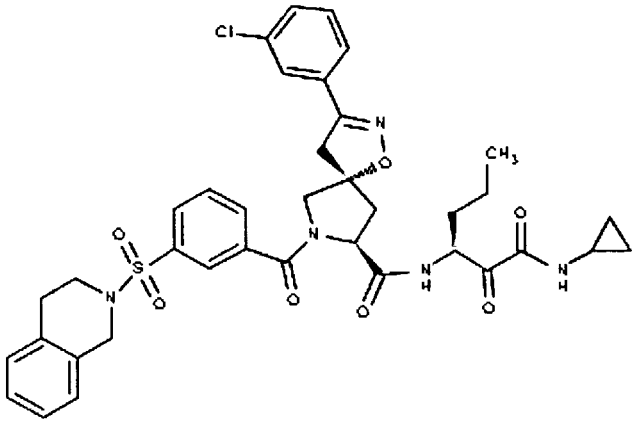
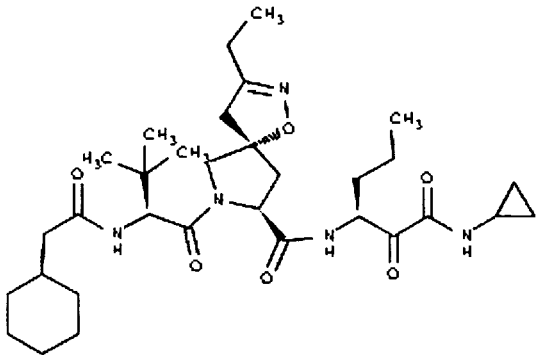
	312
	313
	314

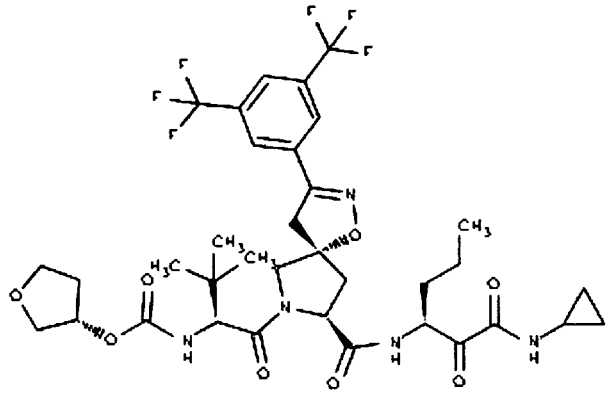
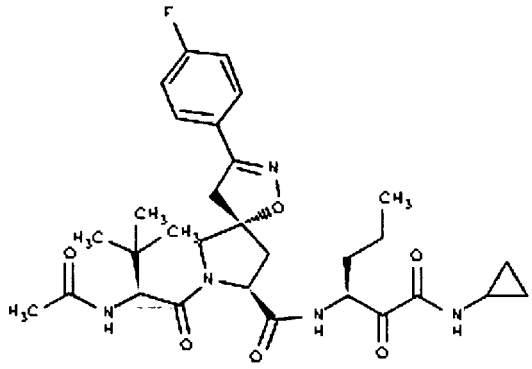
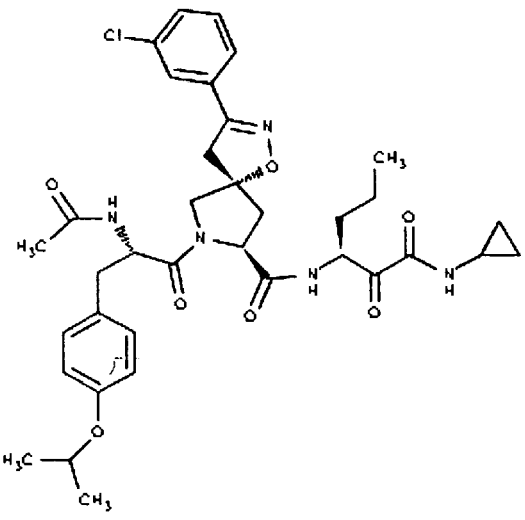
	315
	316
	317

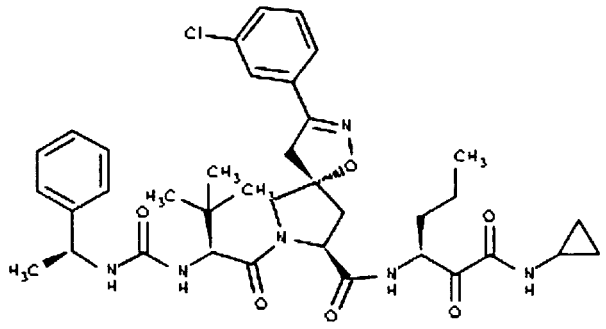
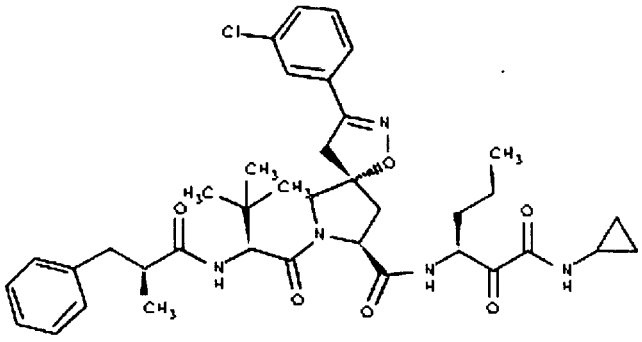
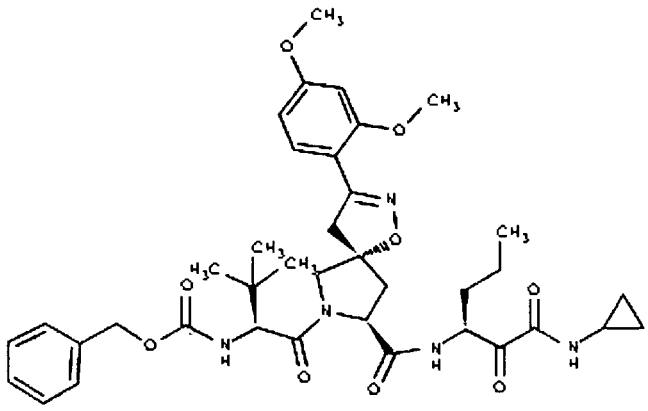
	318
	319
	320

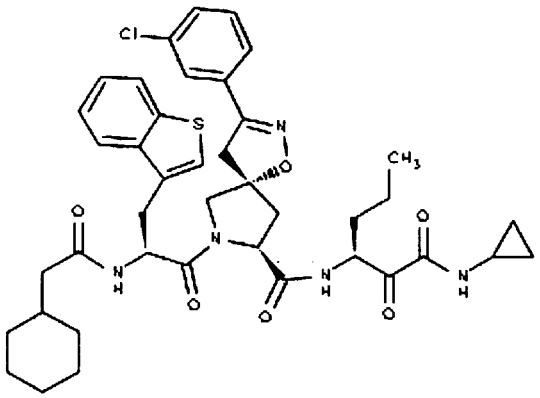
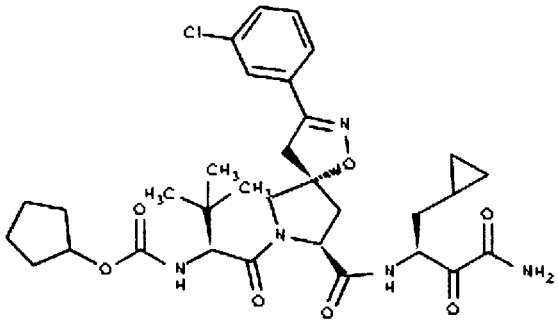
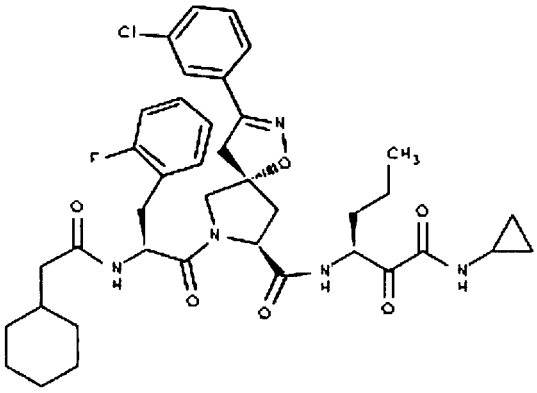
	321
	322
	323

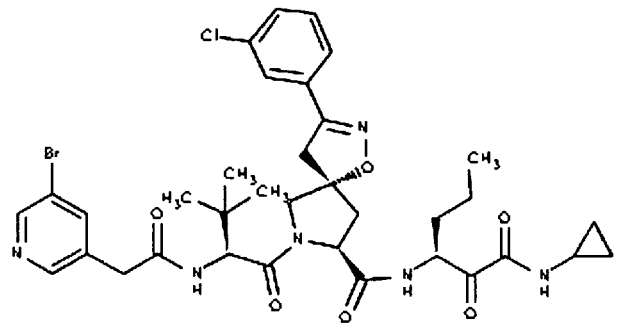
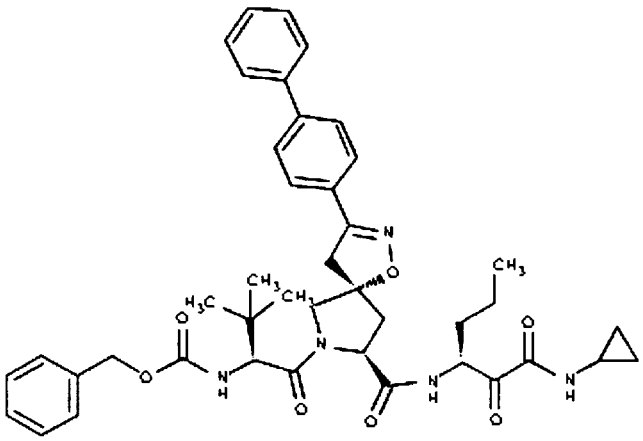
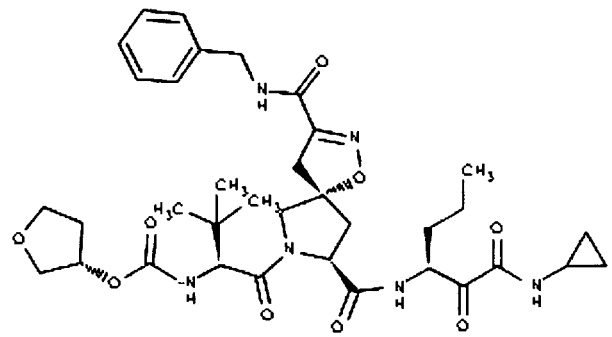
	324
	325
	326

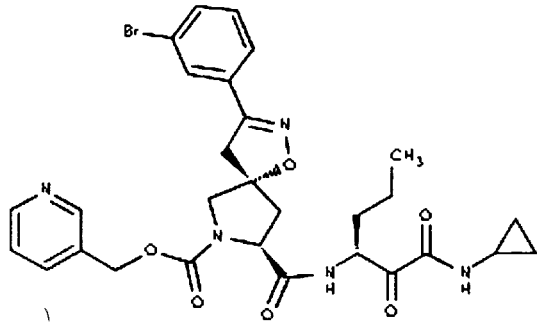
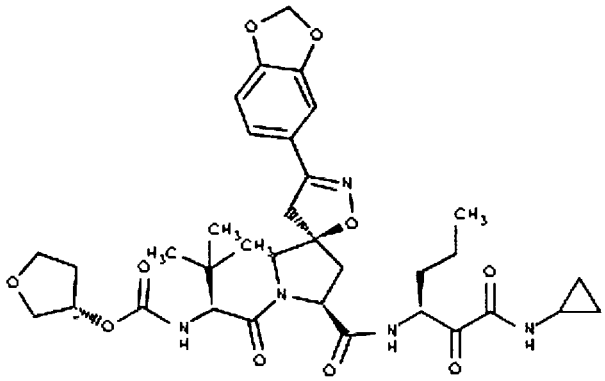
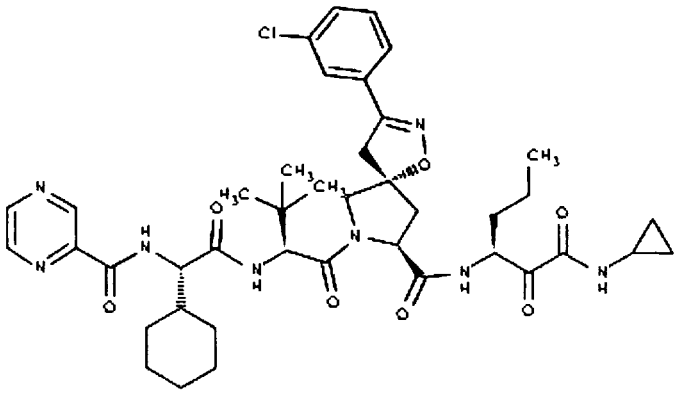
	327
	328
	329

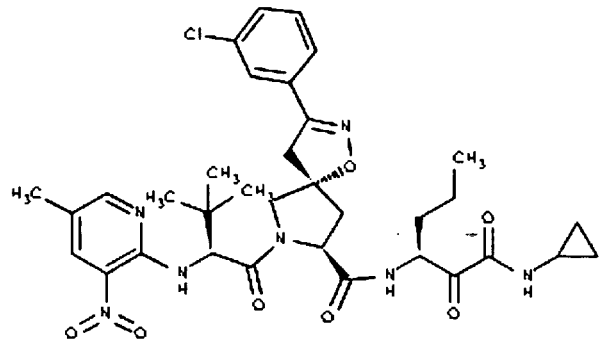
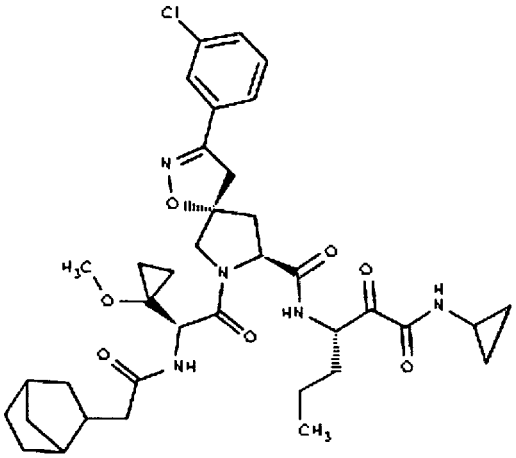
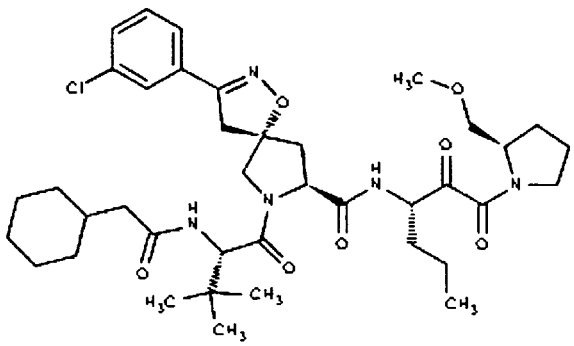
	330
	331
	332

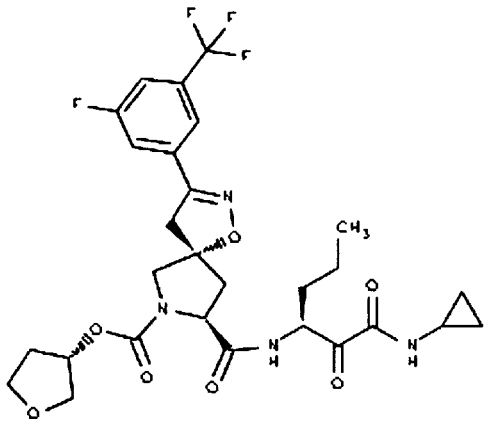
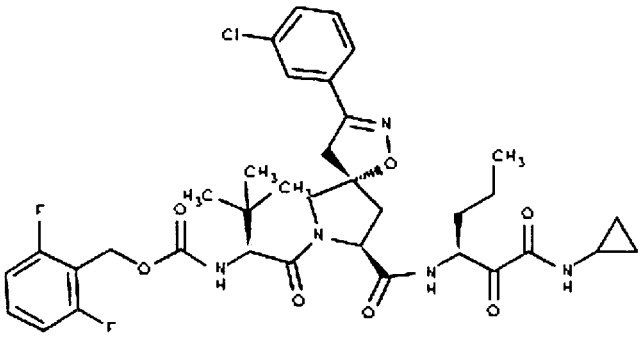
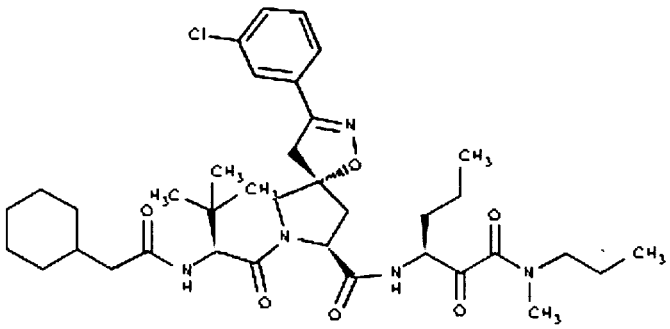
	333
	334
	335

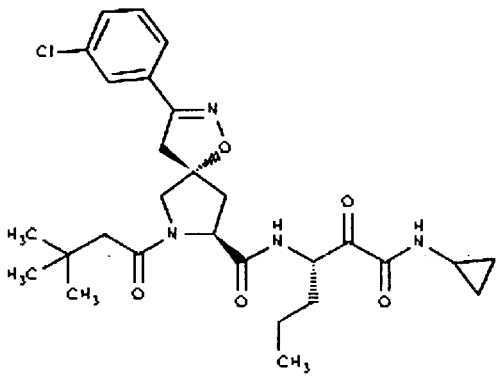
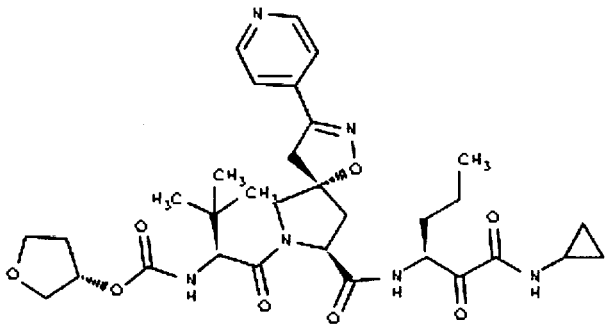
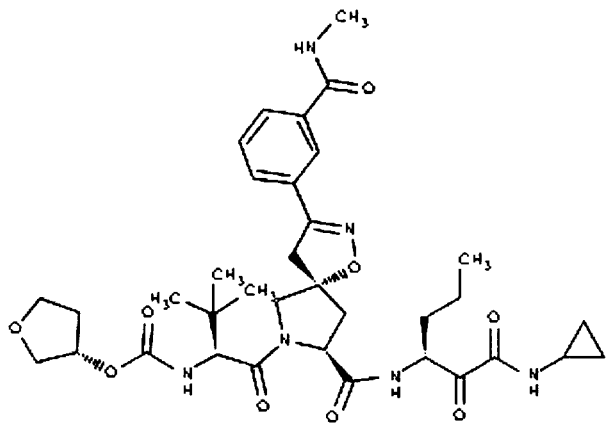
	336
	337
	338

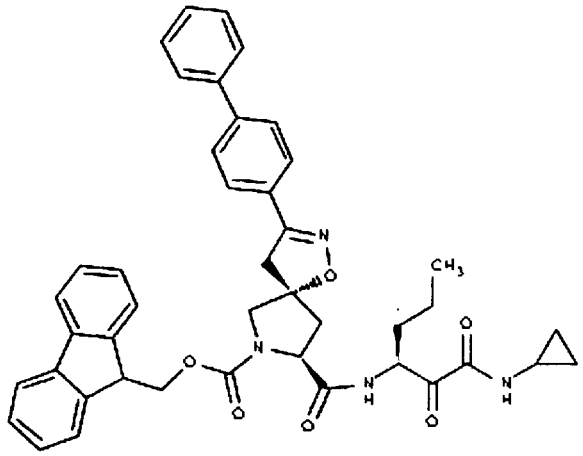
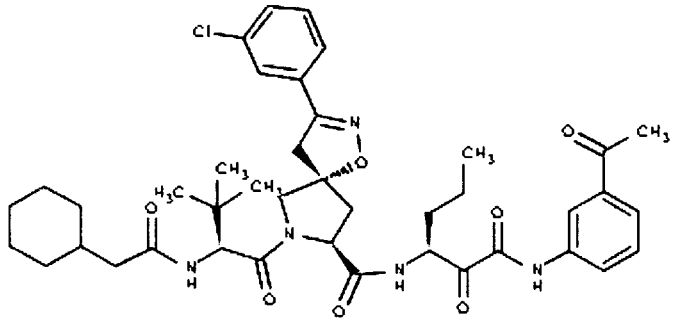
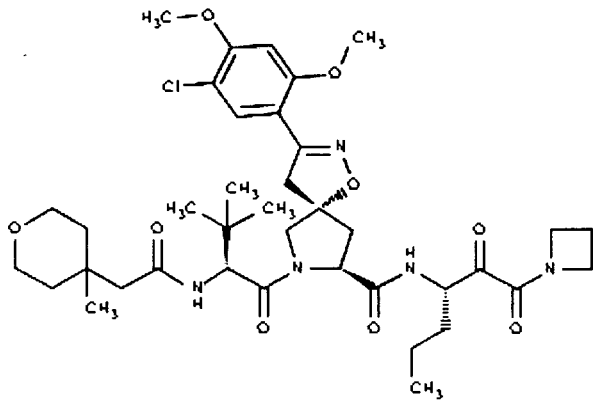
	339
	340
	341

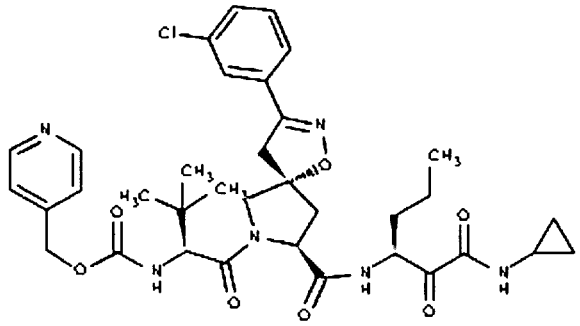
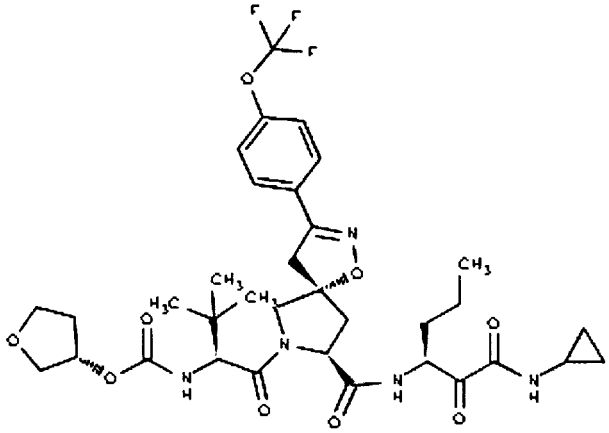
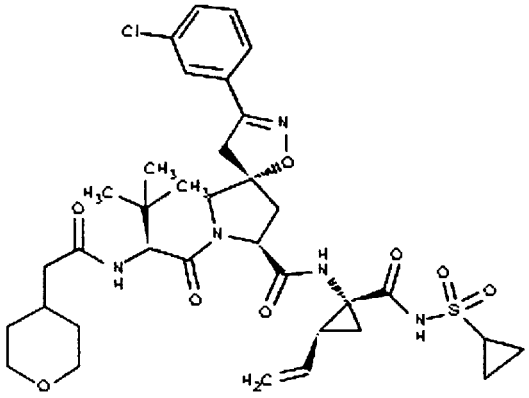
	342
	343
	344

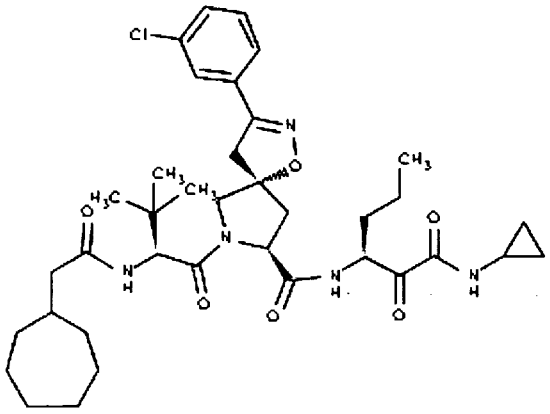
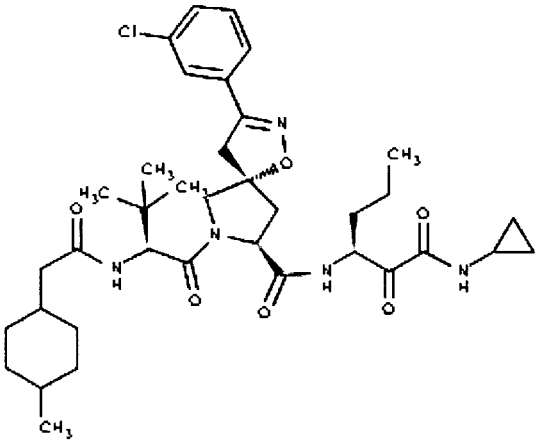
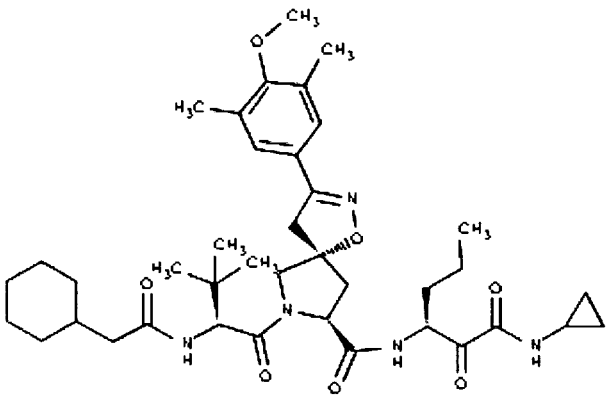
	345
	346
	347

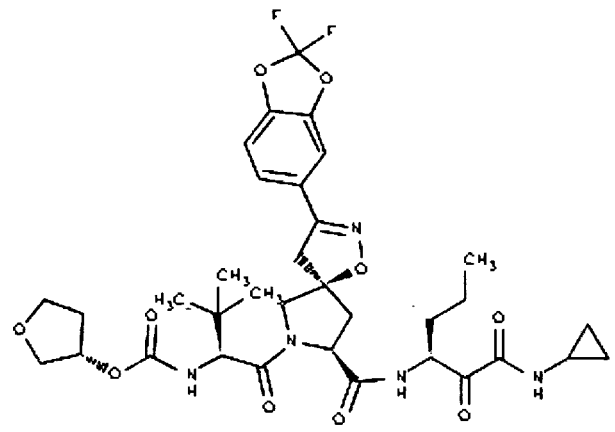
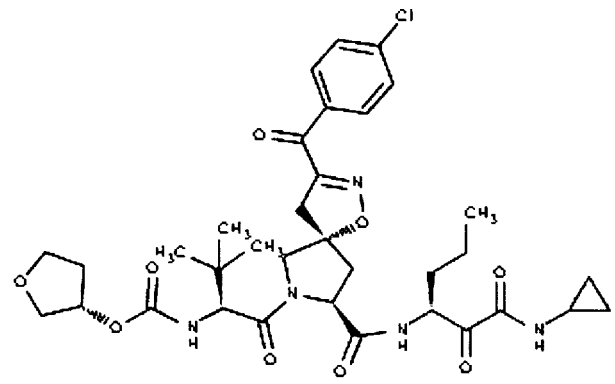
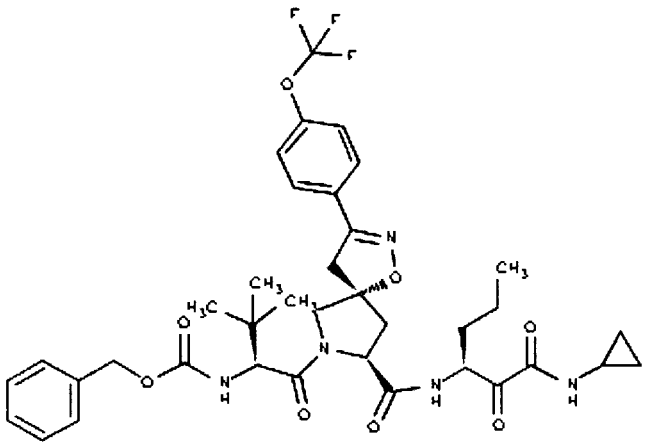
	348
	349
	350

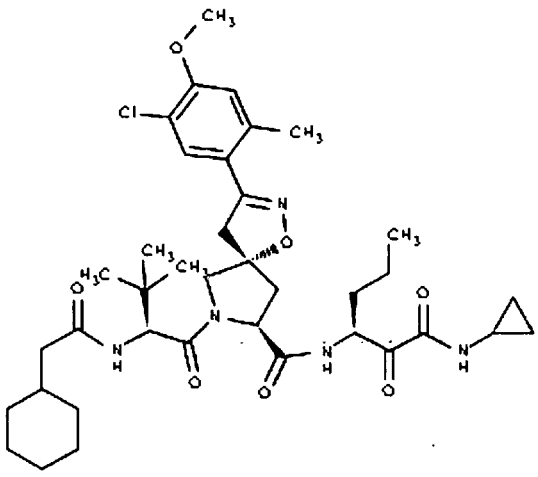
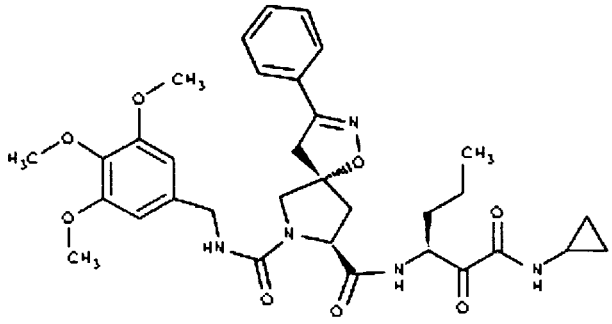
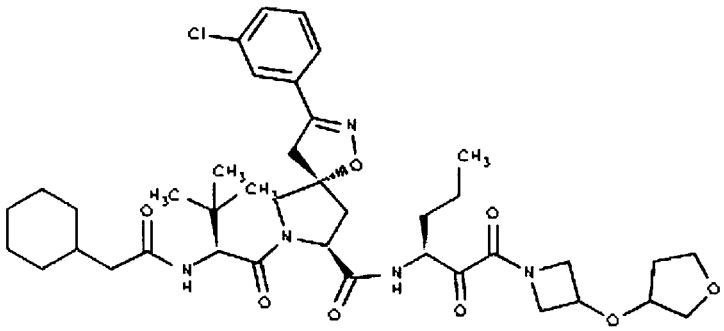
	351
	352
	353

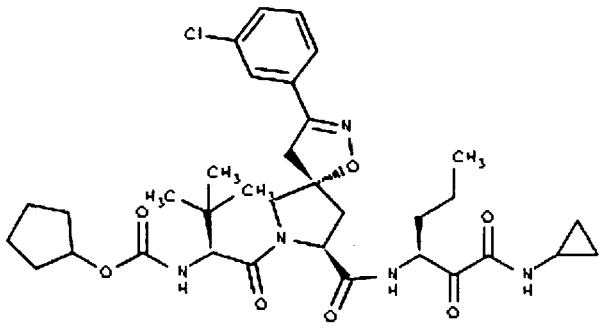
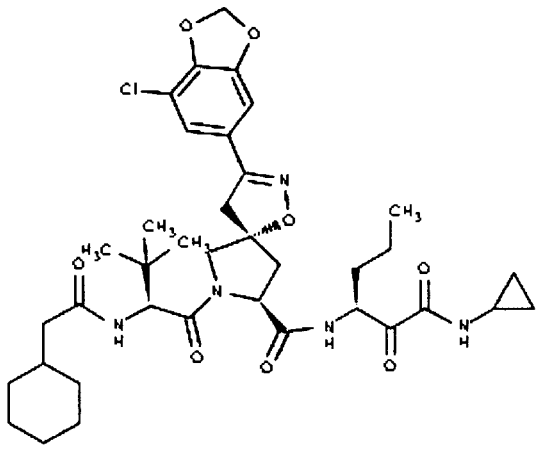
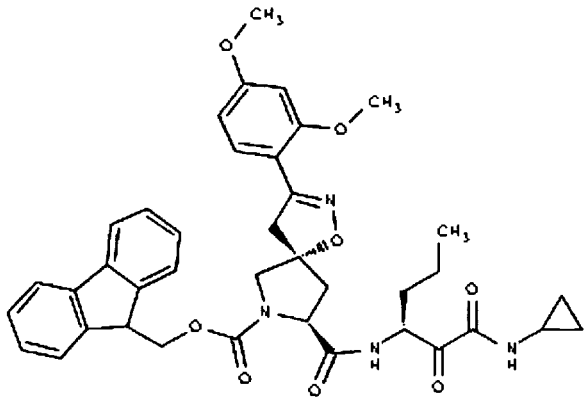
	354
	355
	356

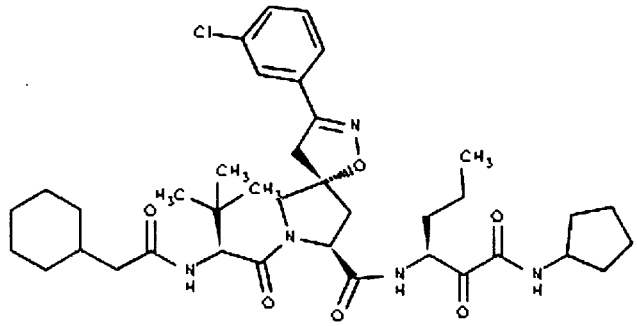
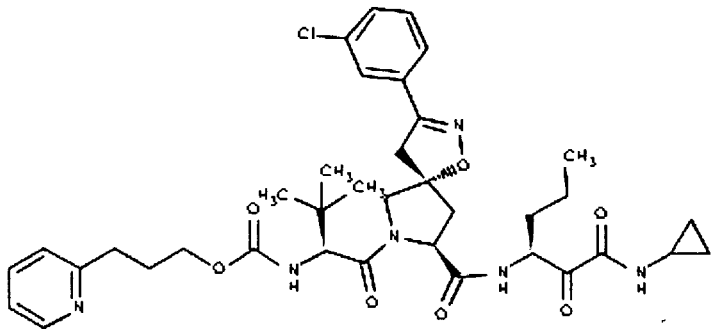
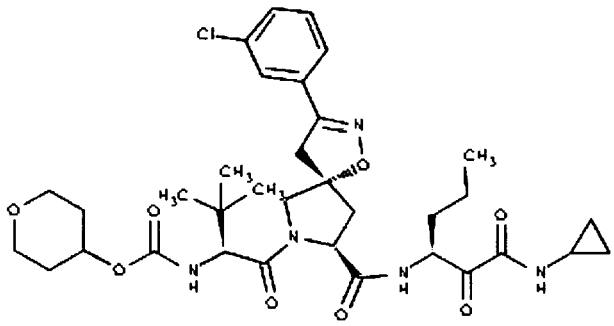
	357
	358
	359

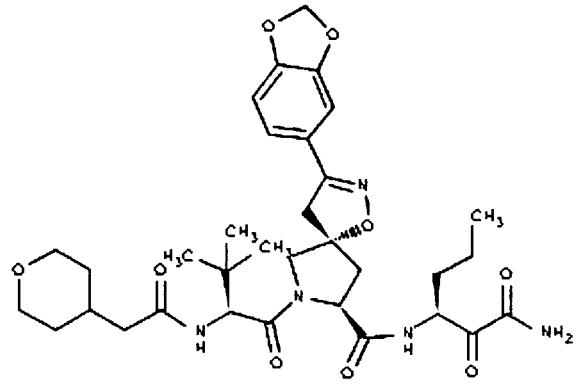
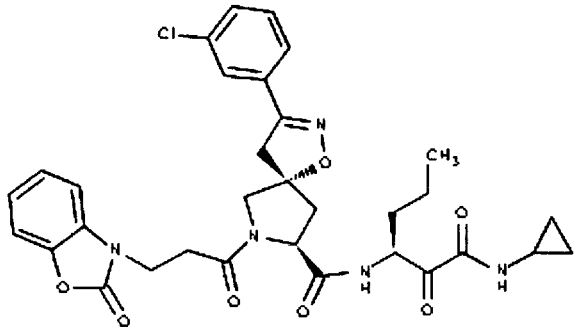
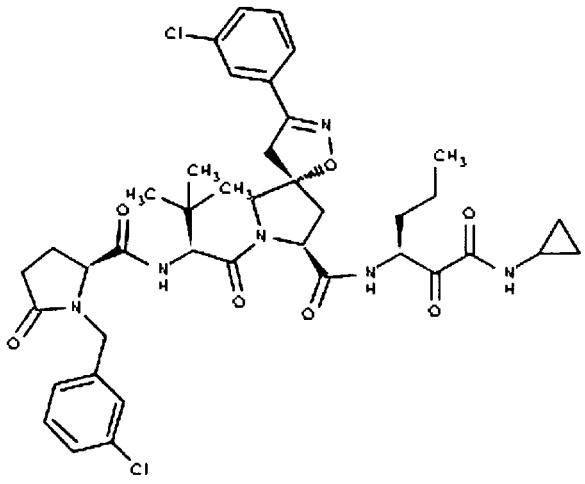
	360
	361
	362

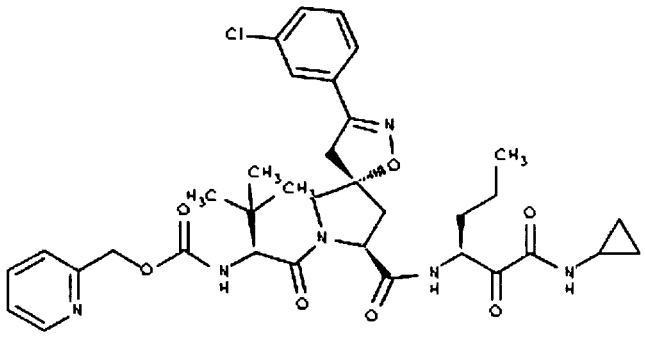
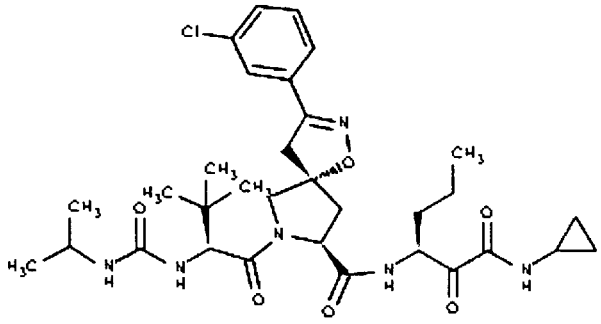
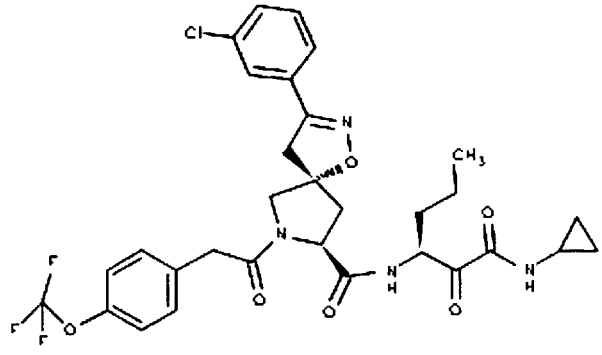
	363
	364
	365

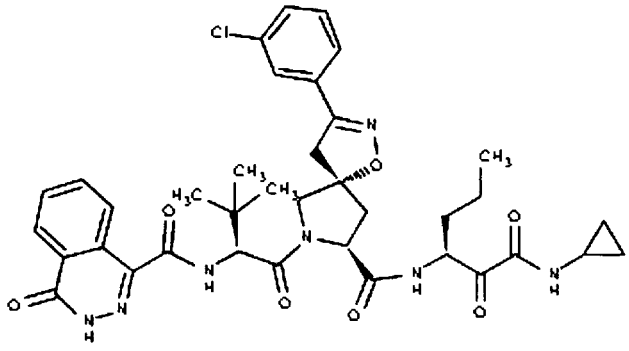
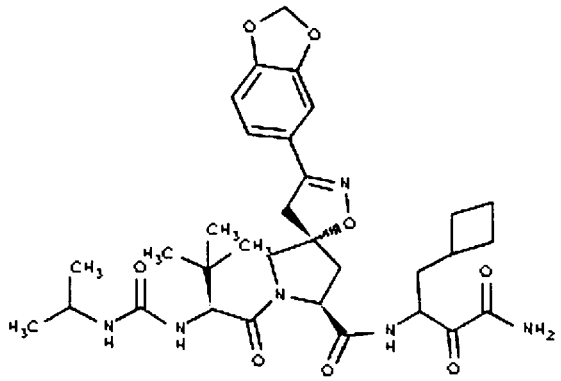
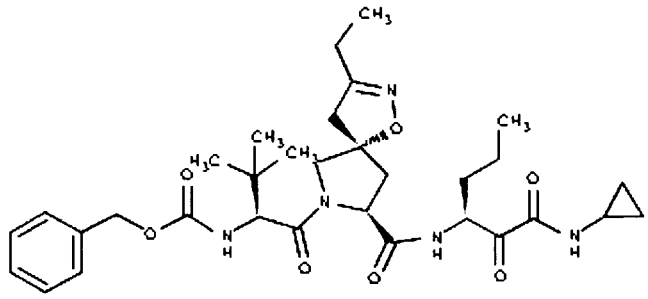
	366
	367
	368

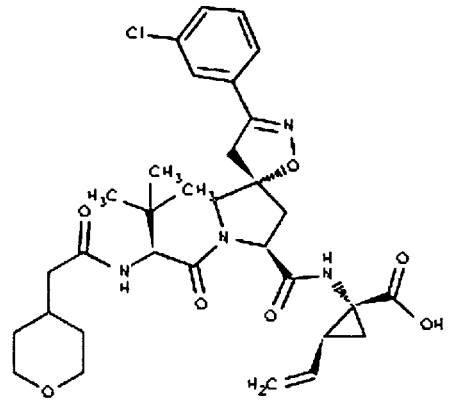
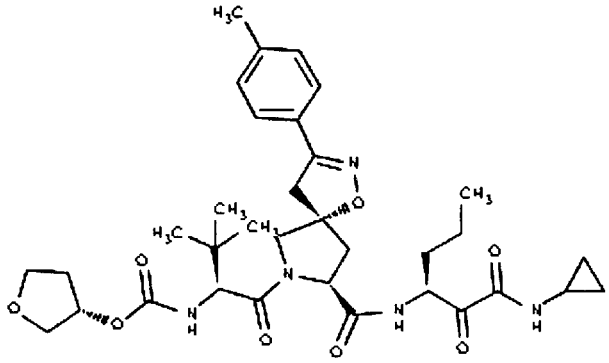
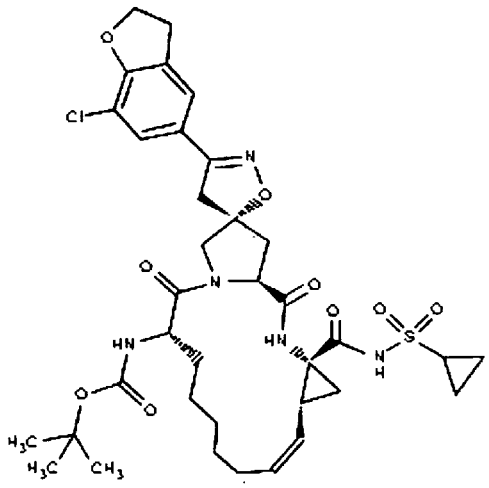
	369
	370
	371

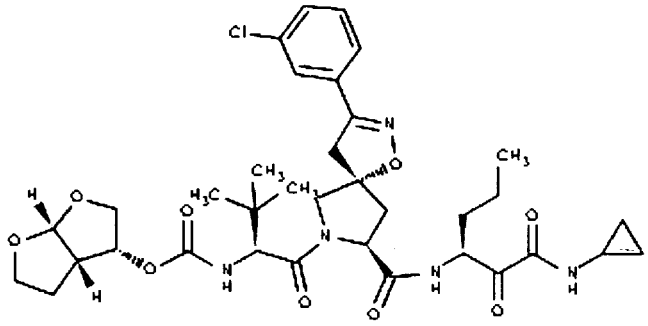
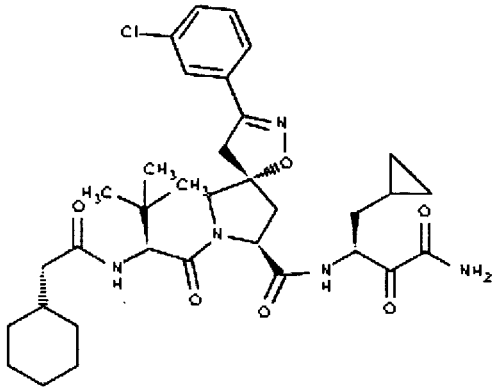
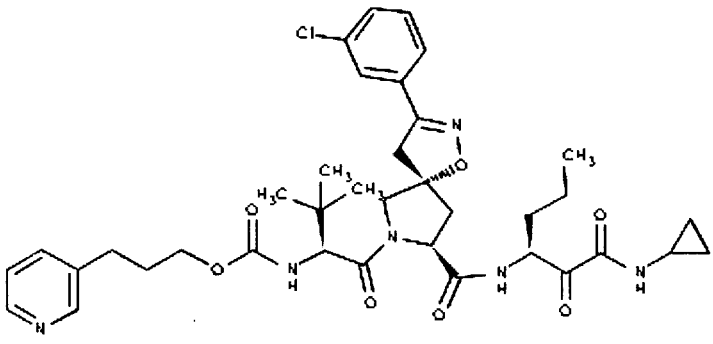
	372
	373
	374

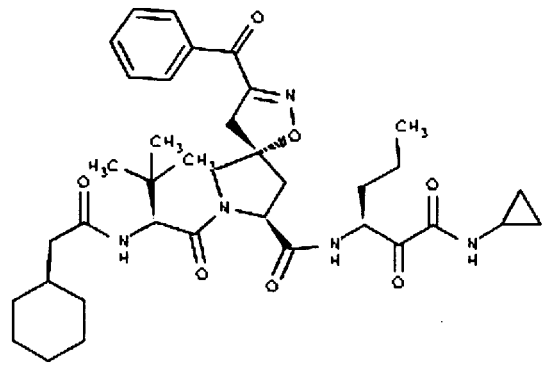
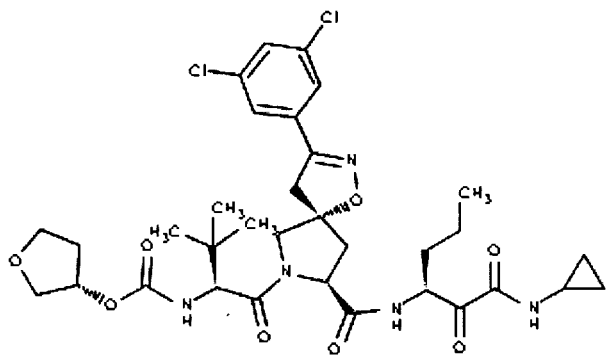
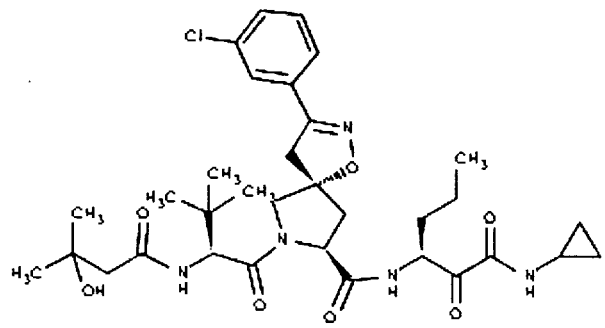
	375
	376
	377

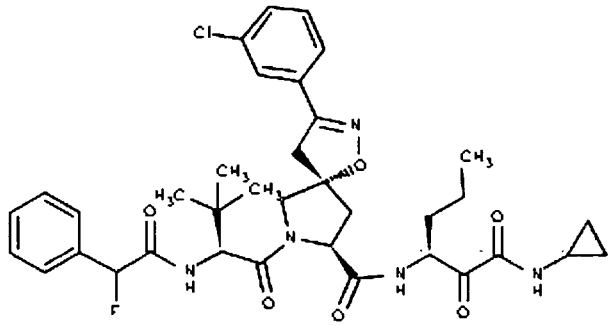
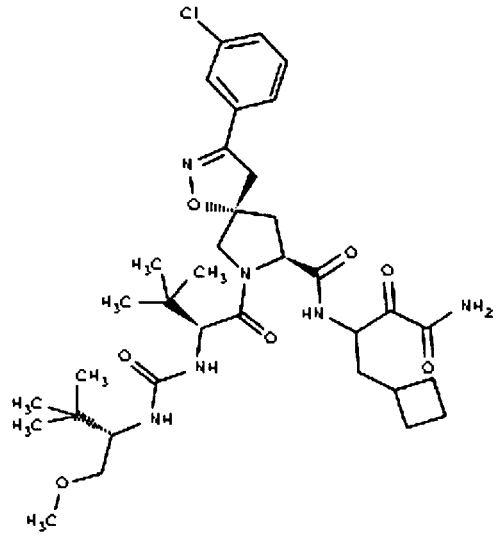
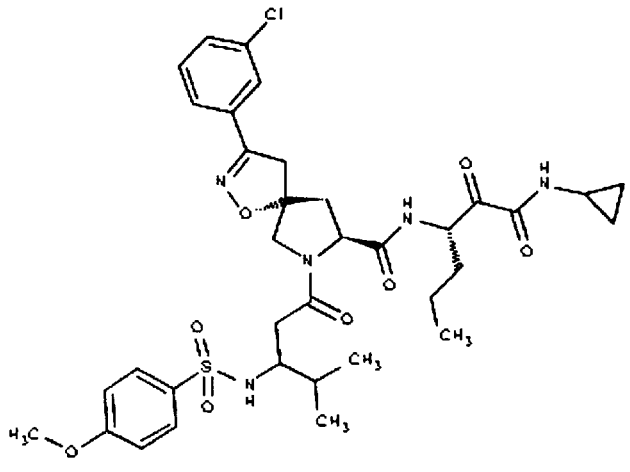
	378
	379
	380

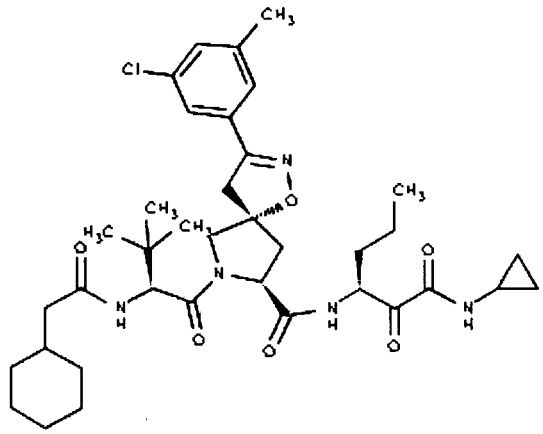
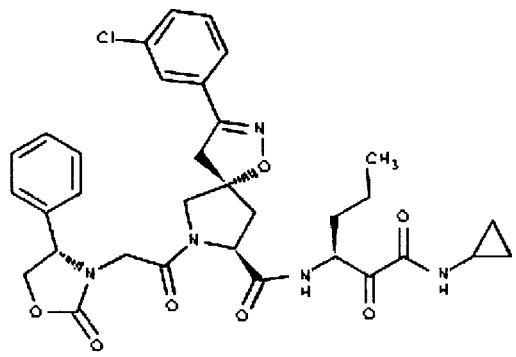
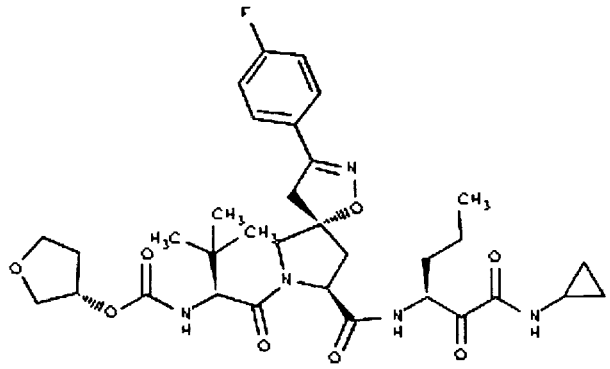
	381
	382
	383

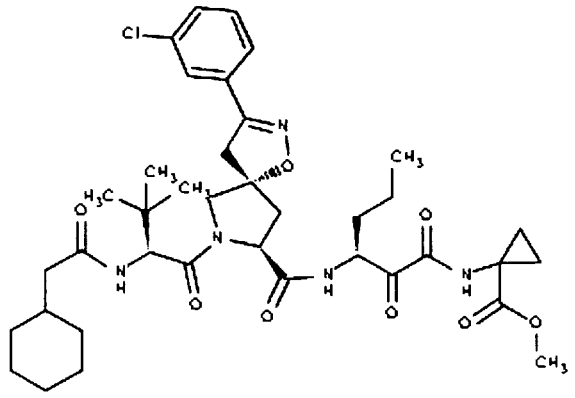
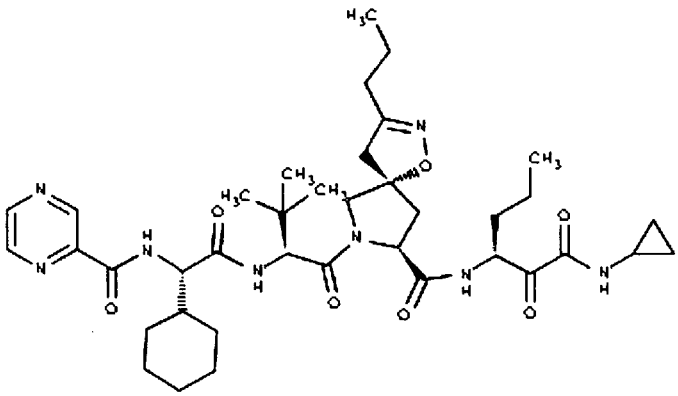
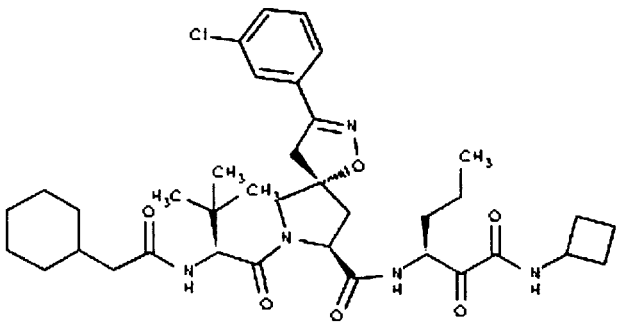
	384
	385
	386

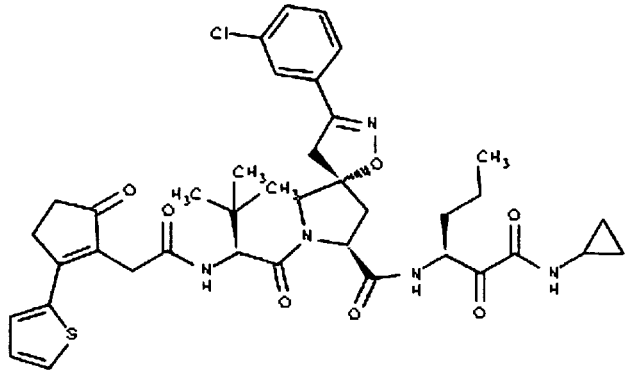
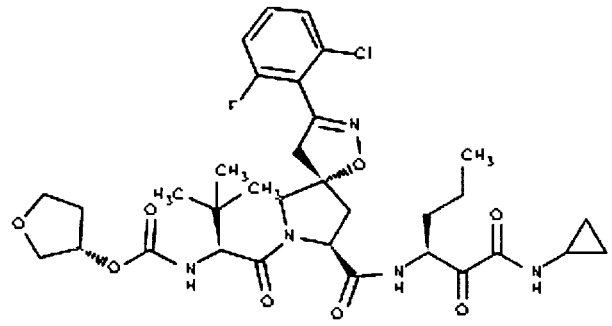
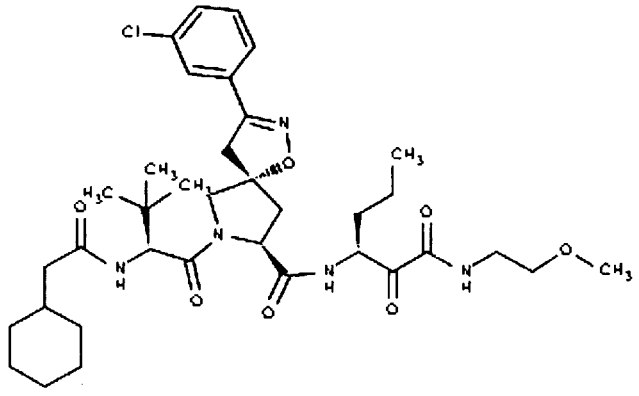
	387
	388
	389

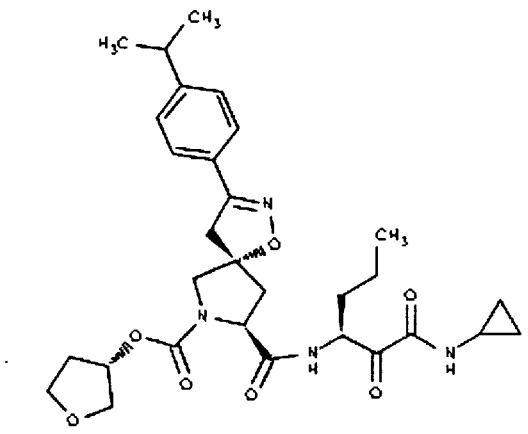
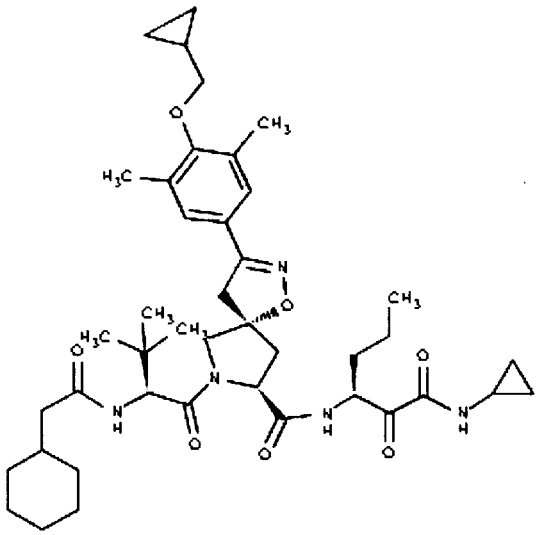
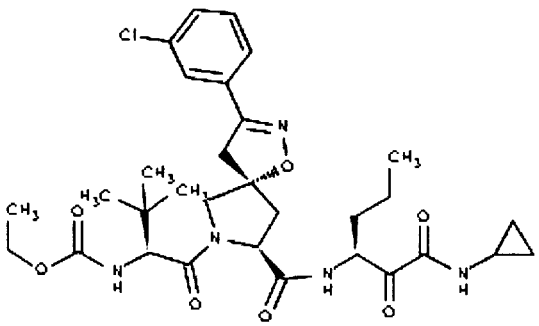
	390
	391
	392

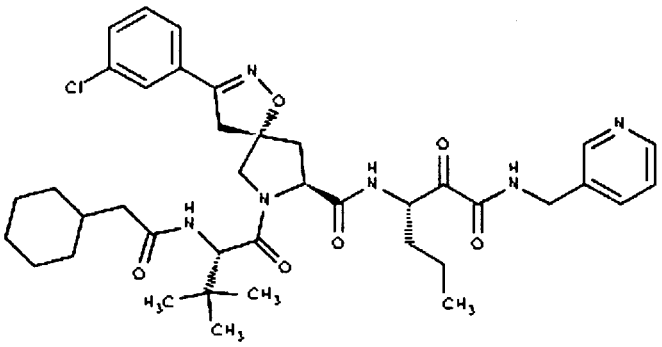
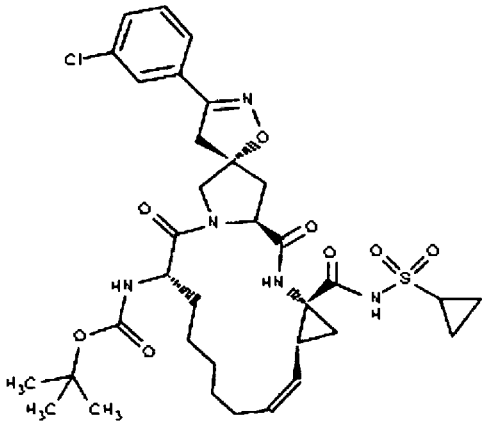
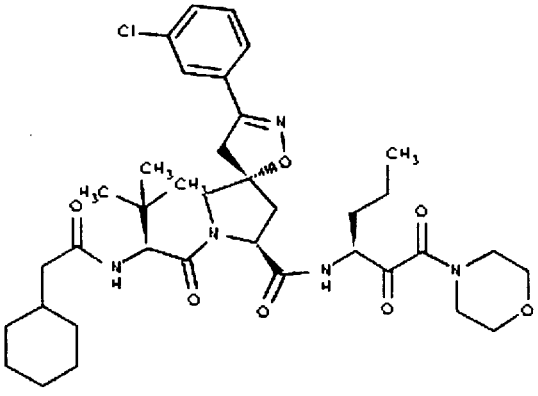
	393
	394
	395

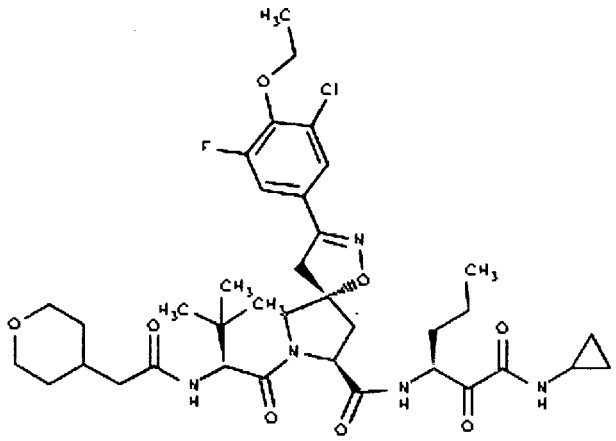
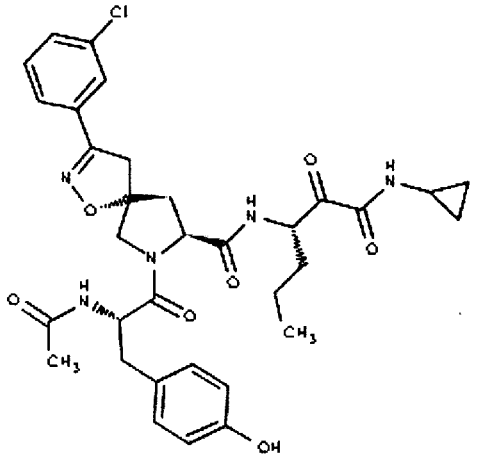
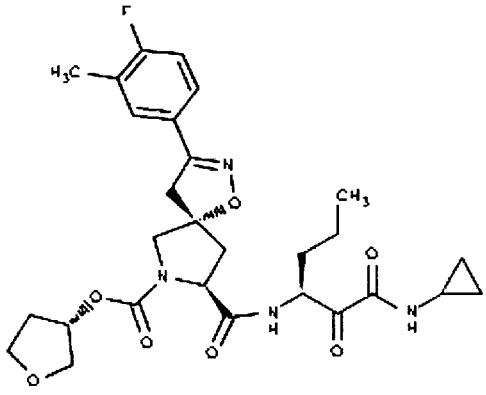
	396
	397
	398

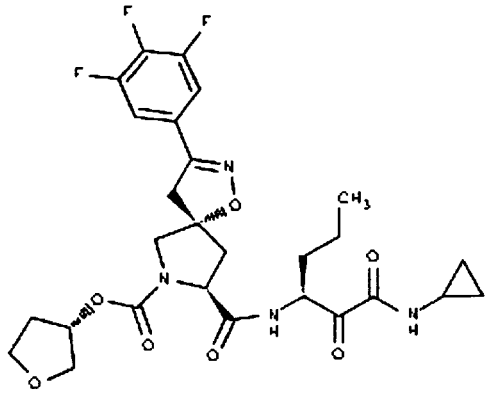
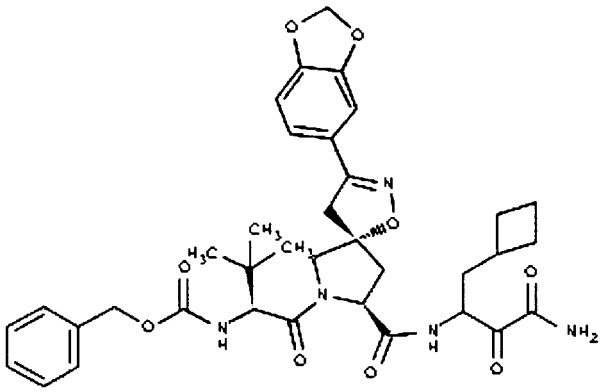
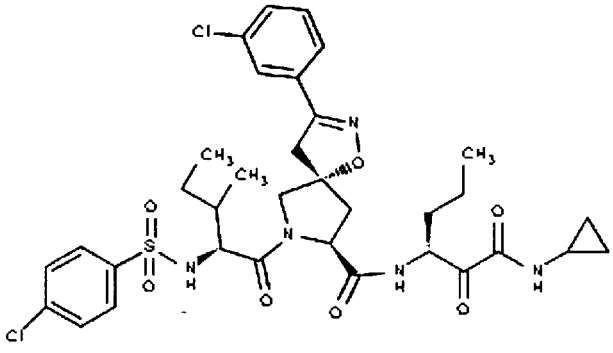
	399
	400
	401

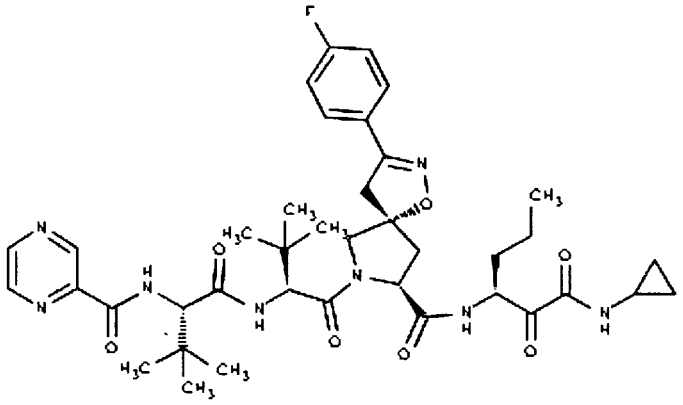
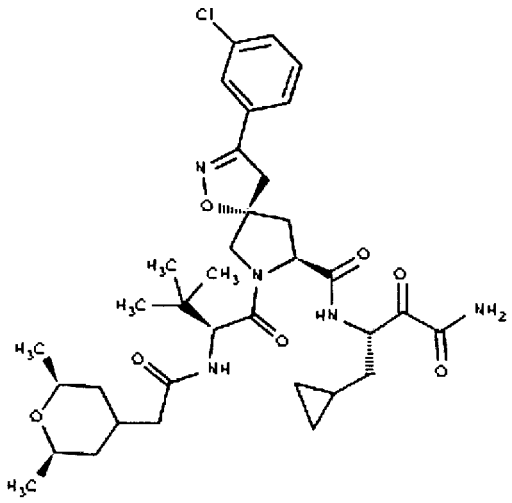
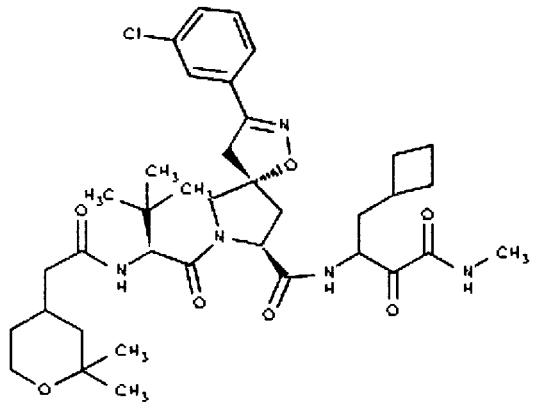
	402
	403
	404

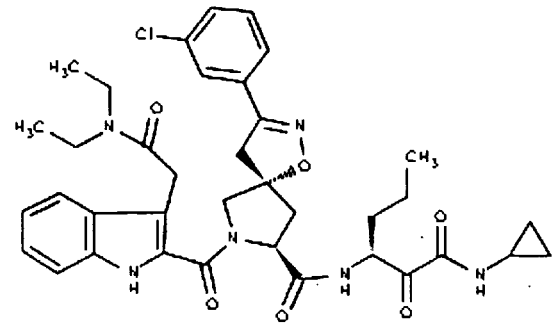
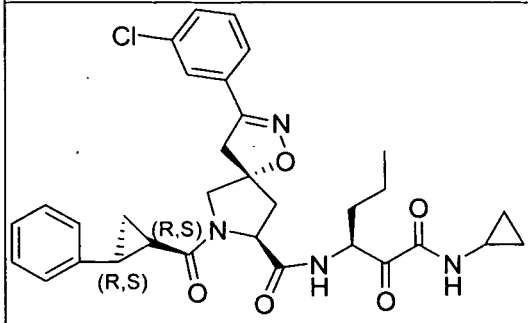
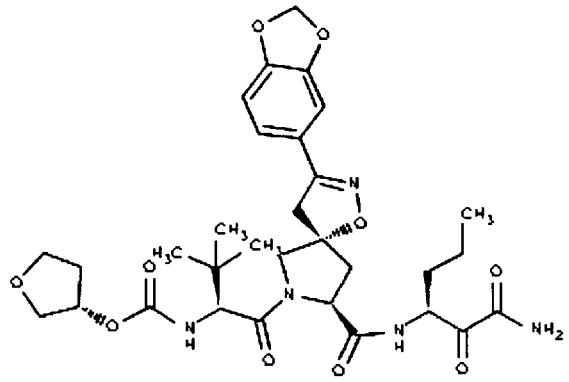
	405
	406
	407

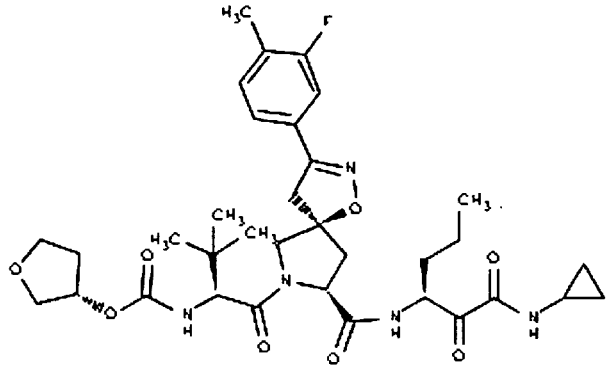
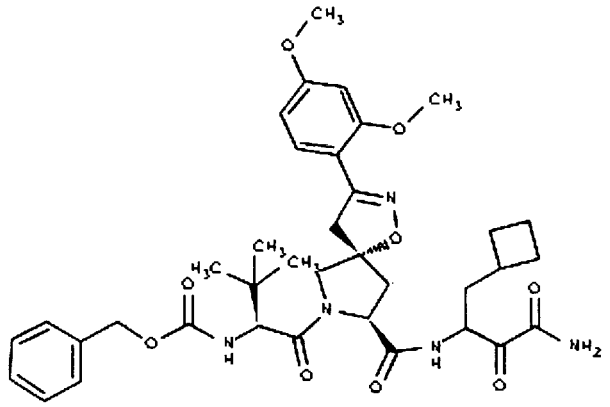
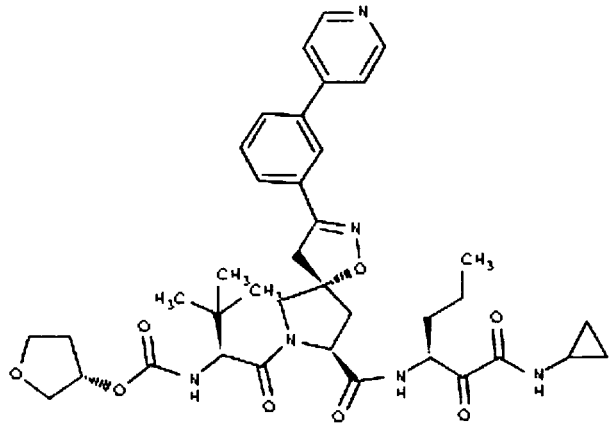
	408
	409
	410

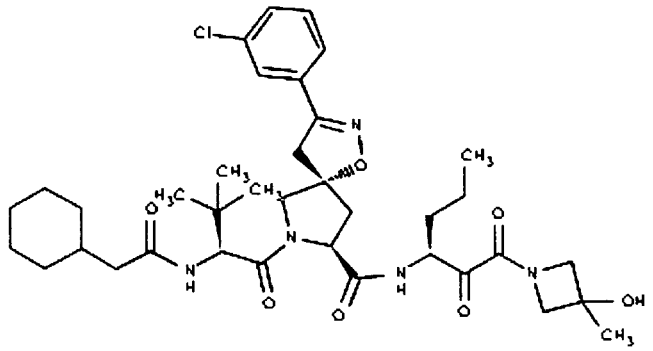
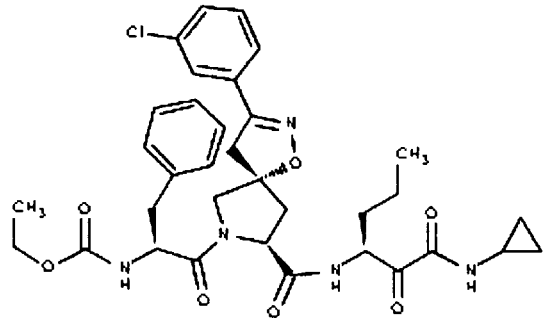
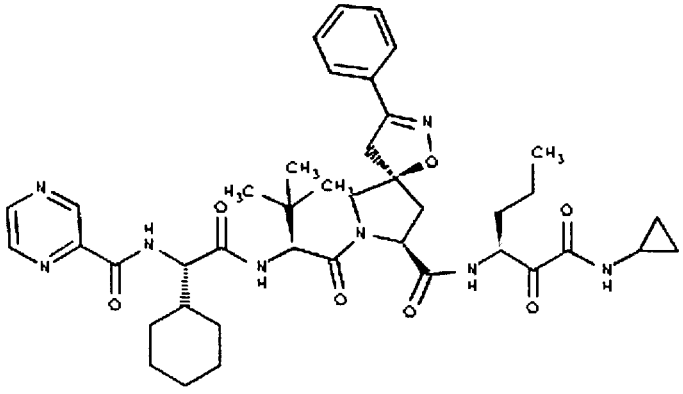
	411
	412
	413

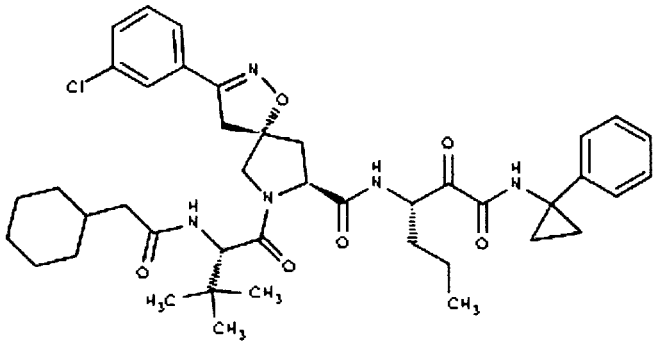
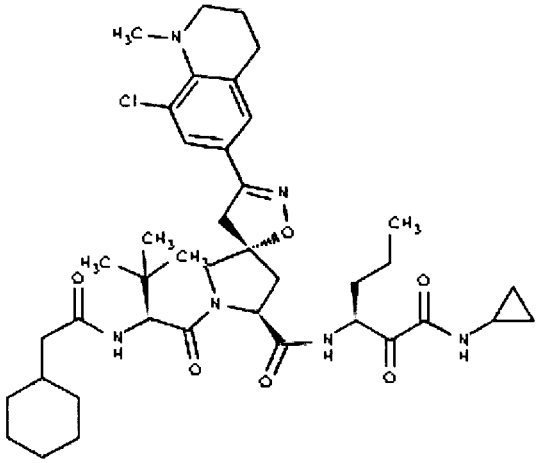
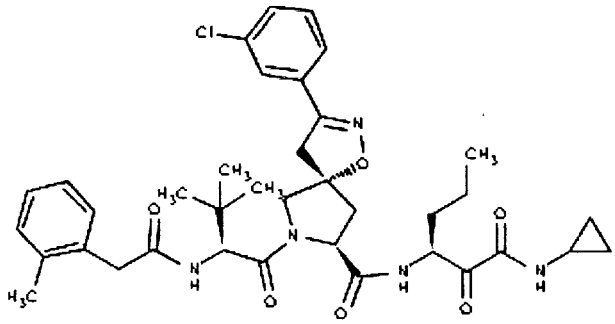
	414
	415
	416

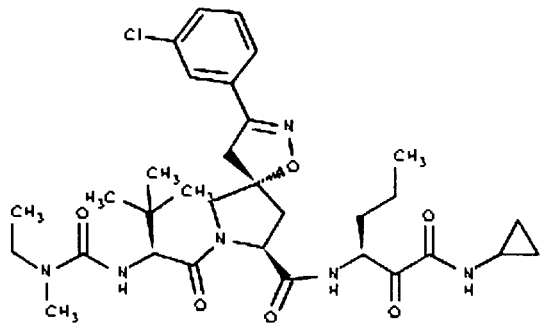
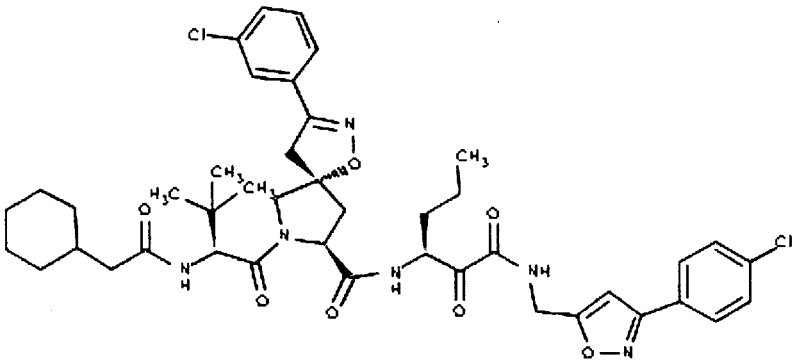
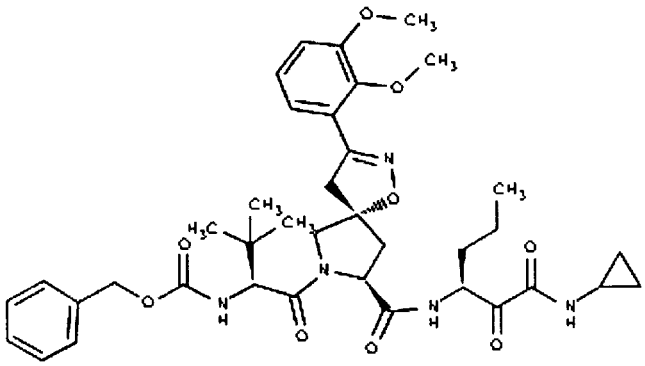
	417
	418
	419

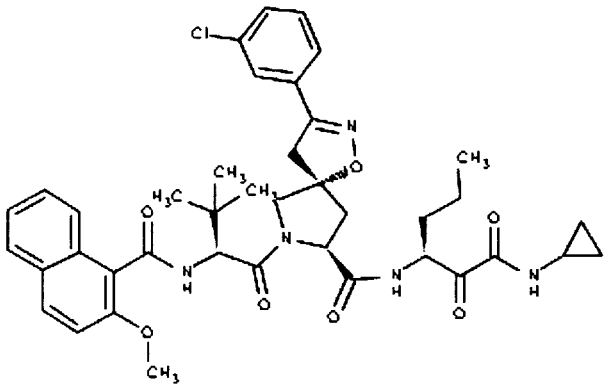
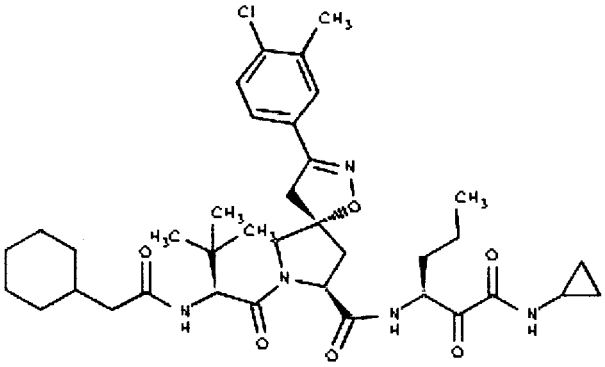
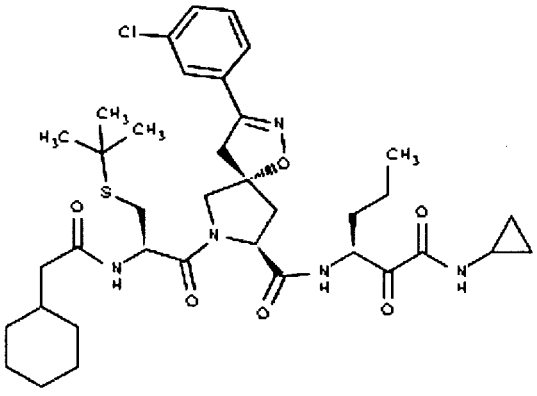
	420
	421
	422

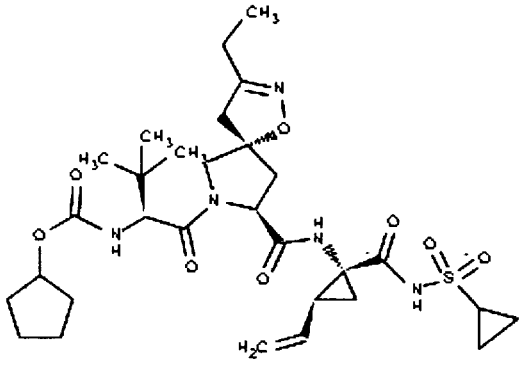
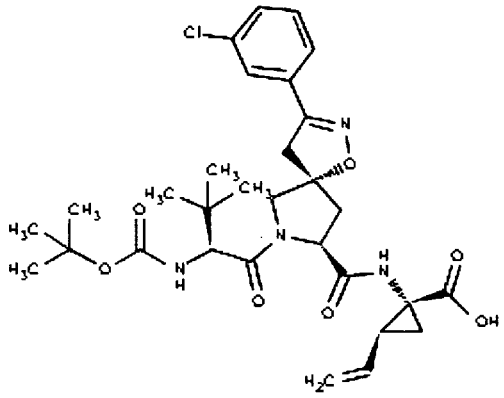
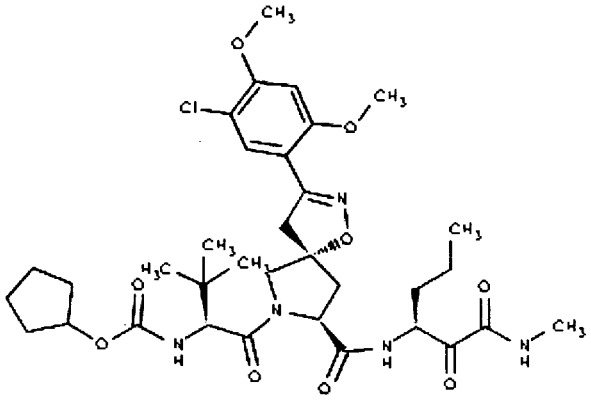
	423
	424
	425

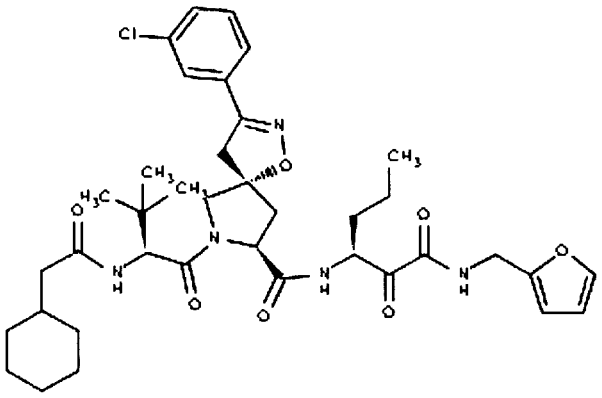
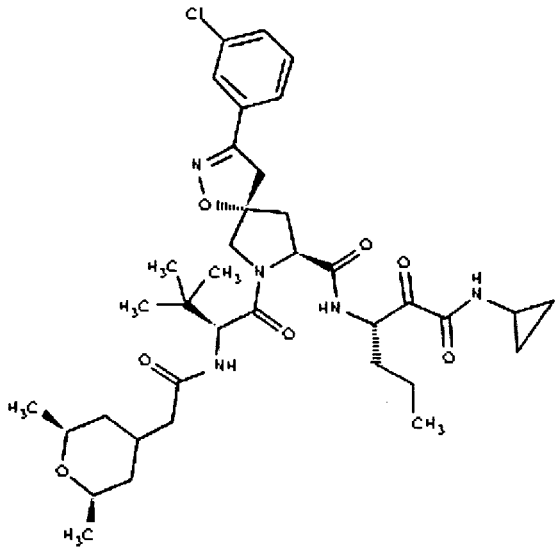
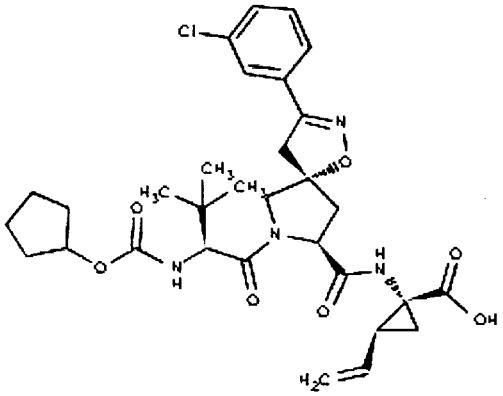
	426
	427
	428

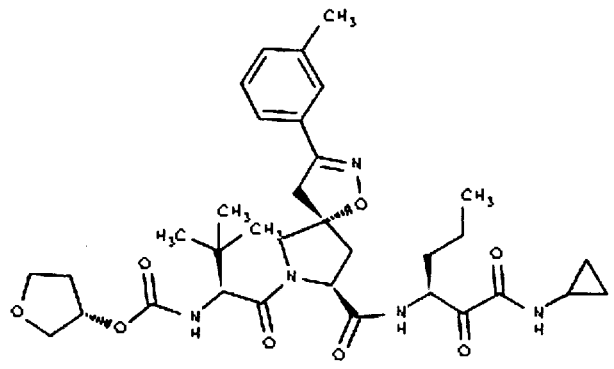
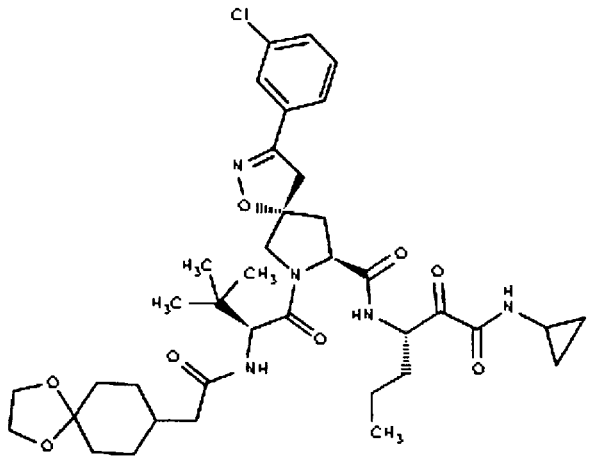
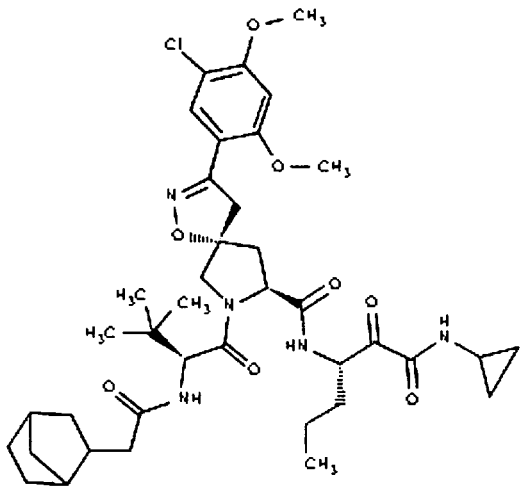
	429
	430
	431

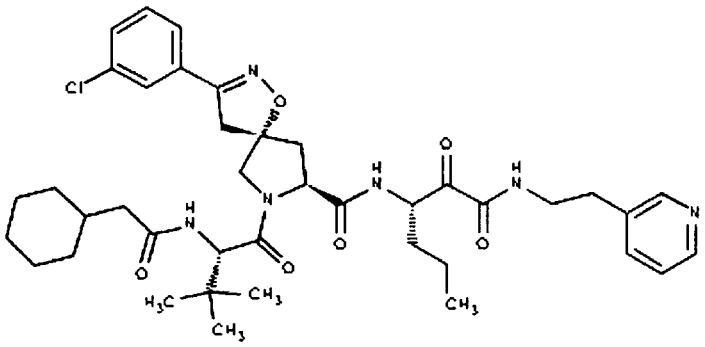
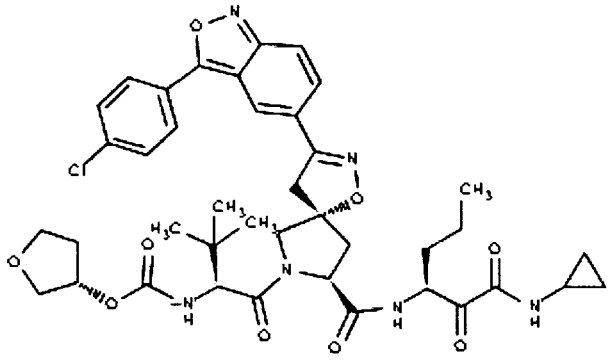
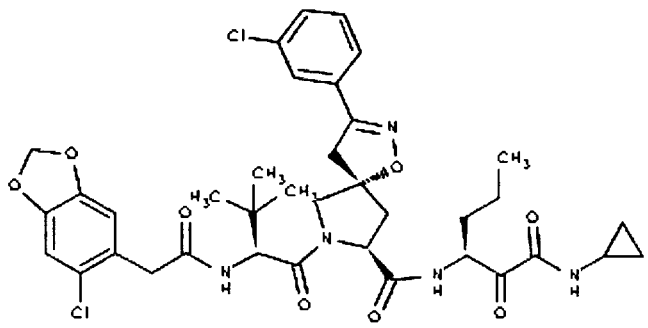
	432
	433
	434

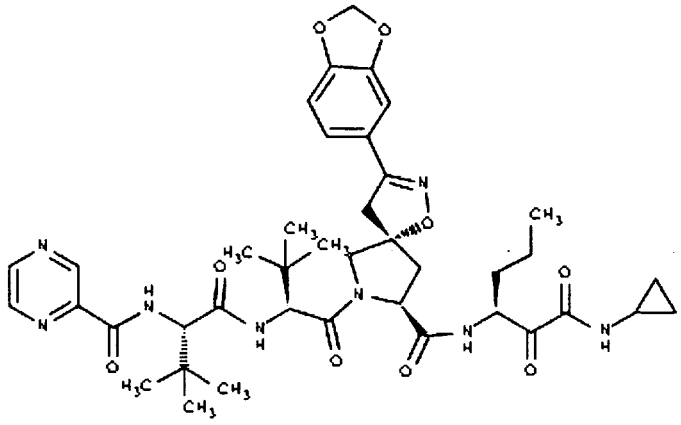
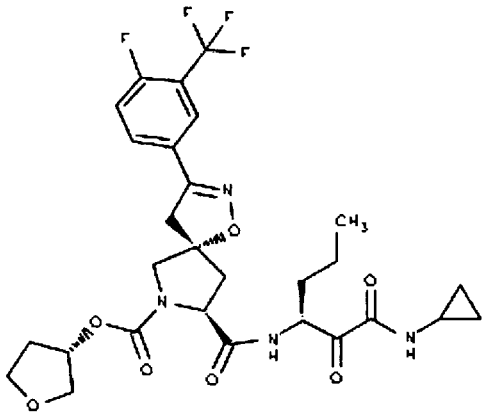
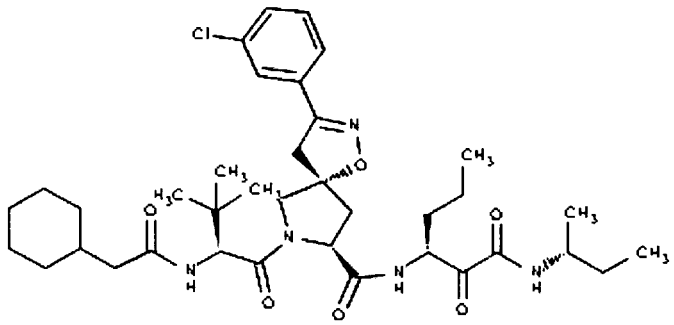
	435
	436
	437

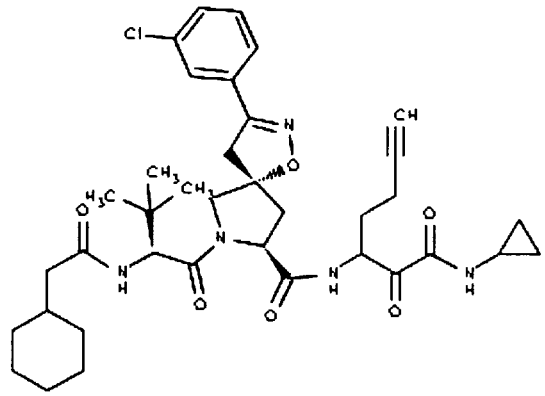
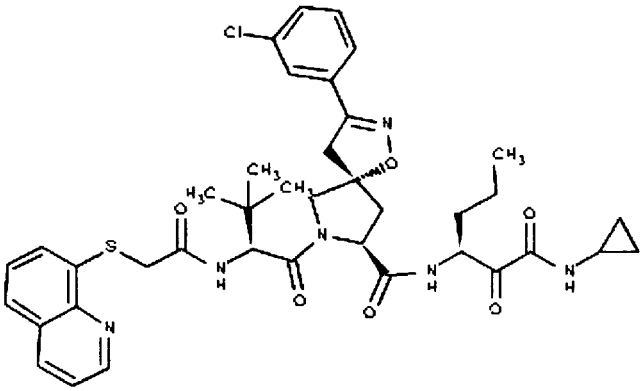
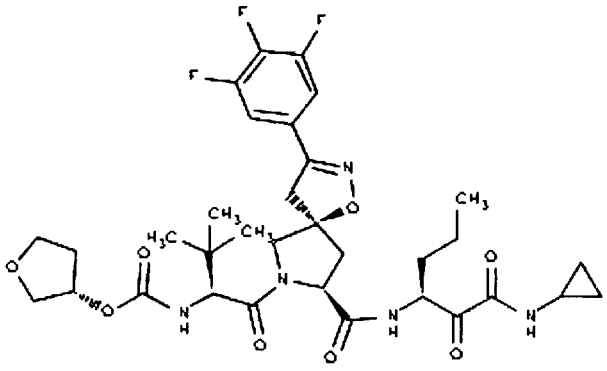
	438
	439
	440

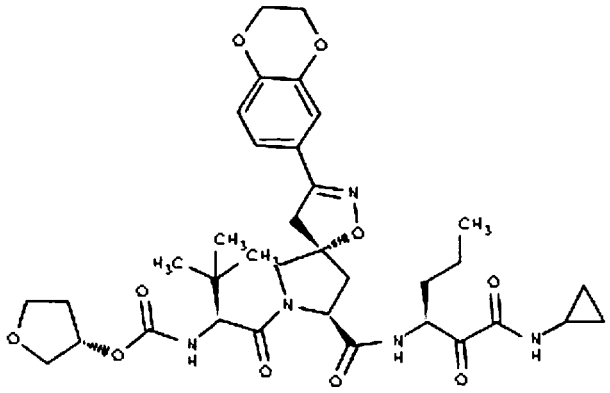
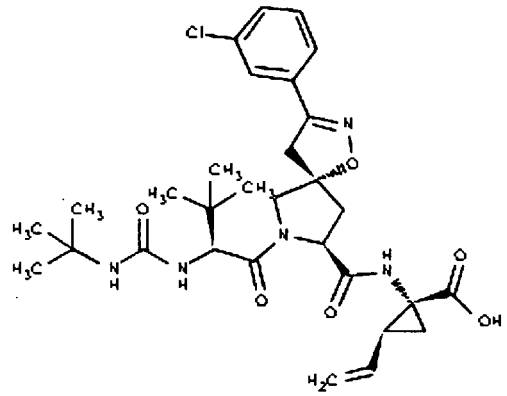
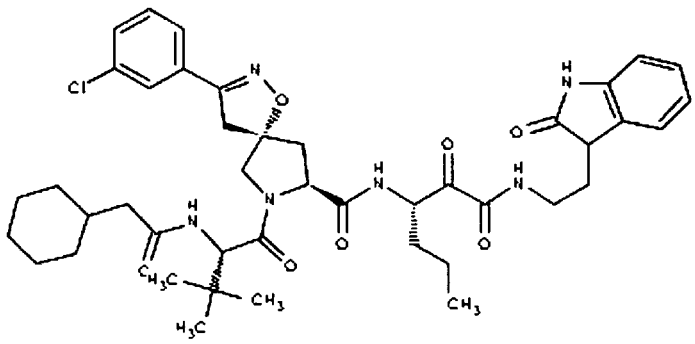
	441
	442
	443

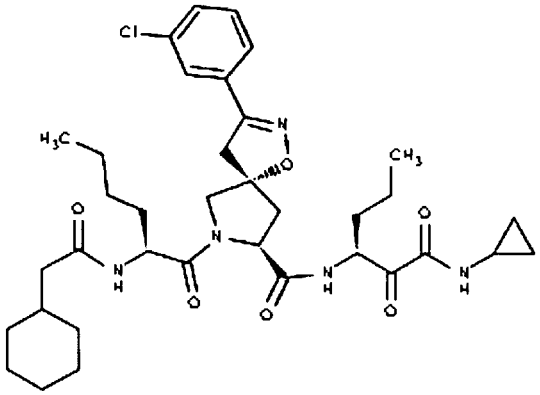
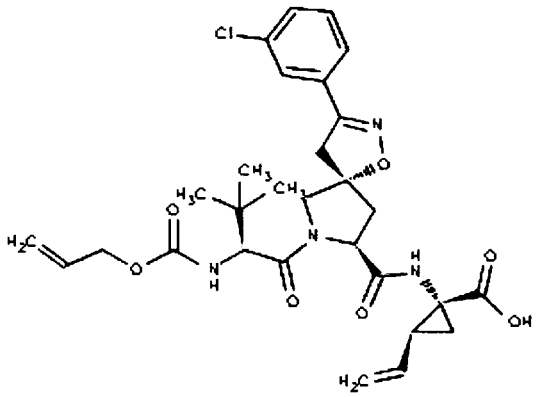
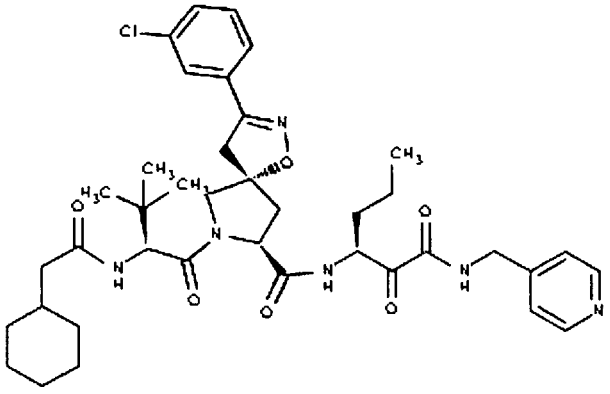
	444
	445
	446

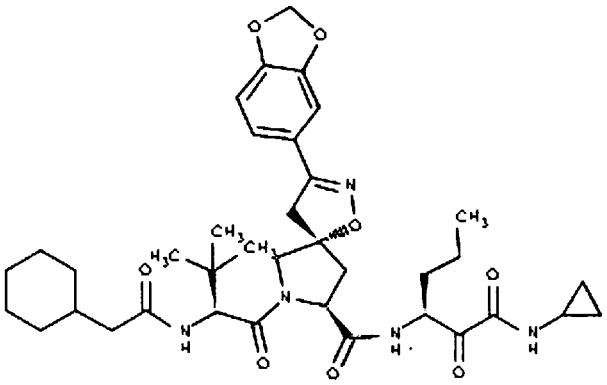
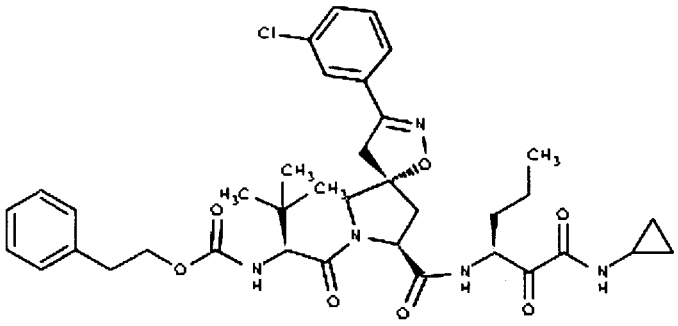
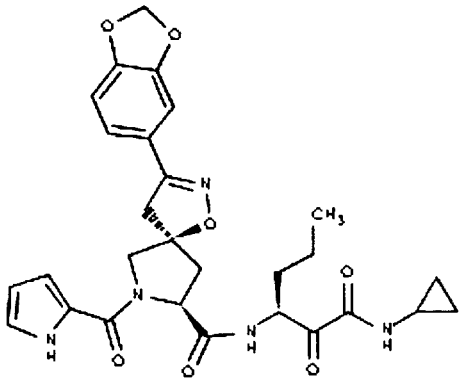
	447
	448
	449

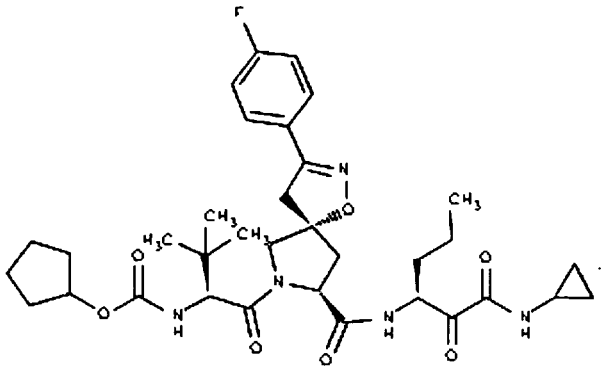
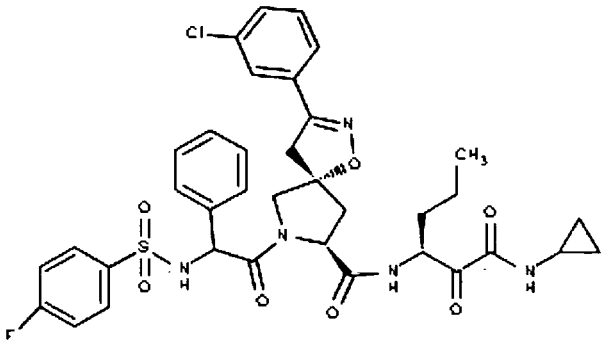
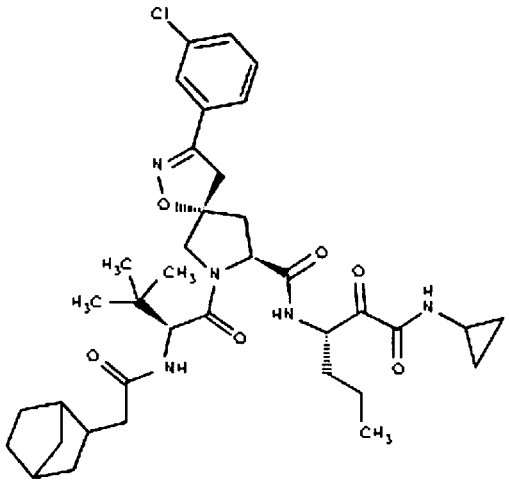
	450
	451
	452

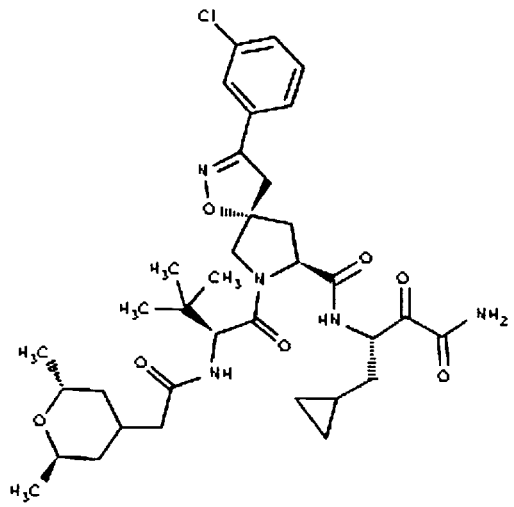
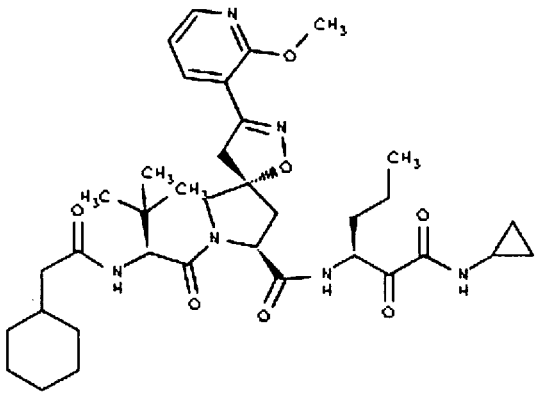
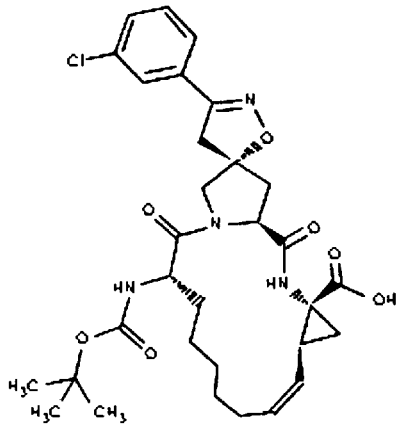
	453
	454
	455

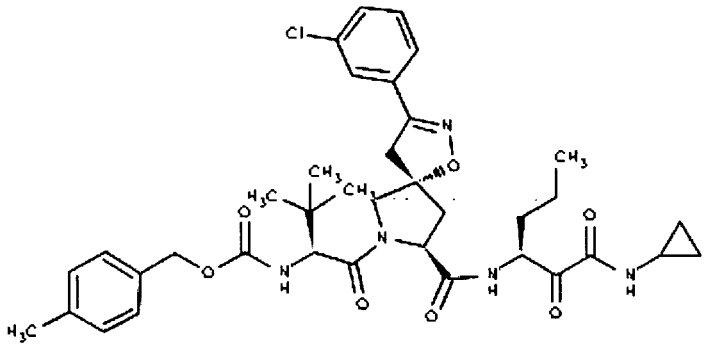
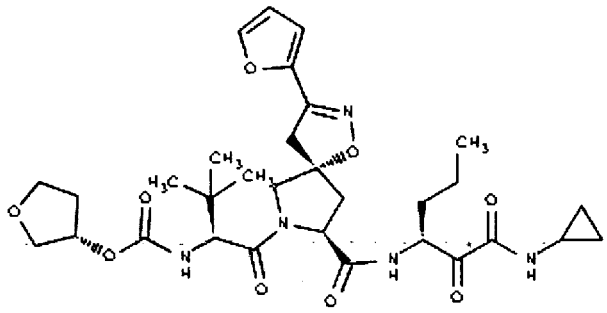
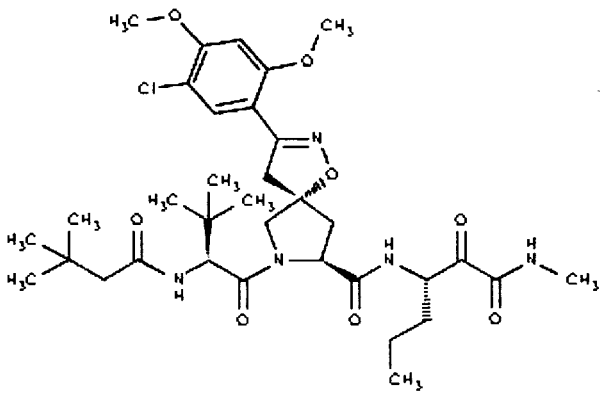
	456
	457
	458

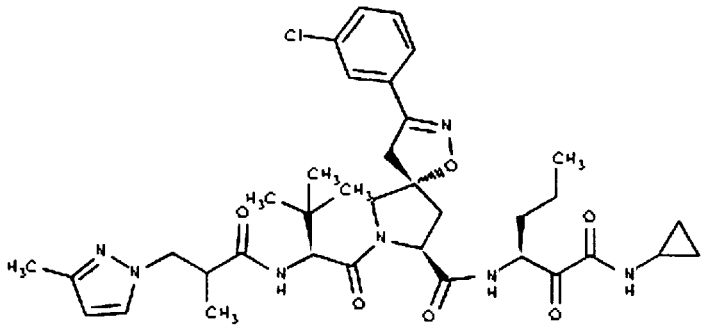
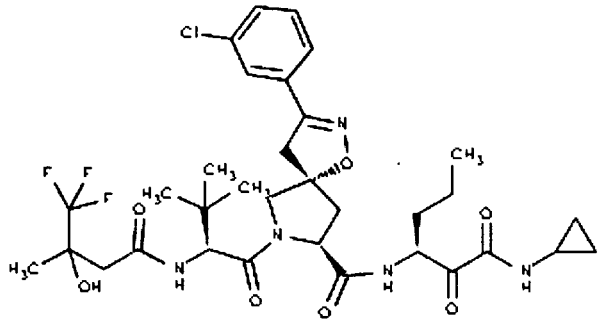
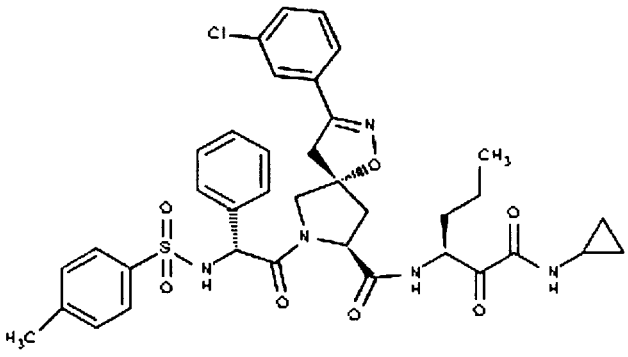
	459
	460
	461

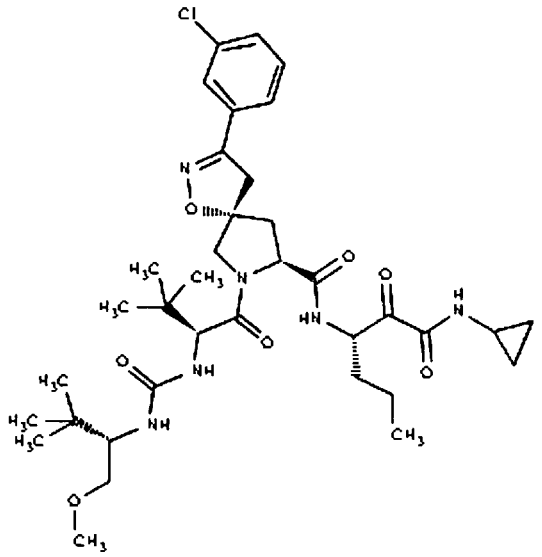
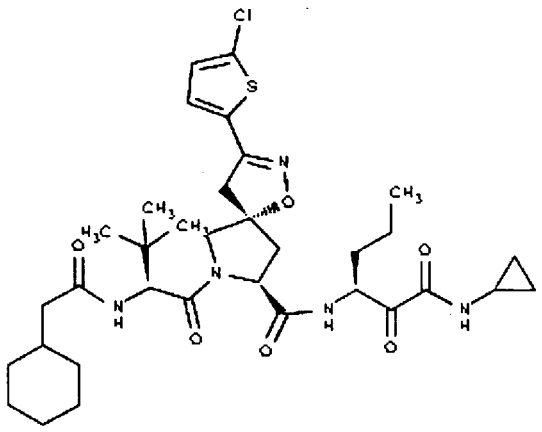
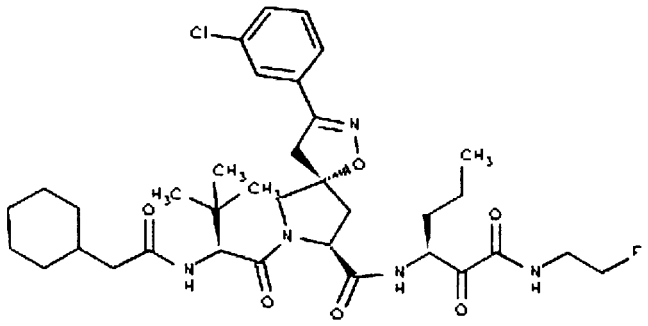
	462
	463
	464

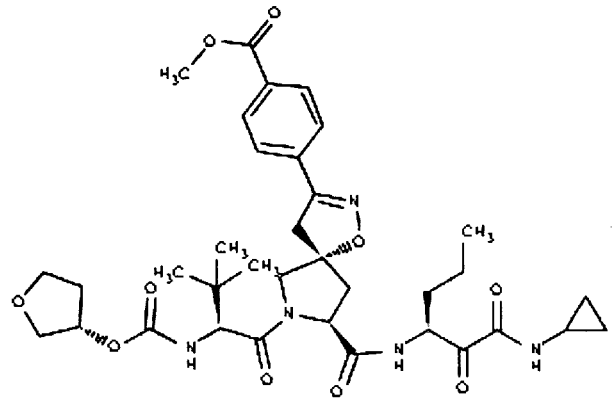
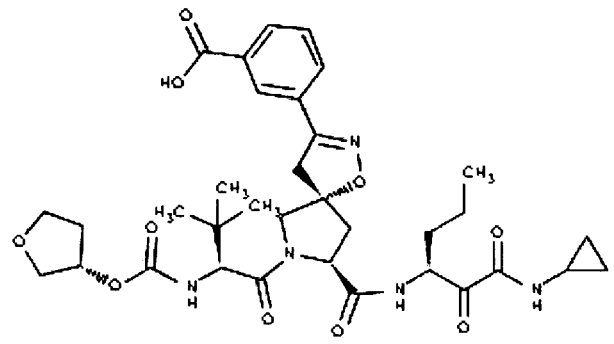
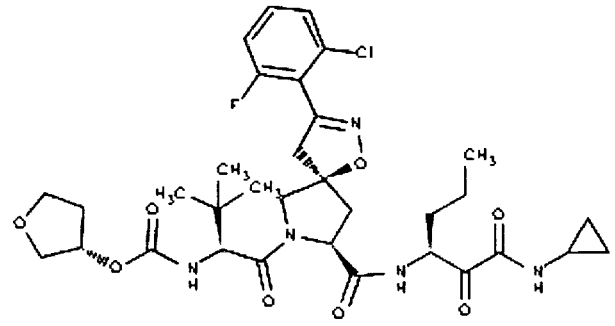
	465
	466
	467

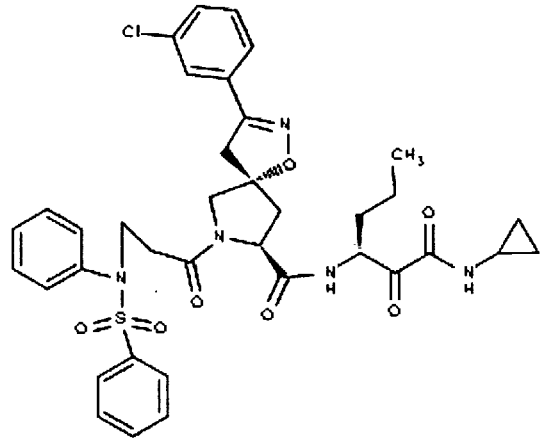
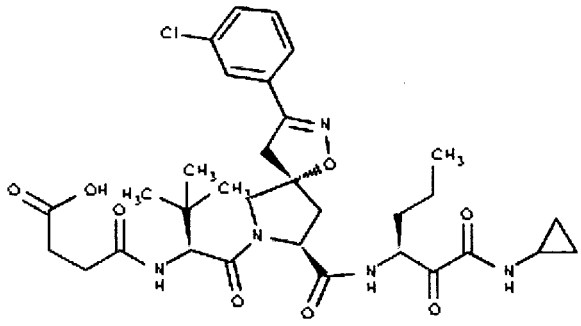
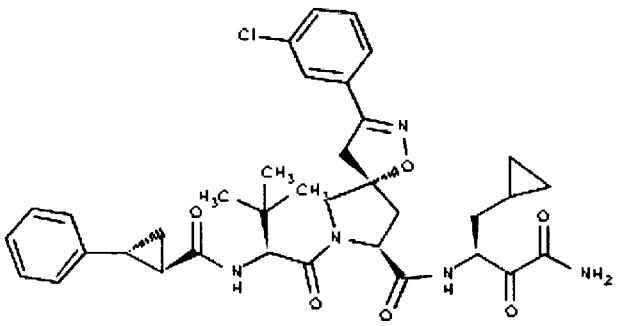
	468
	469
	470

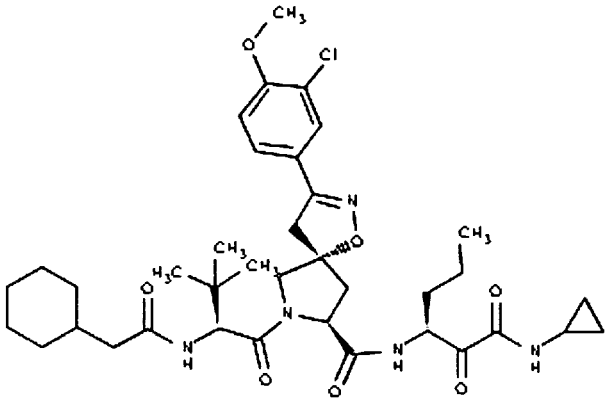
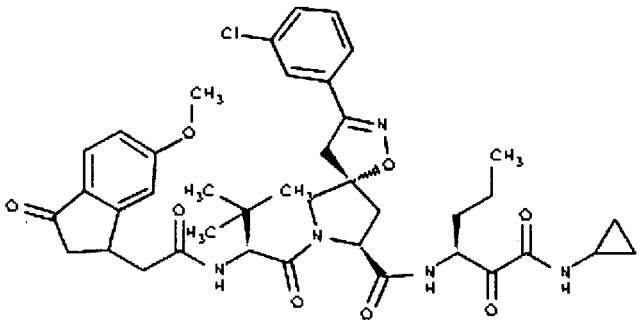
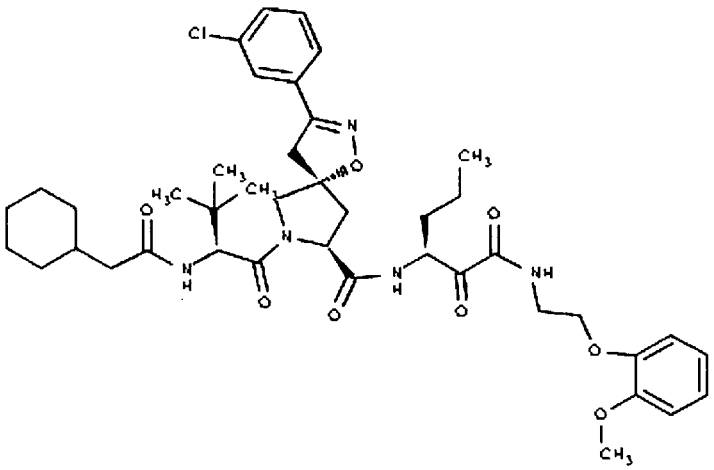
	471
	472
	473

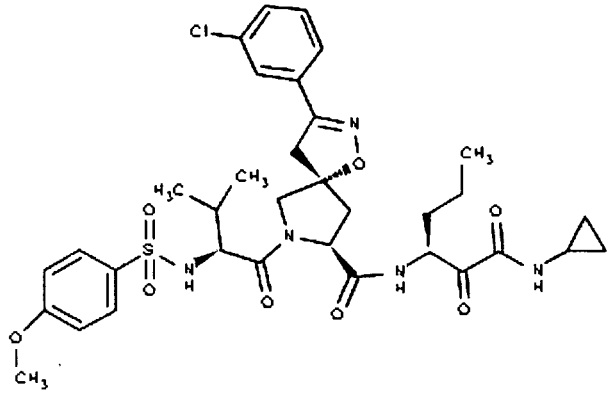
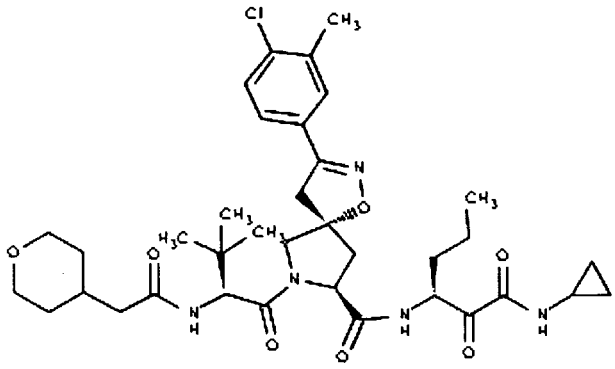
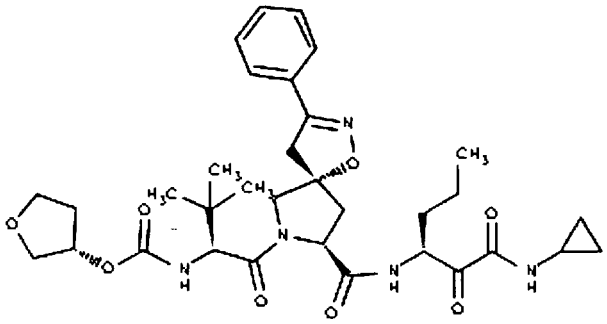
	474
	475
	476

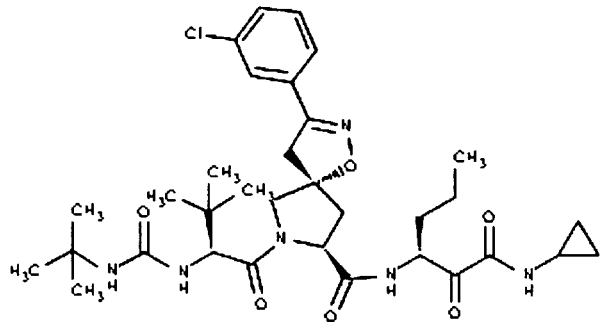
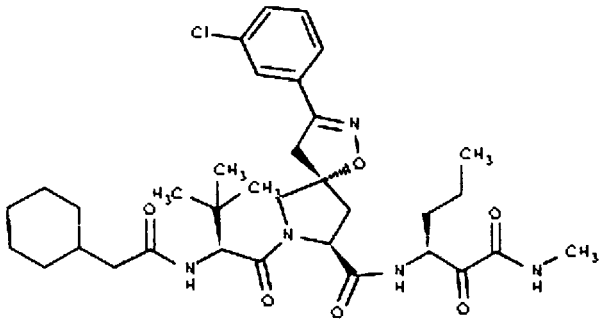
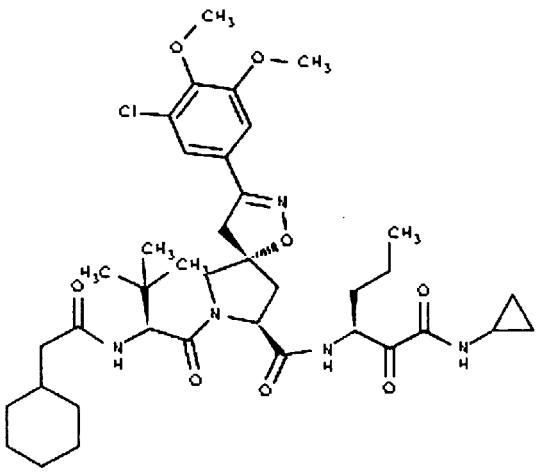
	477
	478
	479

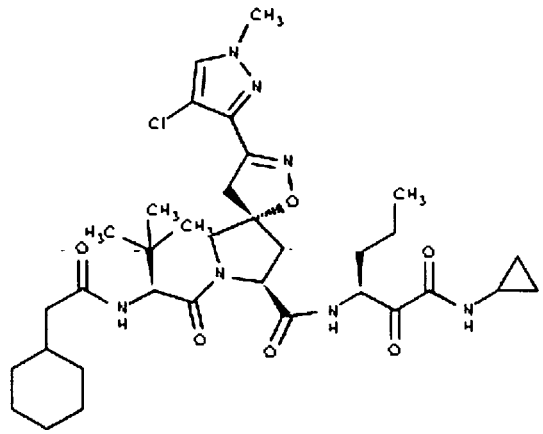
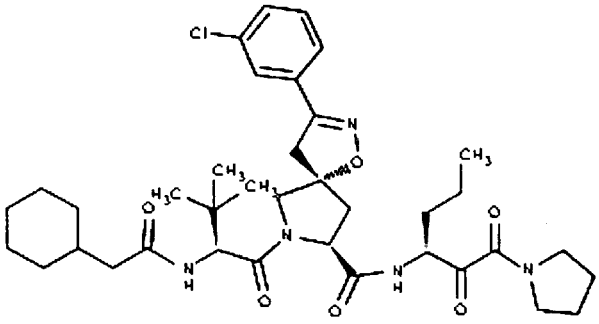
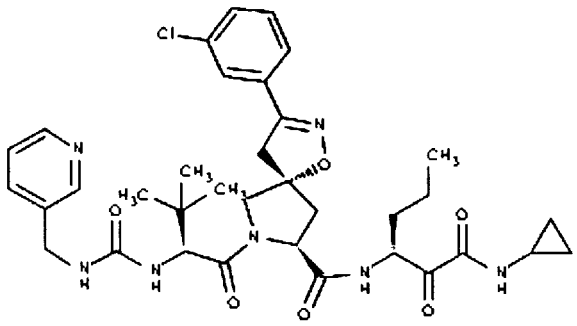
	480
	481
	482

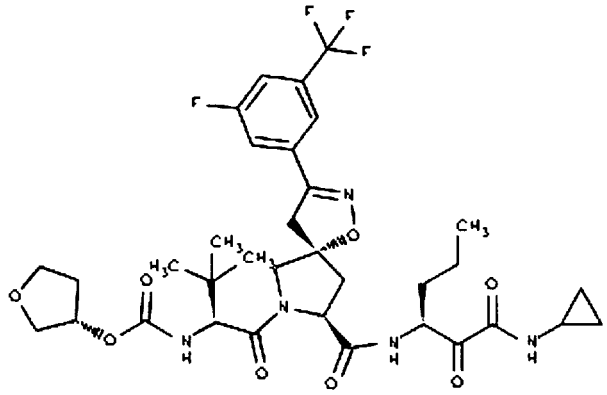
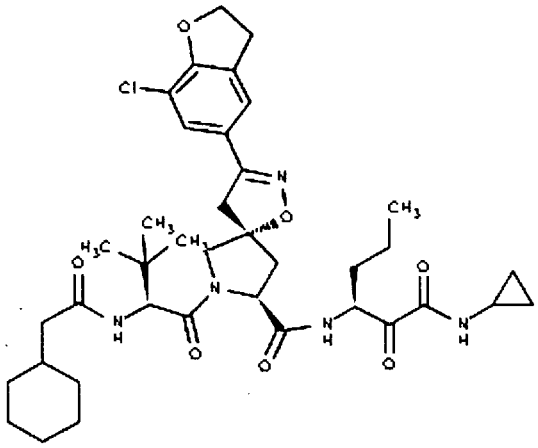
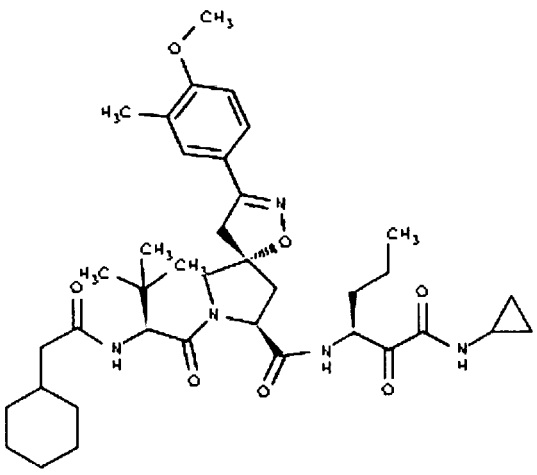
	483
	484
	485

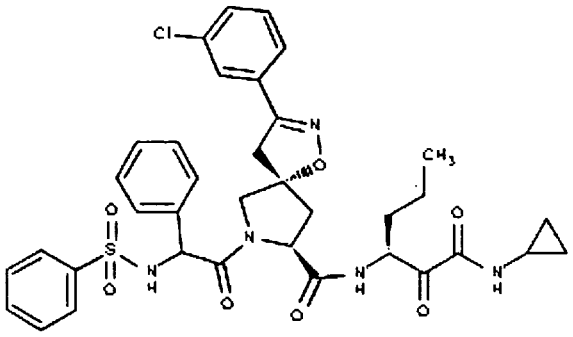
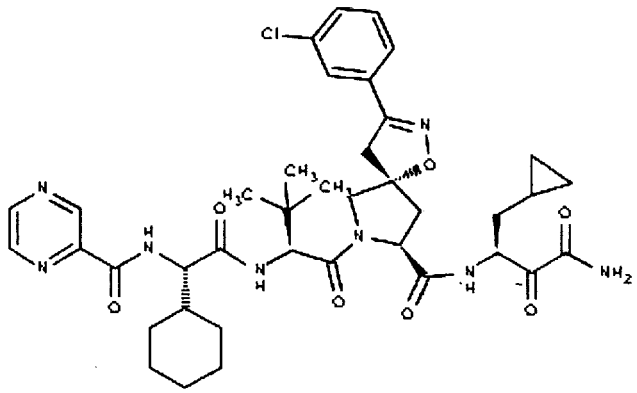
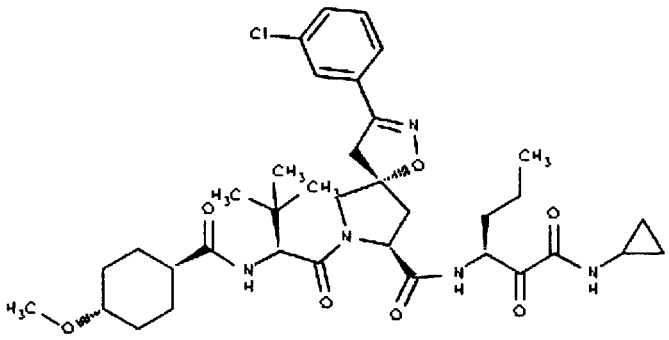
	486
	487
	488

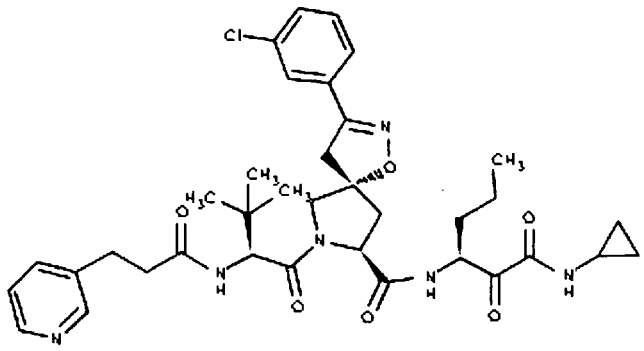
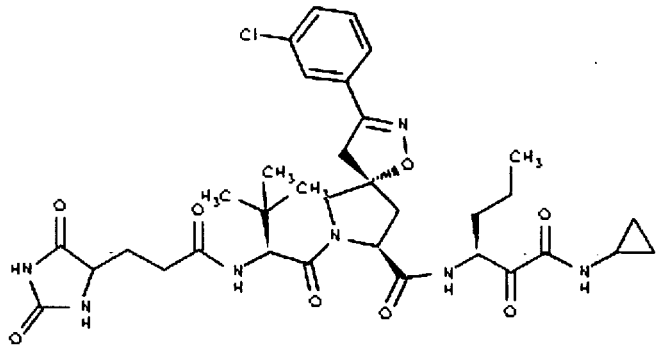
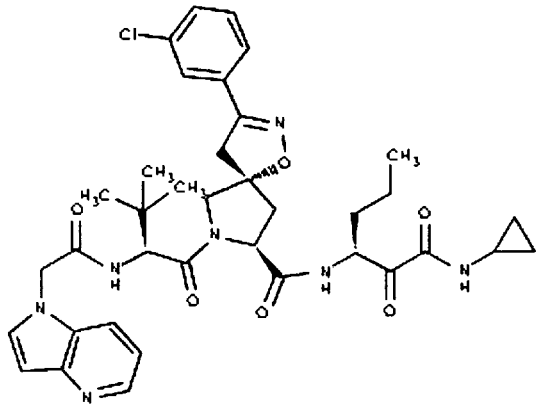
	489
	490
	491

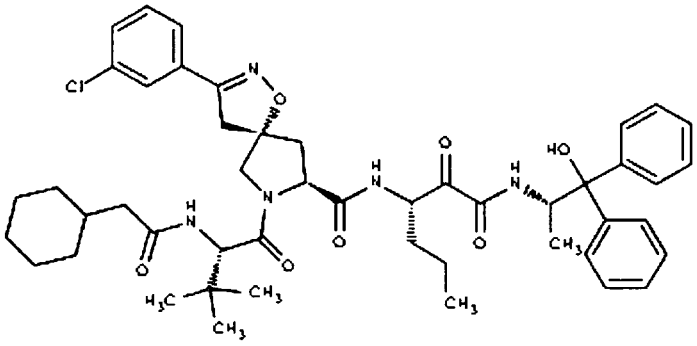
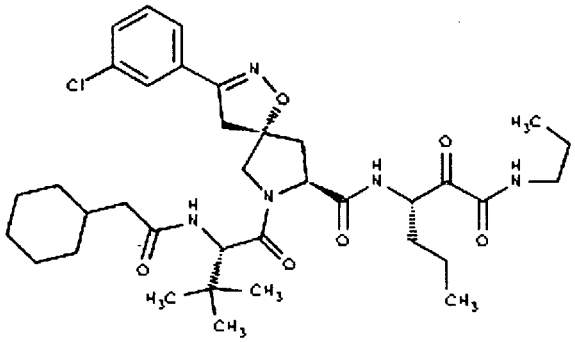
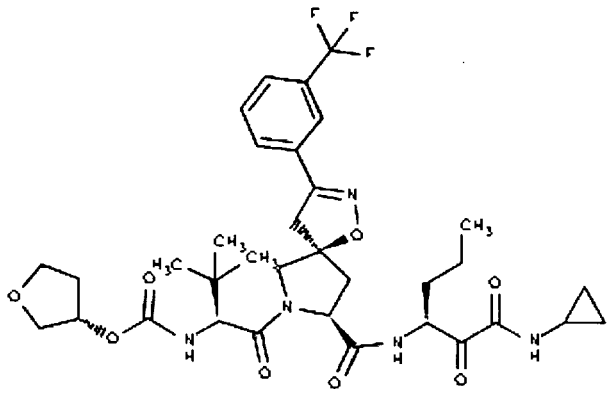
	492
	493
	494

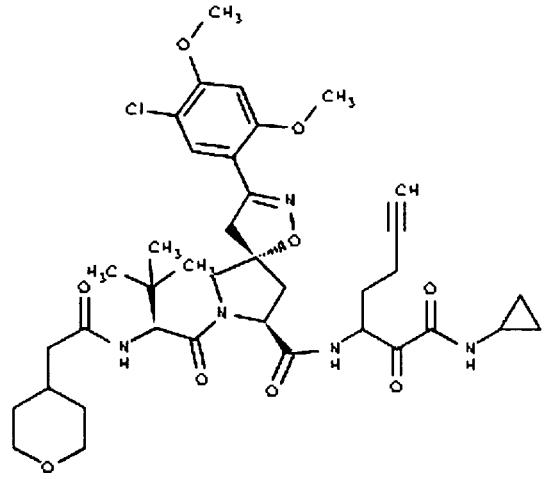
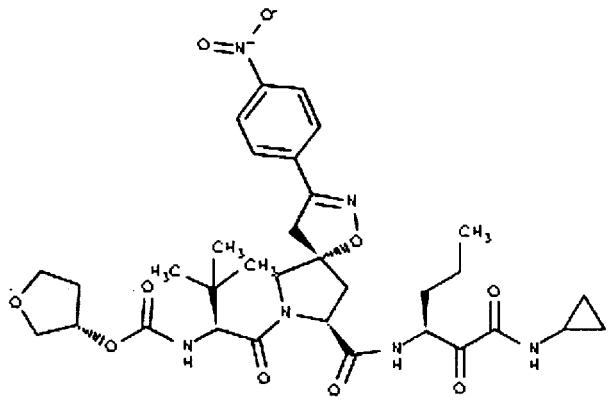
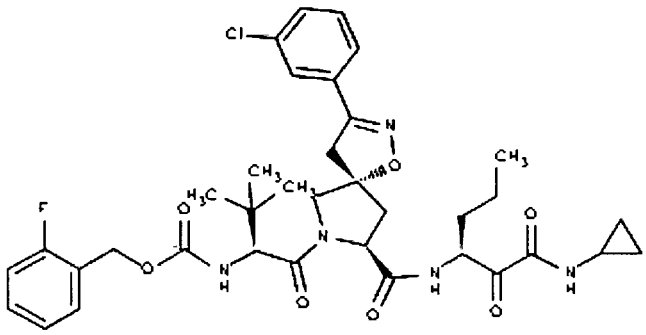
	495
	496
	497

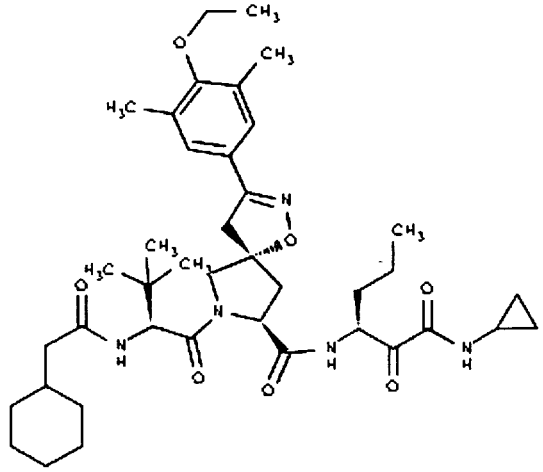
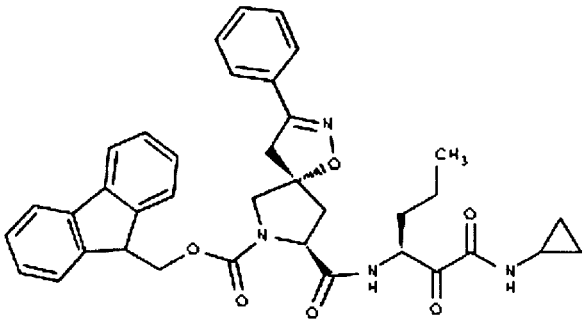
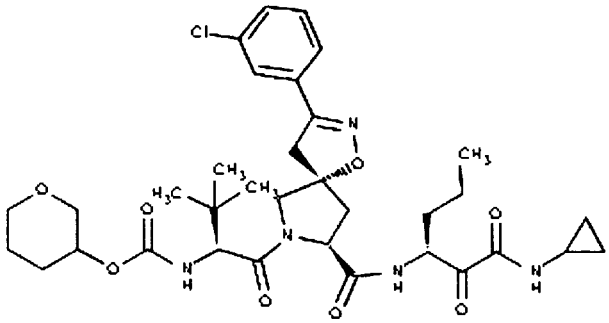
	498
	499
	500

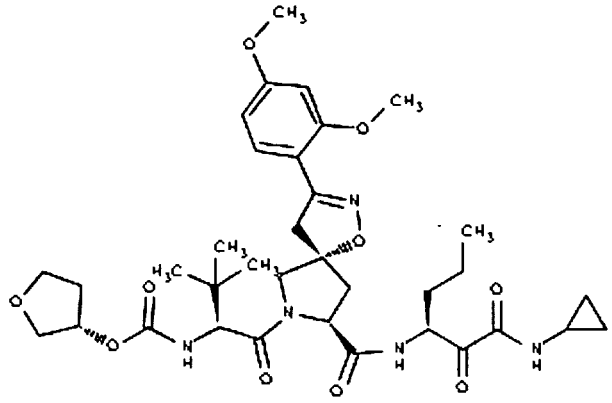
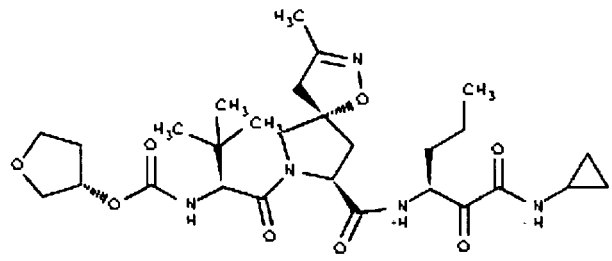
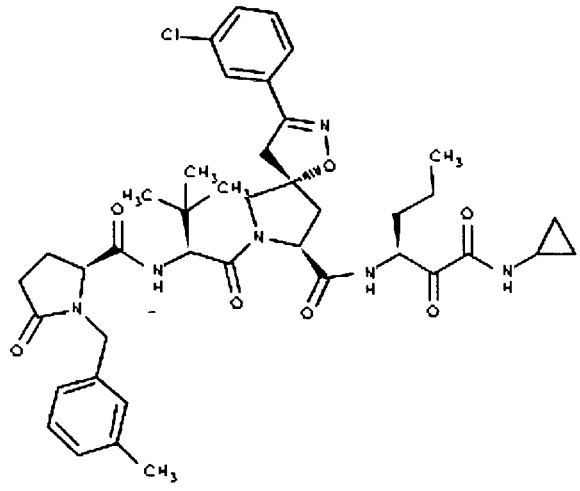
	501
	502
	503

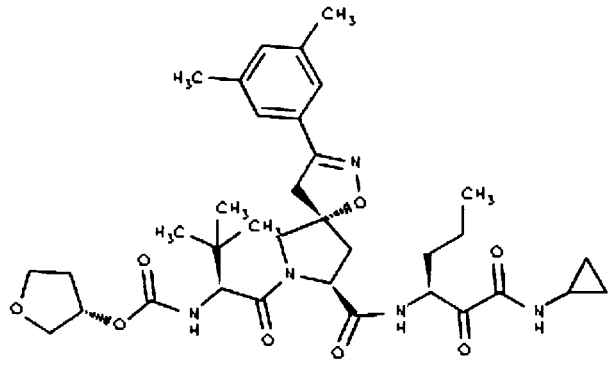
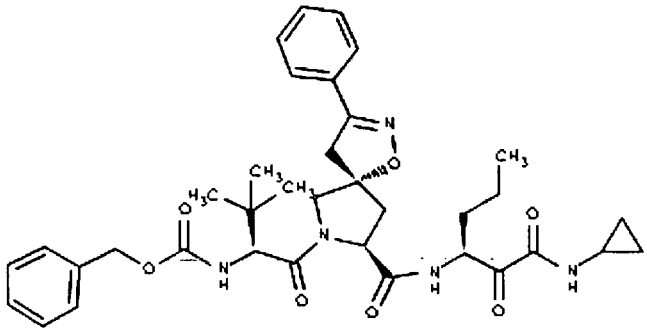
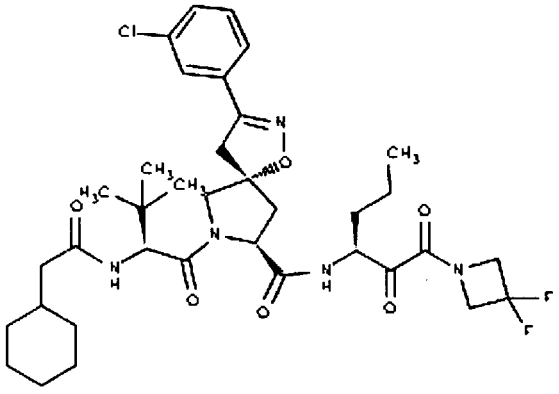
	504
	505
	506

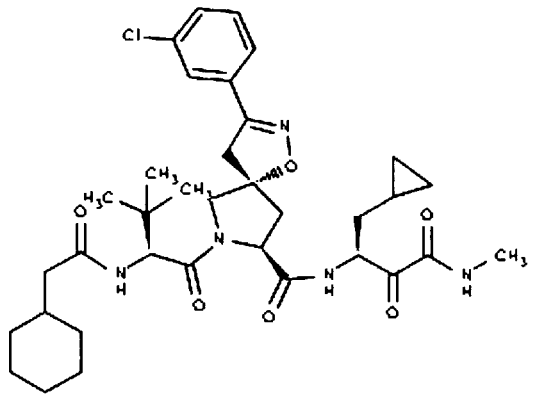
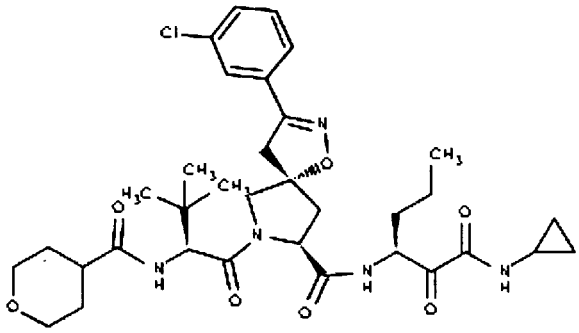
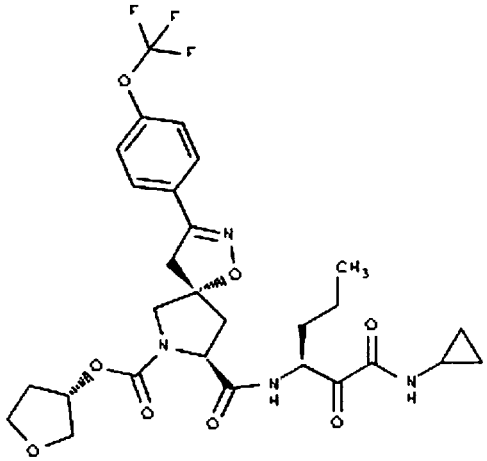
	507
	508
	509

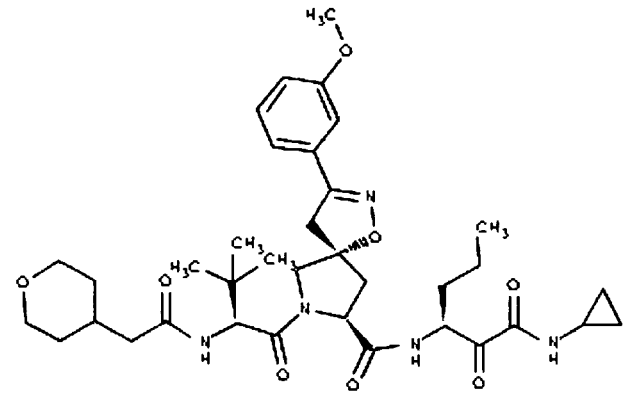
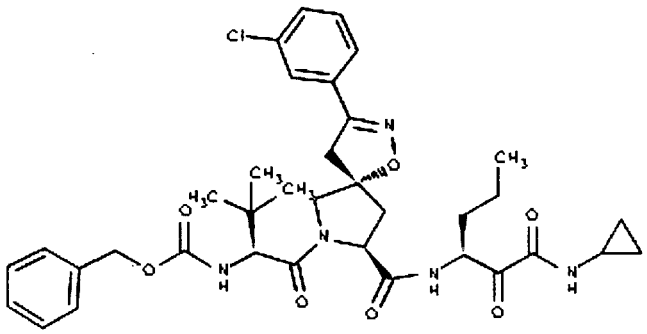
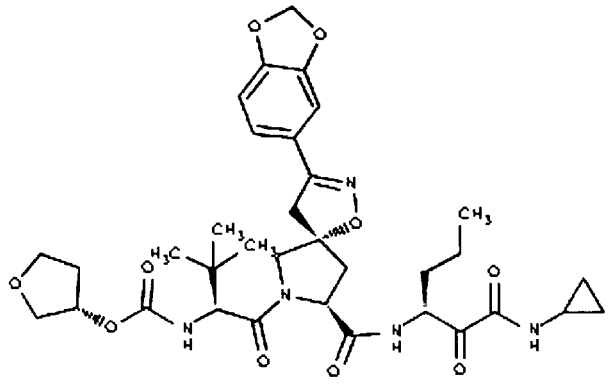
	510
	511
	512

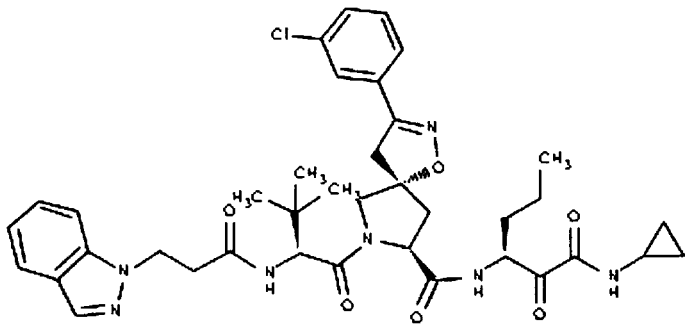
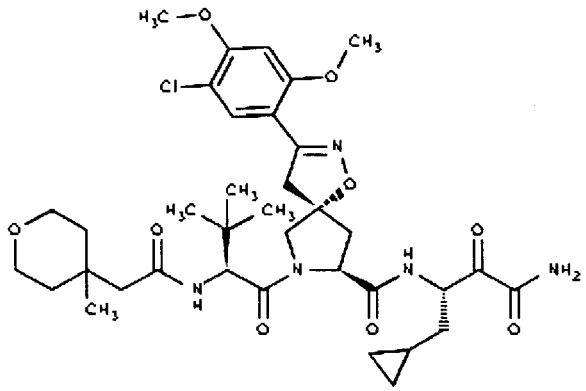
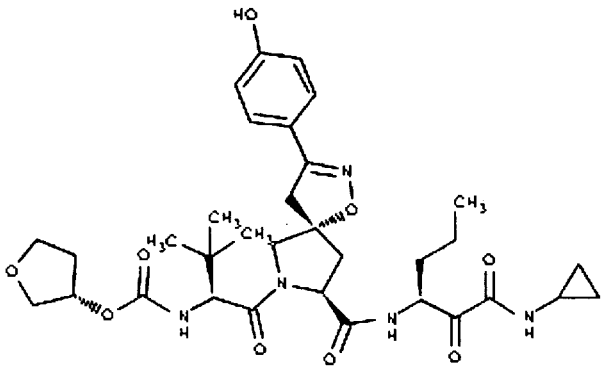
	513
	514
	515

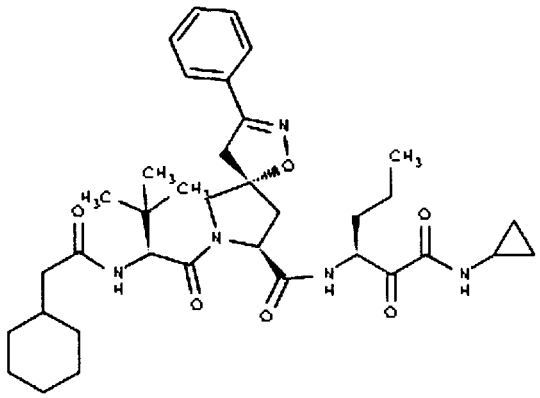
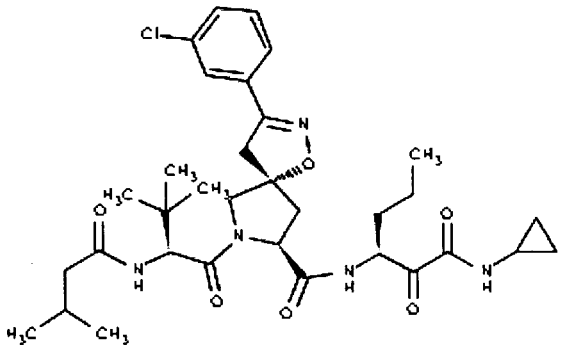
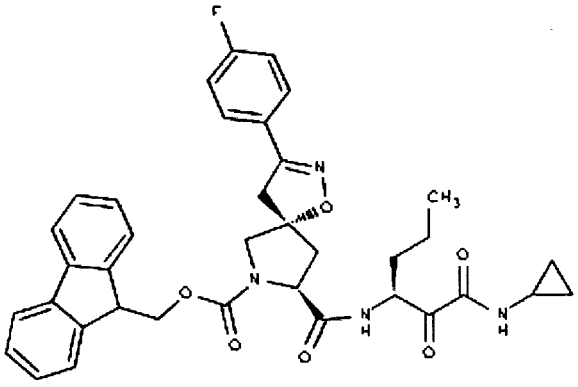
	516
	517
	518

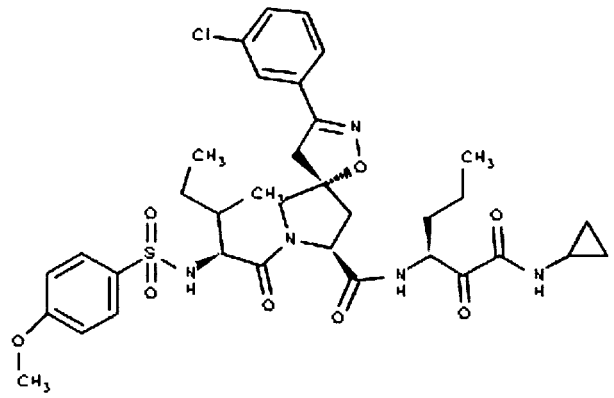
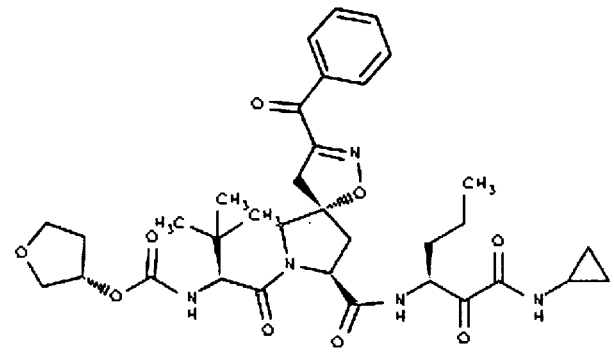
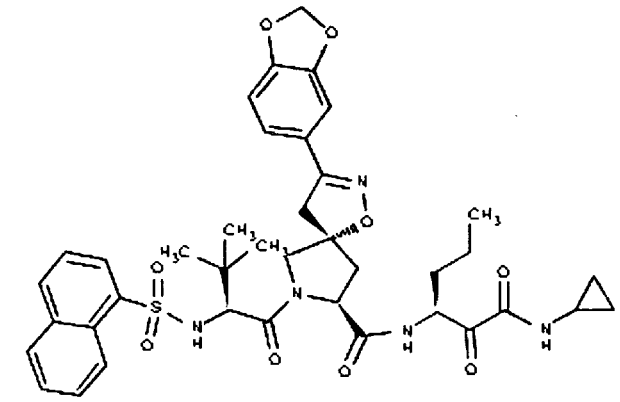
	519
	520
	521

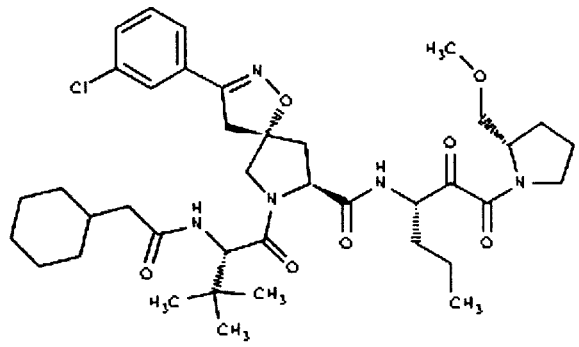
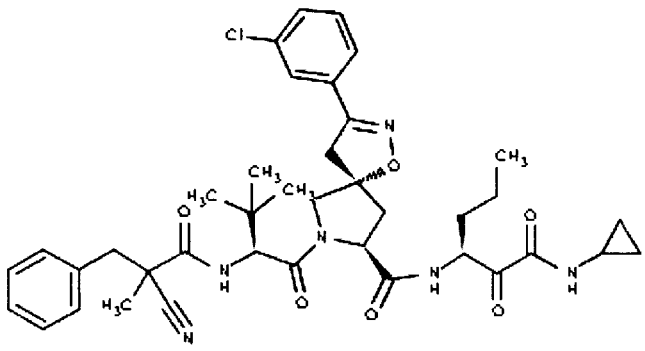
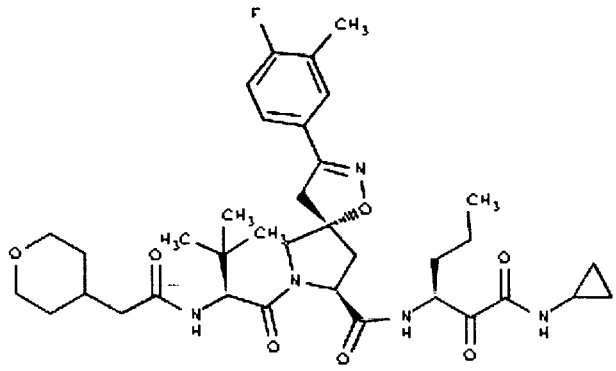
	522
	523
	524

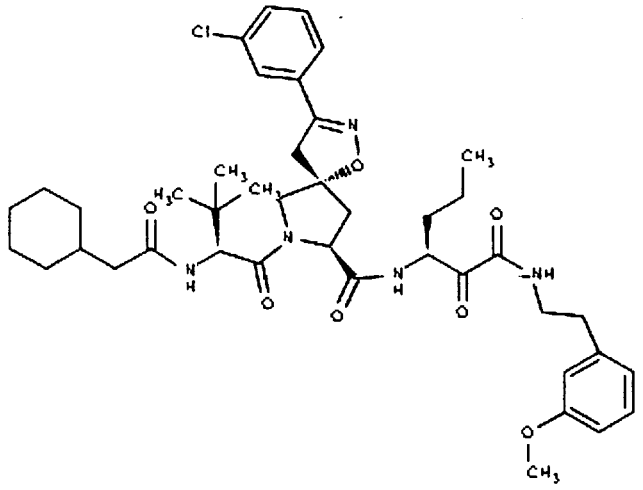
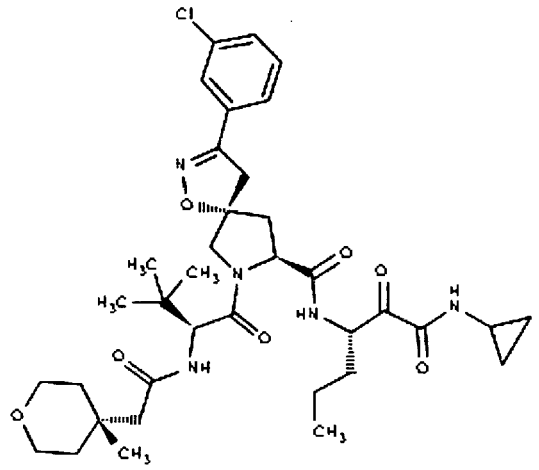
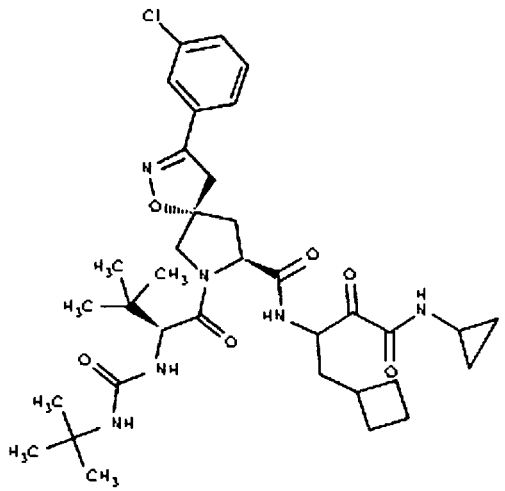
	525
	526
	527

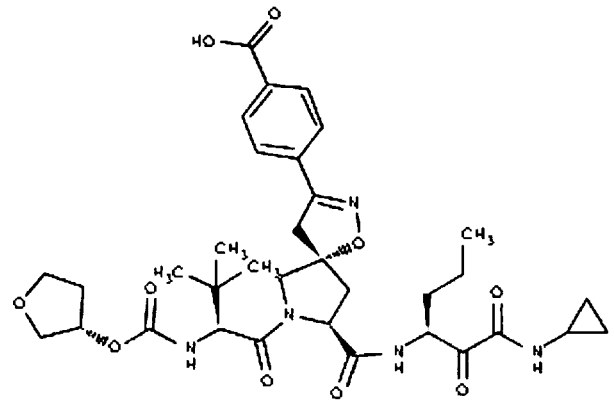
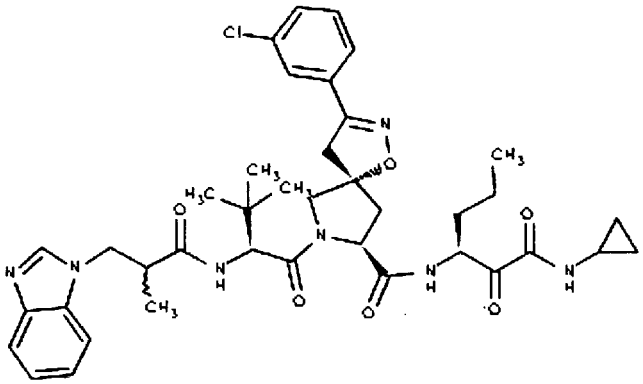
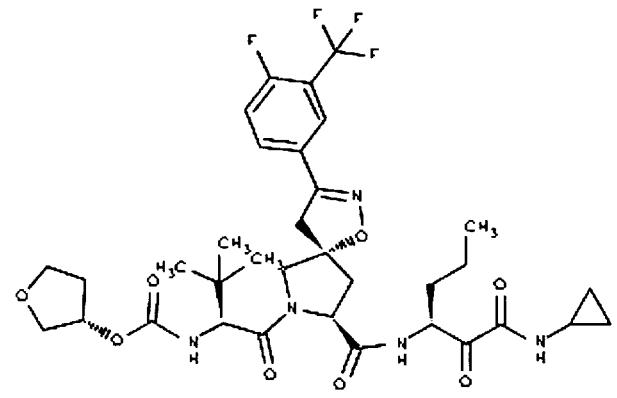
	528
	529
	530

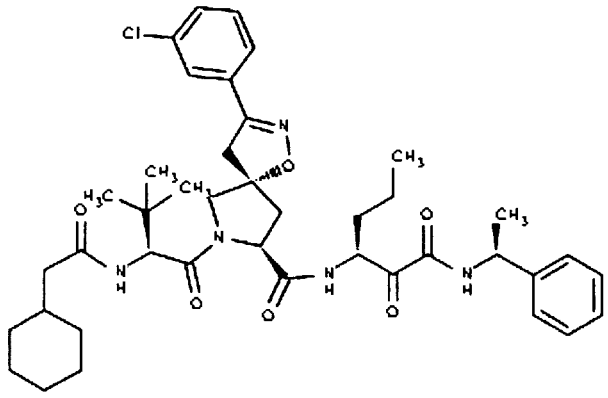
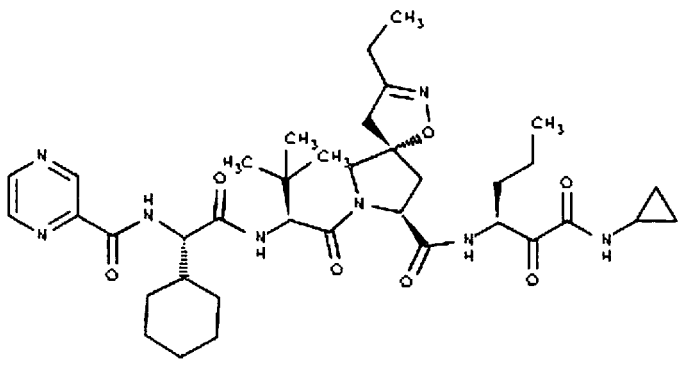
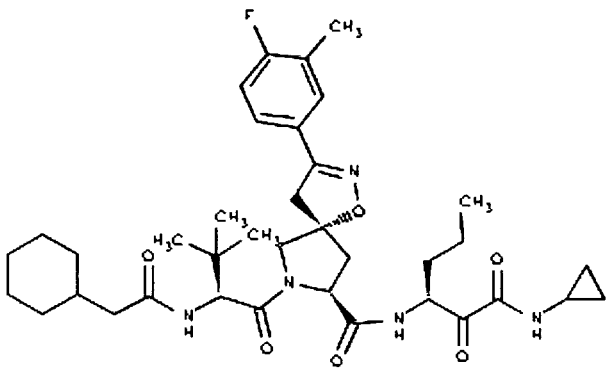
	531
	532
	533

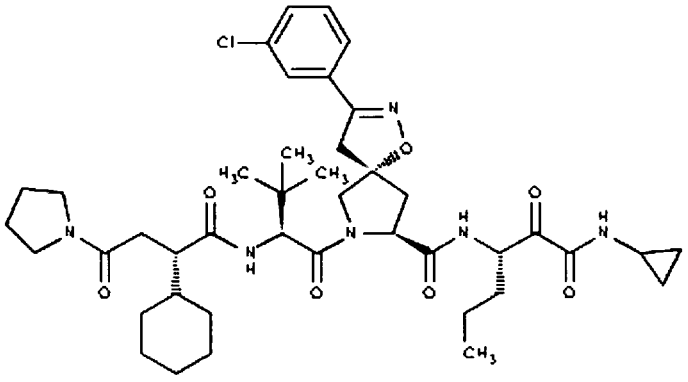
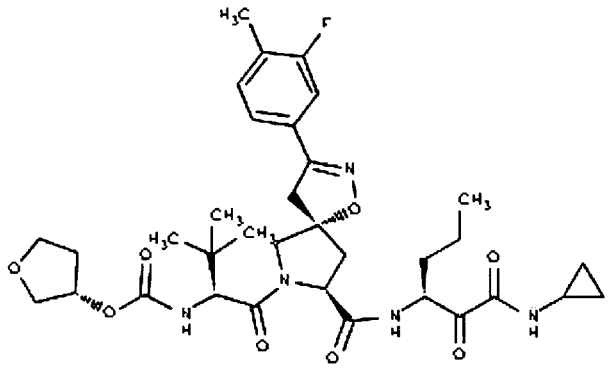
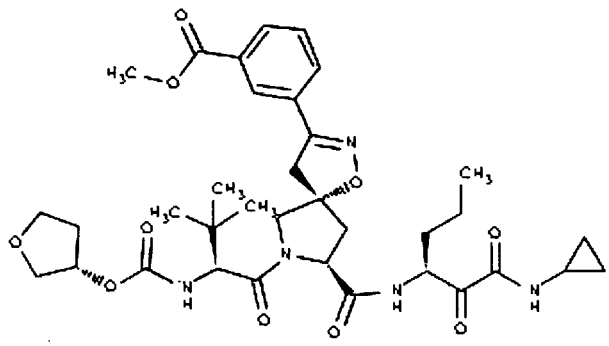
	534
	535
	536

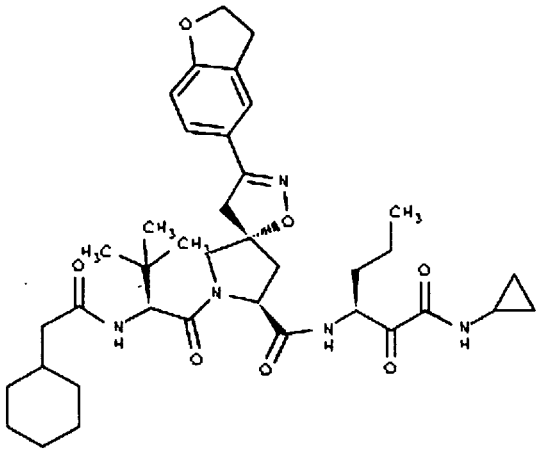
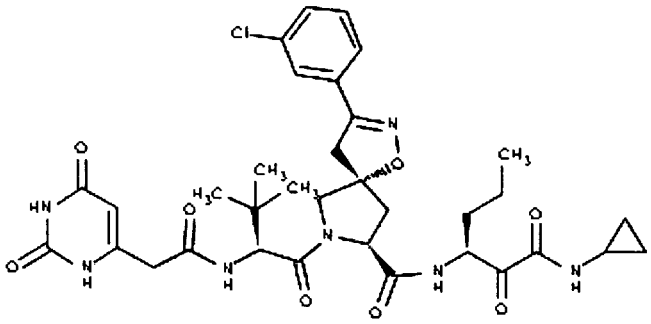
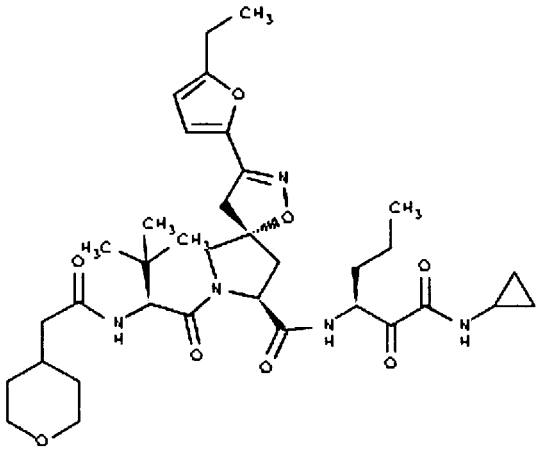
	537
	538
	539

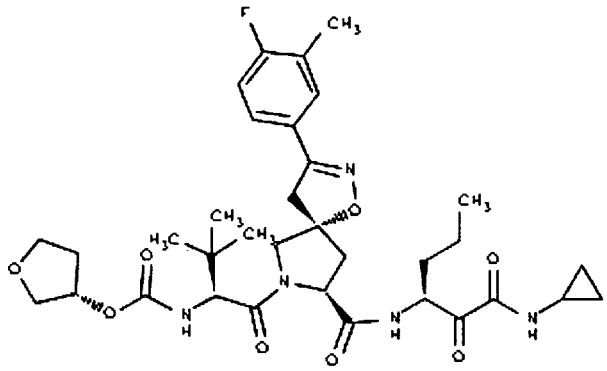
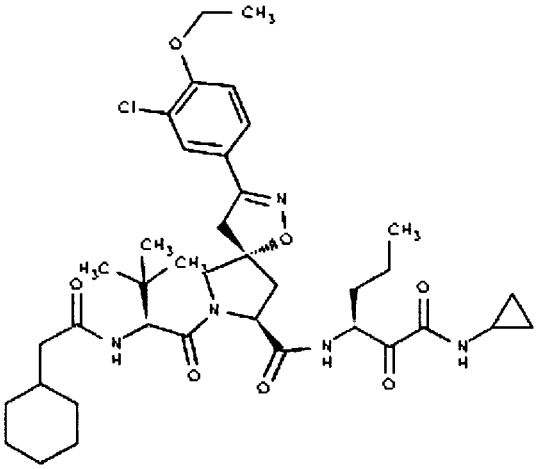
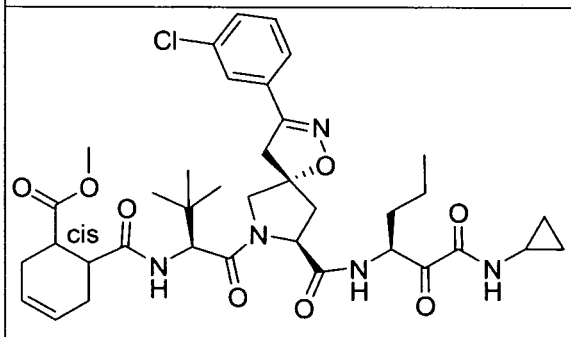
	540
	541
	542

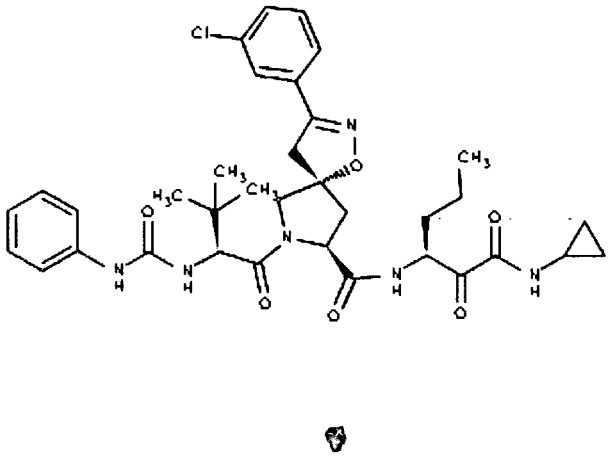
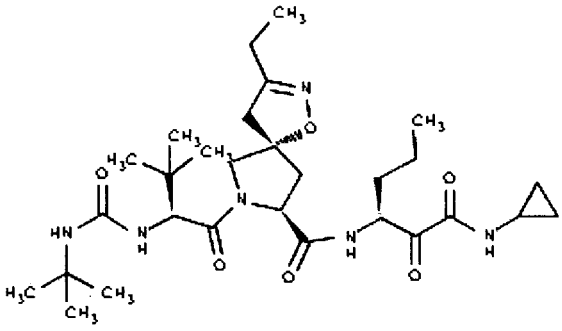
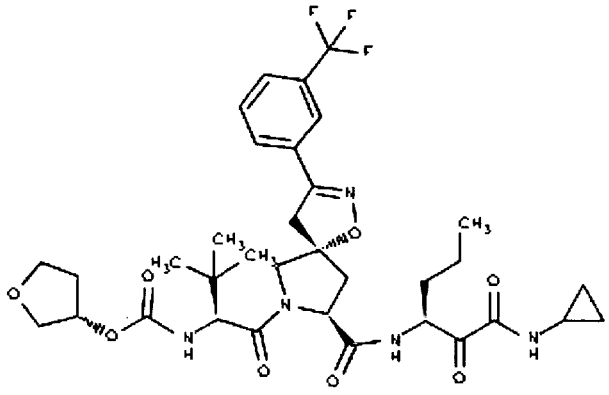
	543
	544
	545

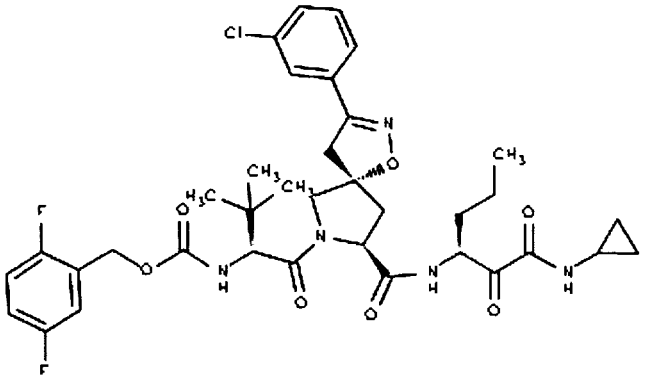
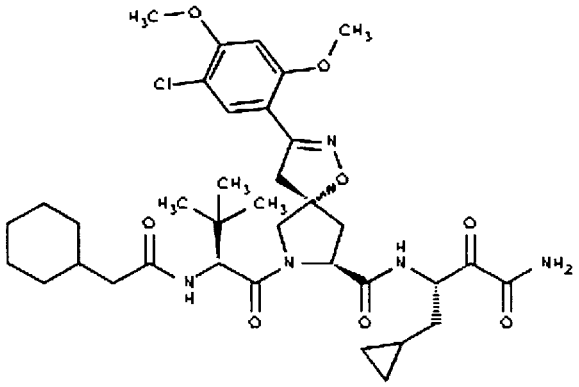
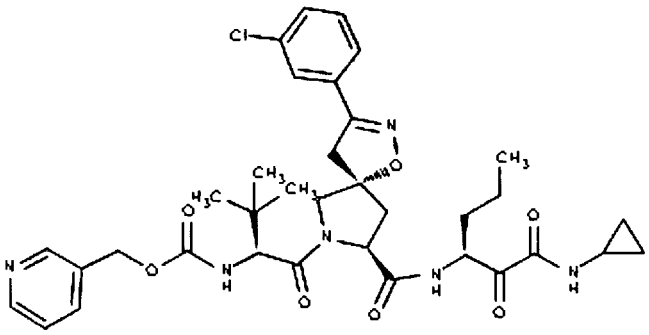
	546
	547
	548

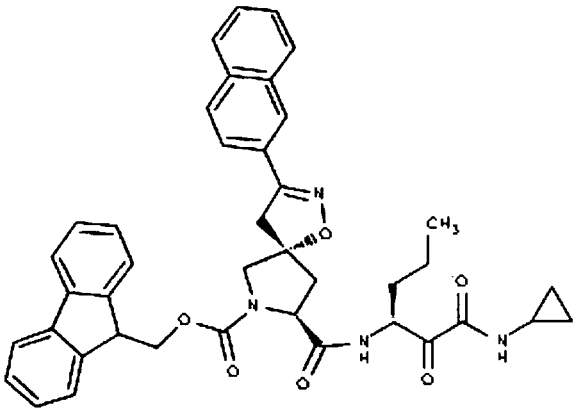
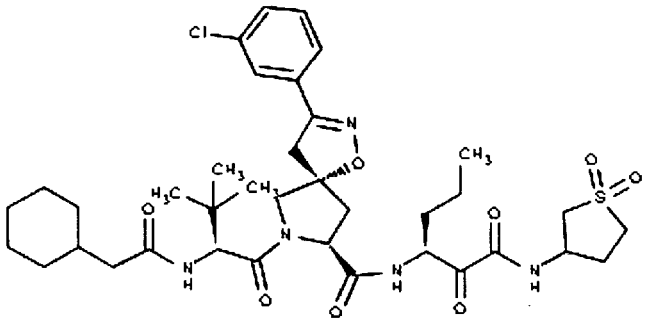
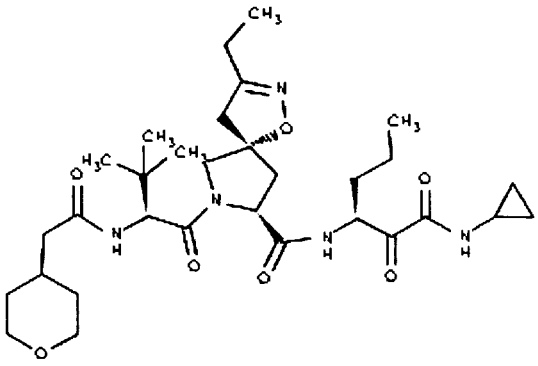
	549
	550
	551

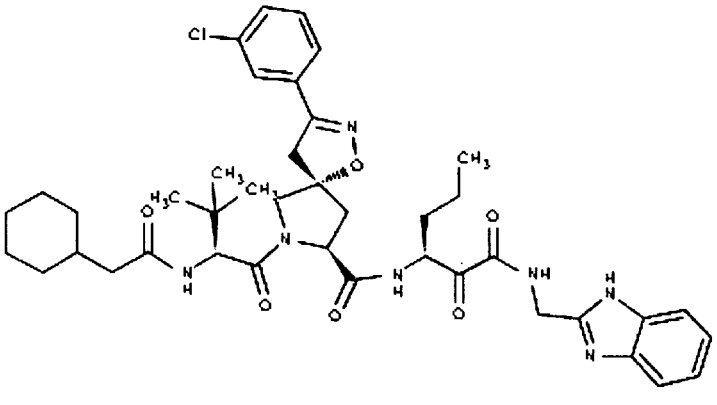
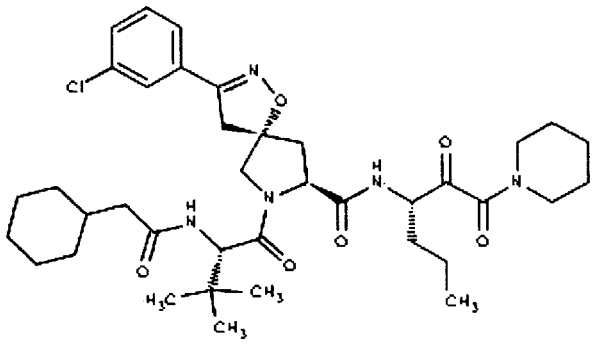
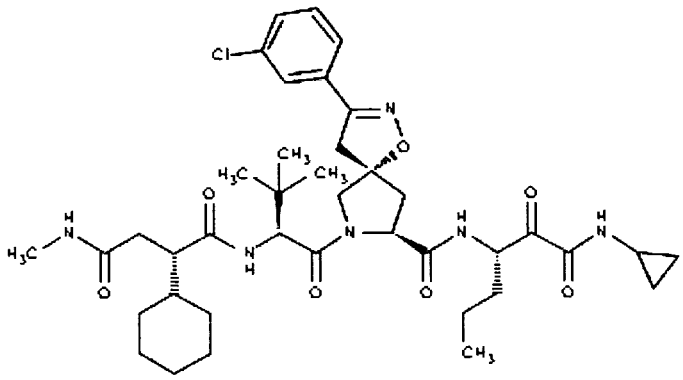
	552
	553
	554

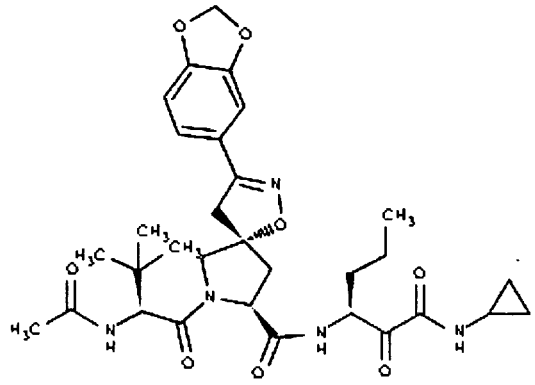
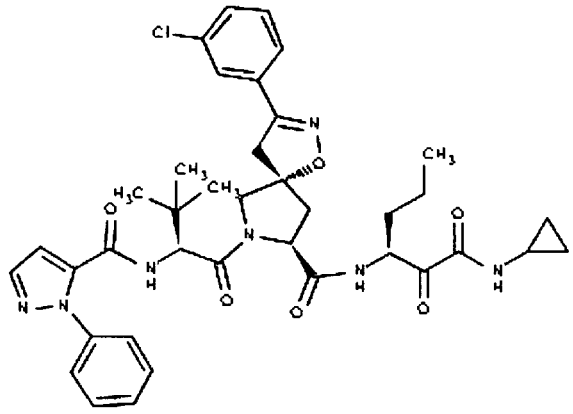
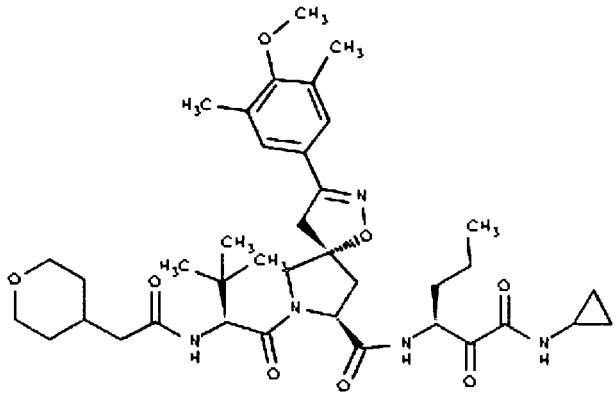
	555
	556
	557

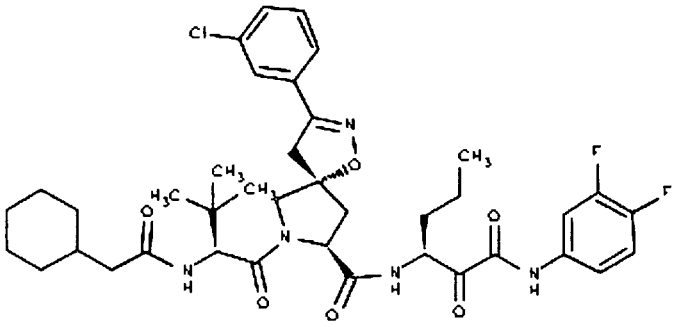
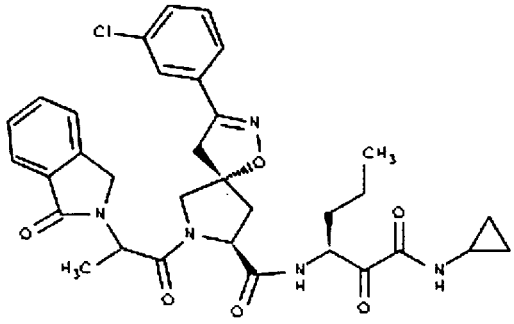
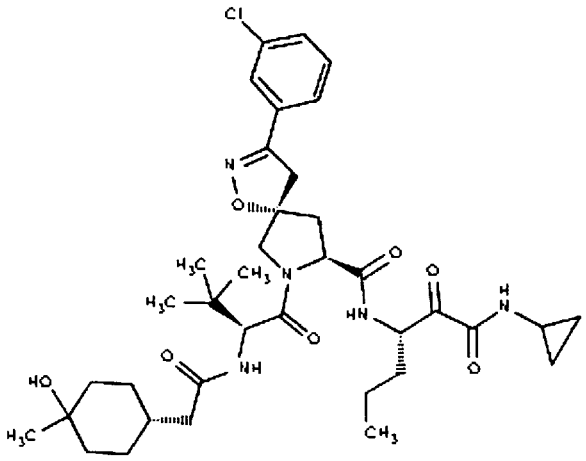
	558
	559
	560

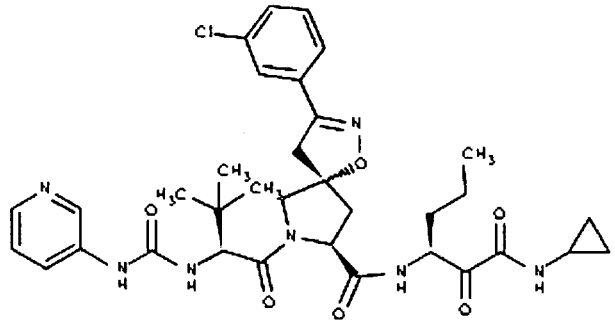
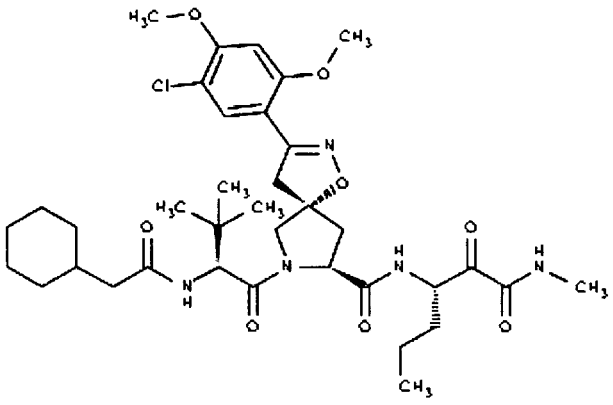
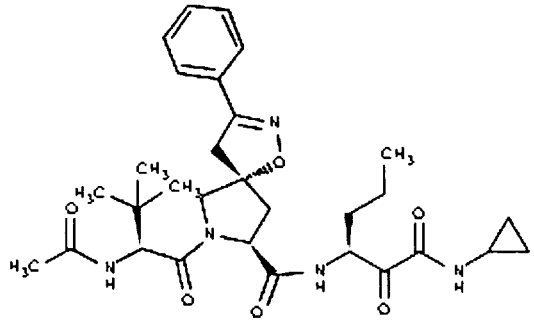
	561
	562
	563

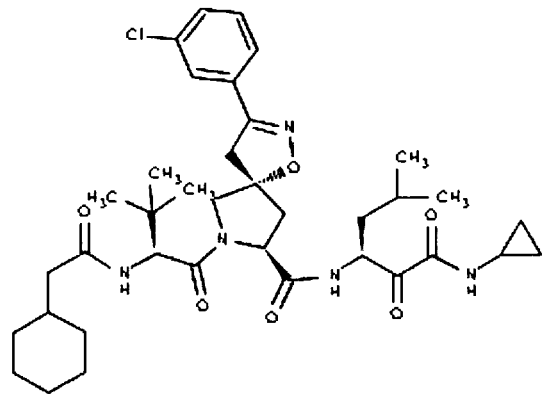
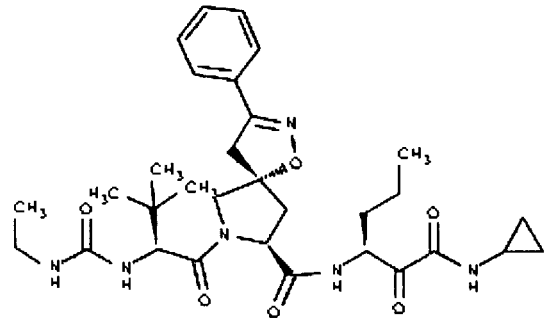
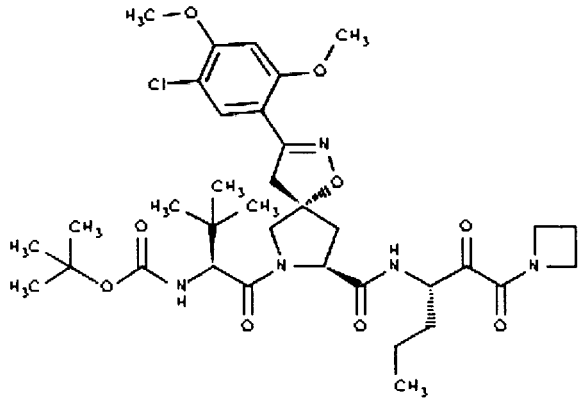
	564
	565
	566

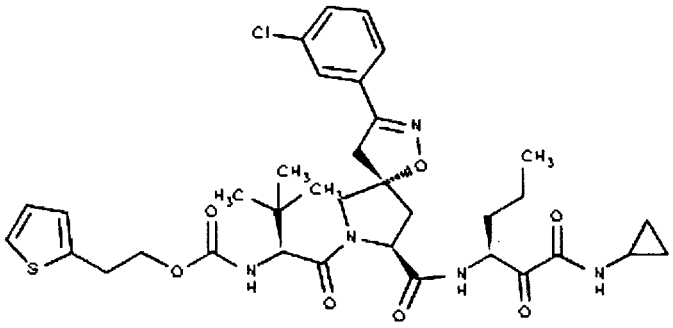
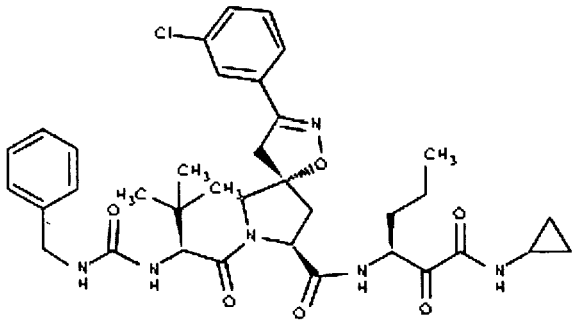
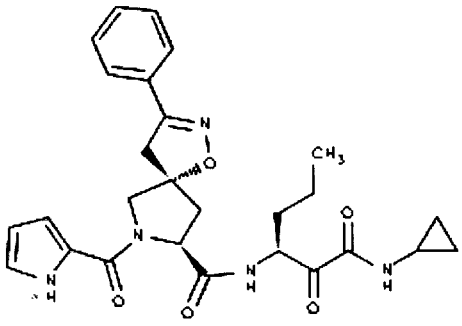
	567
	568
	569

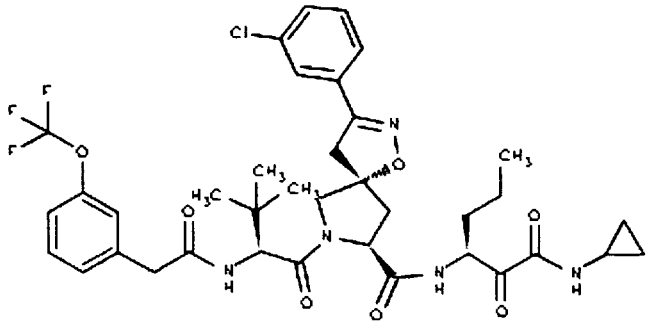
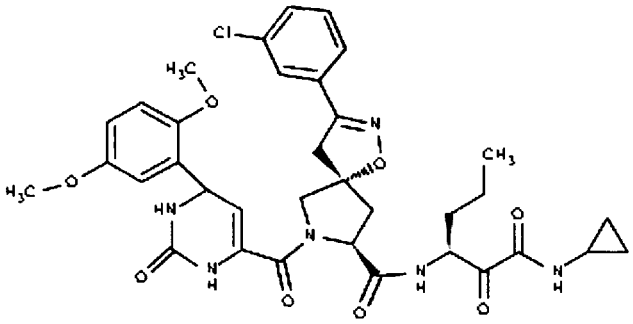
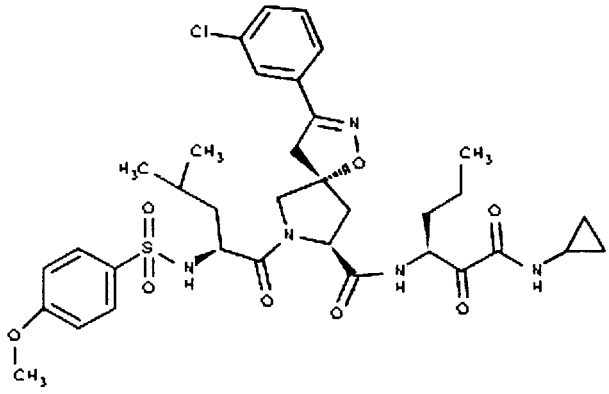
	570
	571
	572

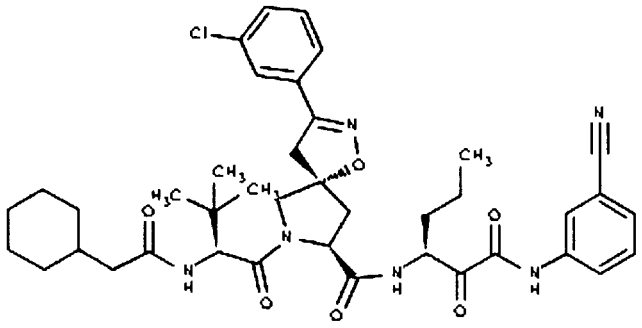
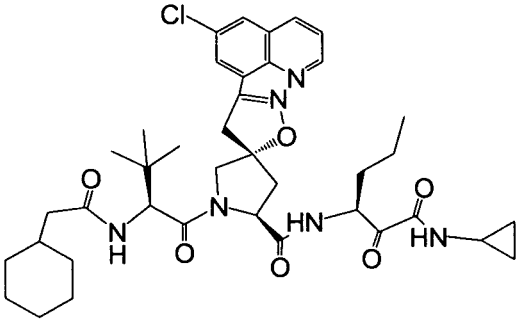
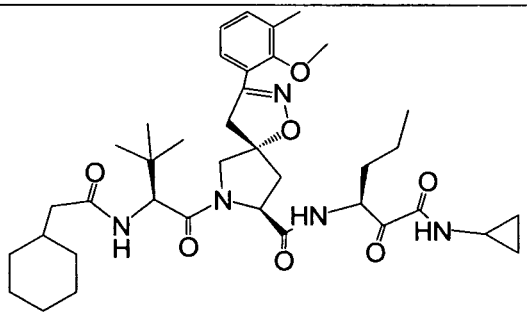
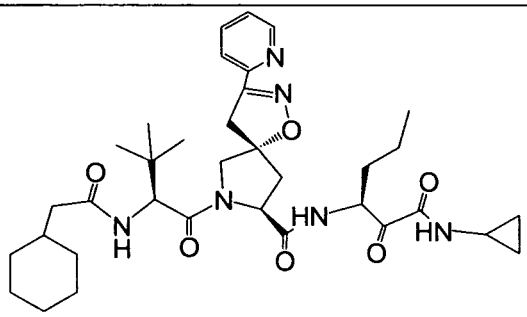
	573
	574
	575

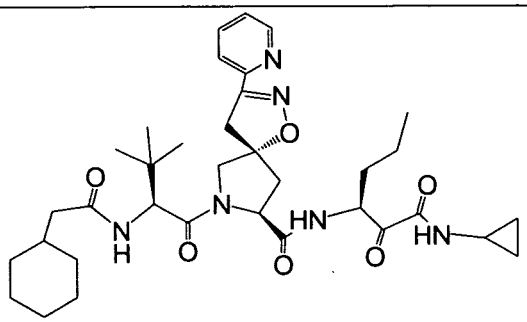
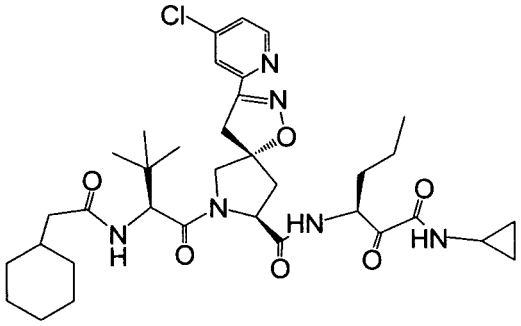
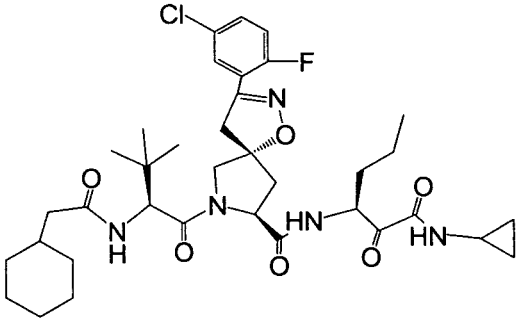
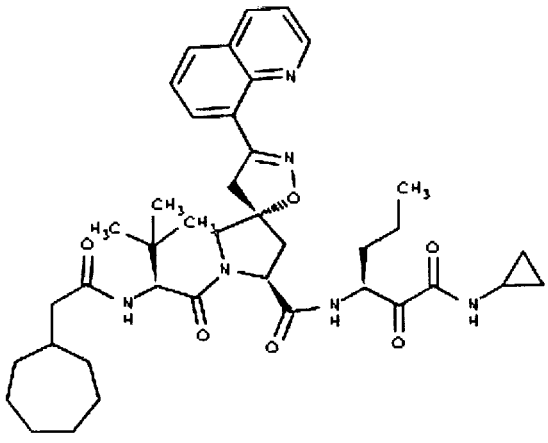
	576
	577
	578

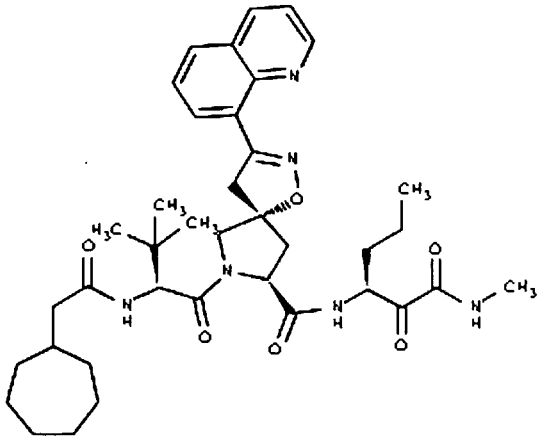
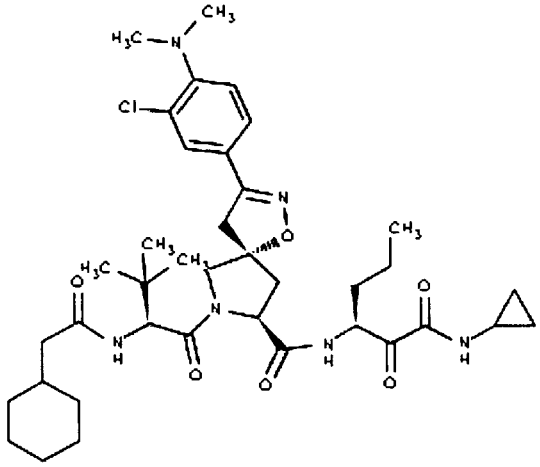
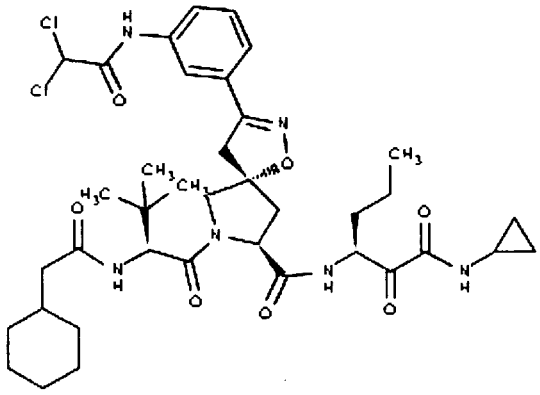
	579
	580
	581

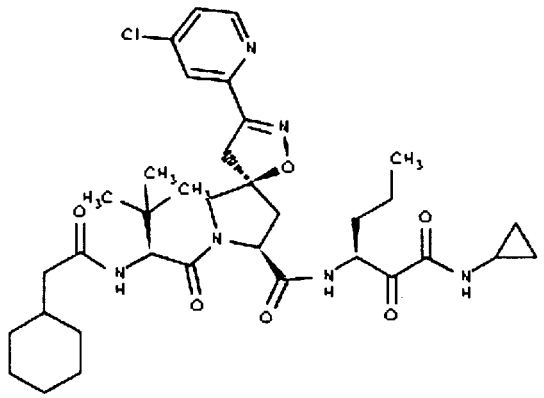
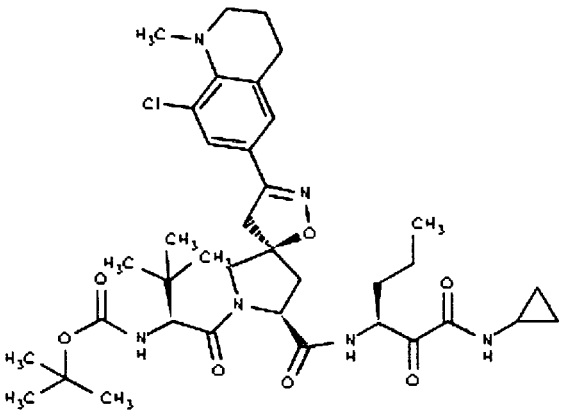
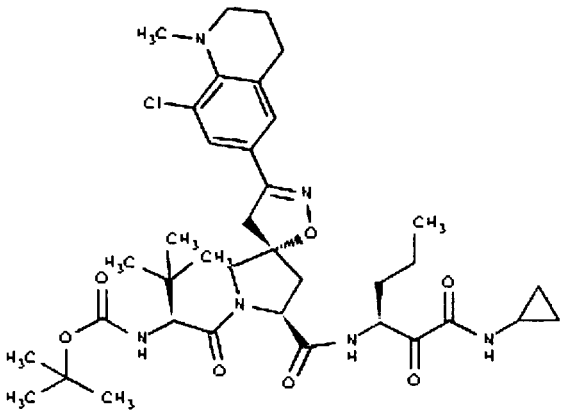
	582
	583
	584

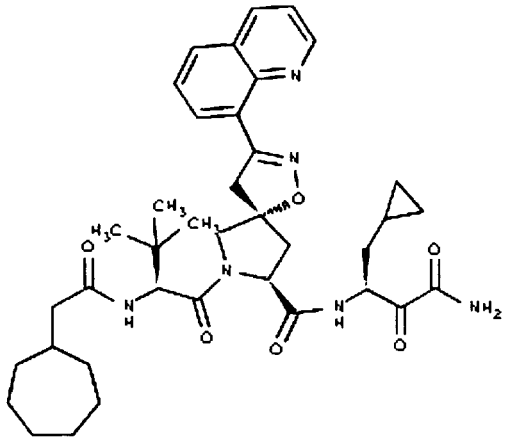
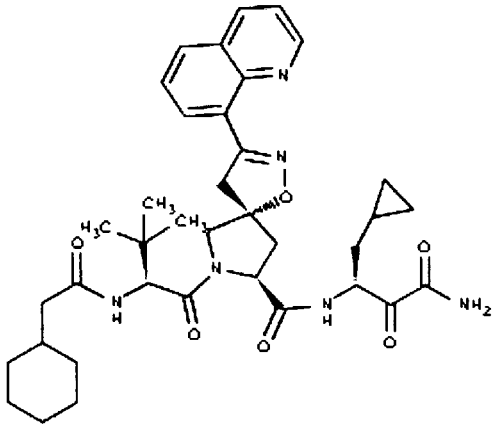
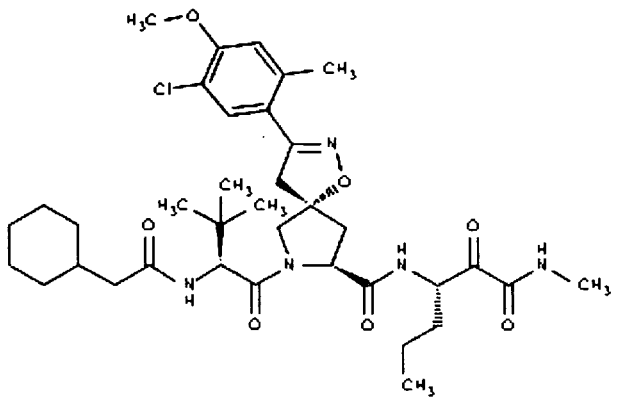
	585
	586
	587

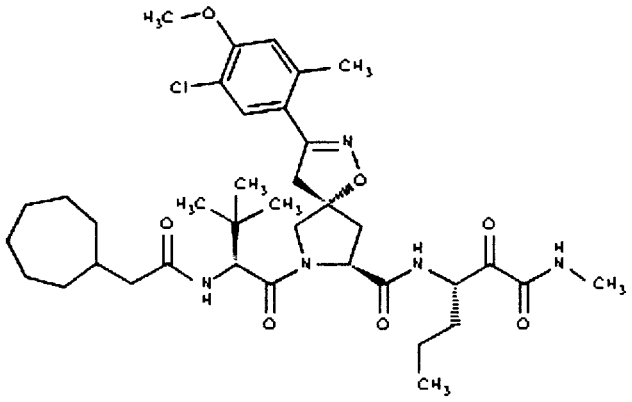
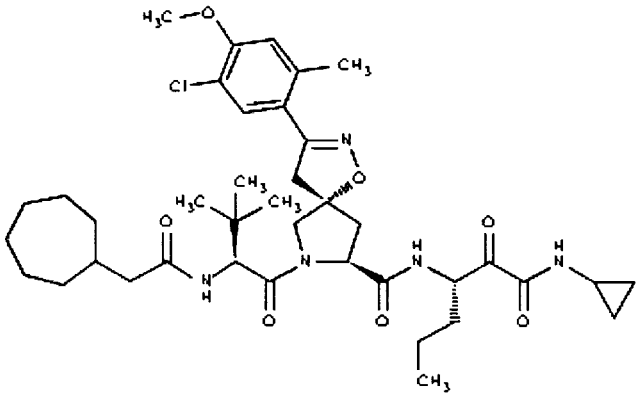
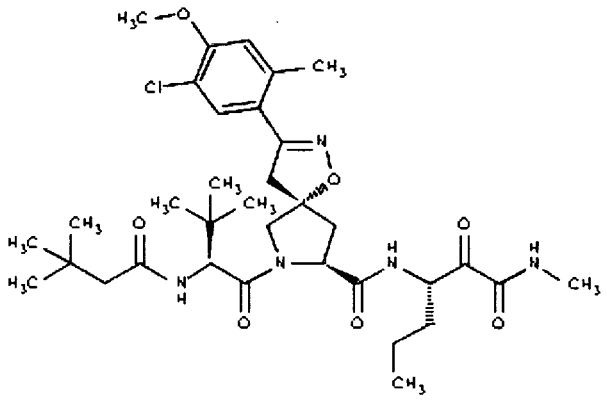
	588
	589
	590
	591

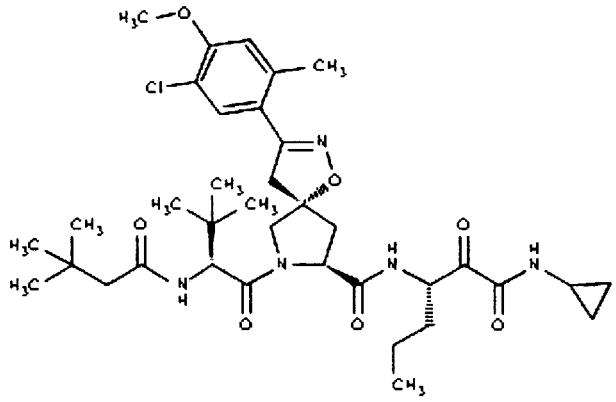
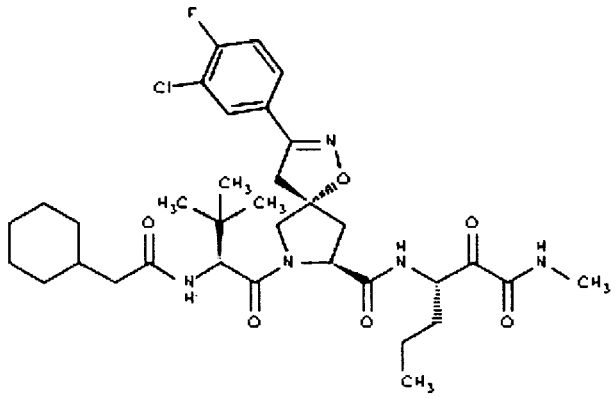
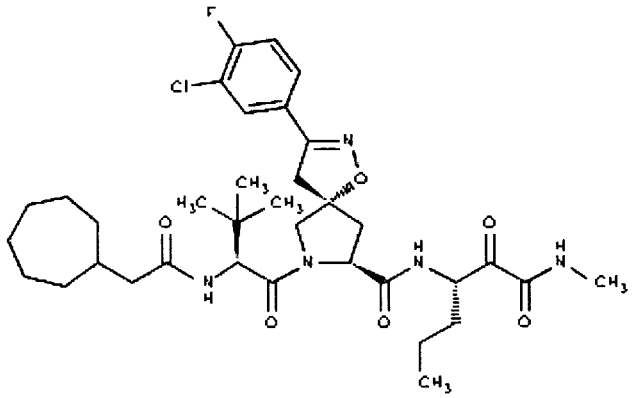
	592
	593
	594
	595

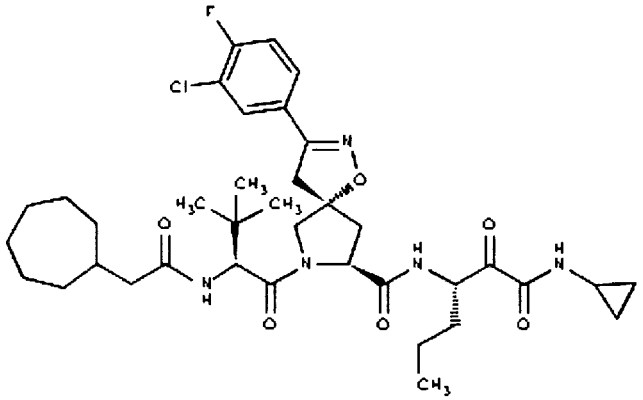
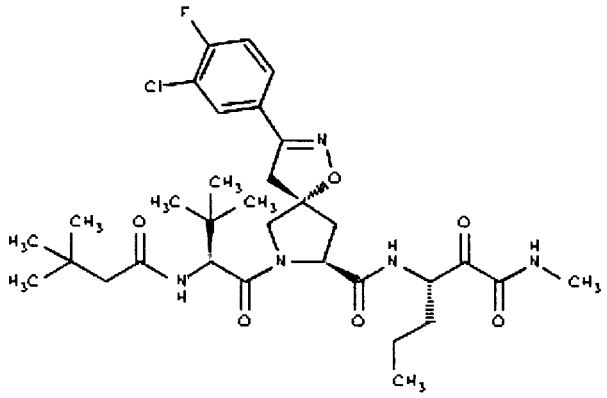
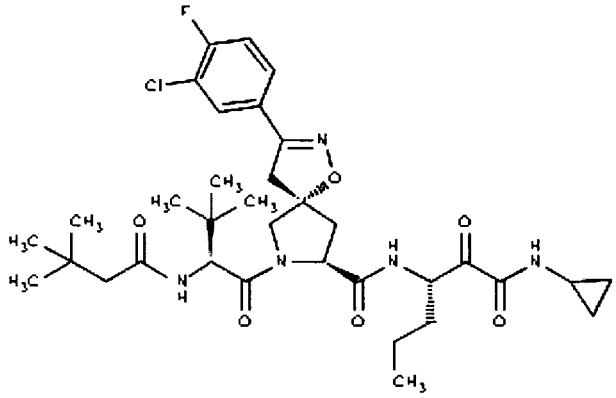
	596
	597
	598

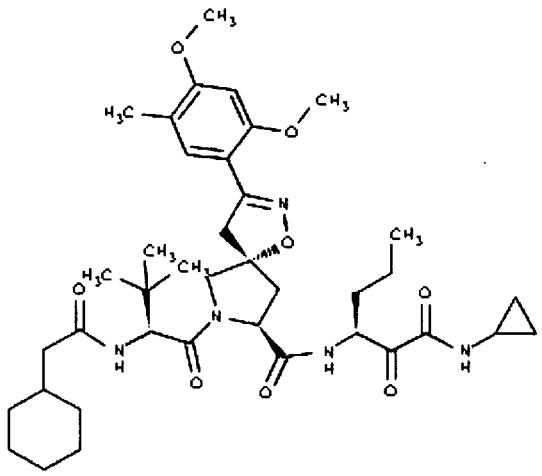
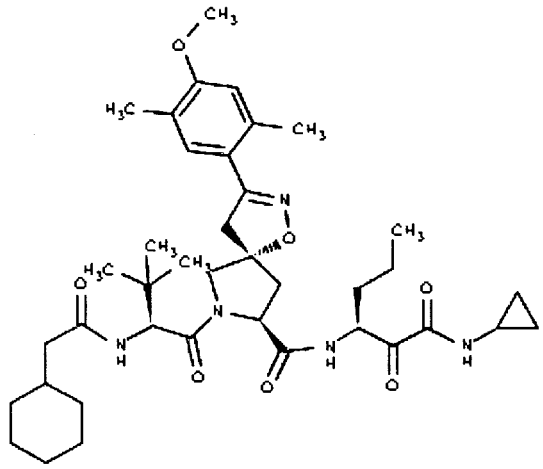
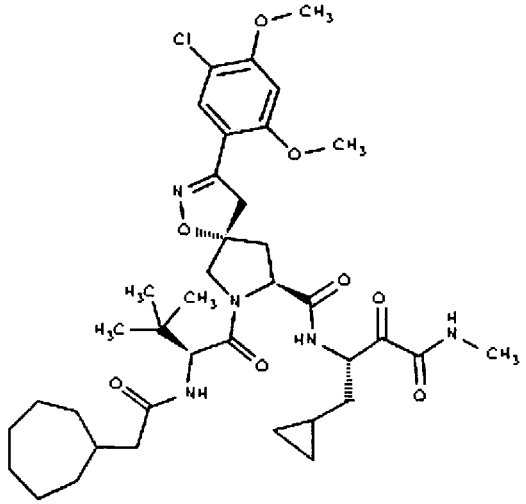
	599
	600
	601

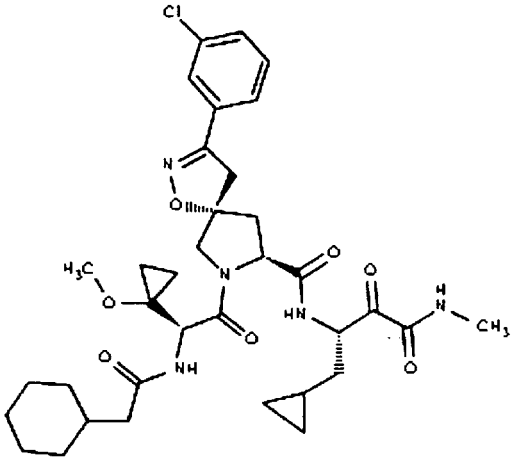
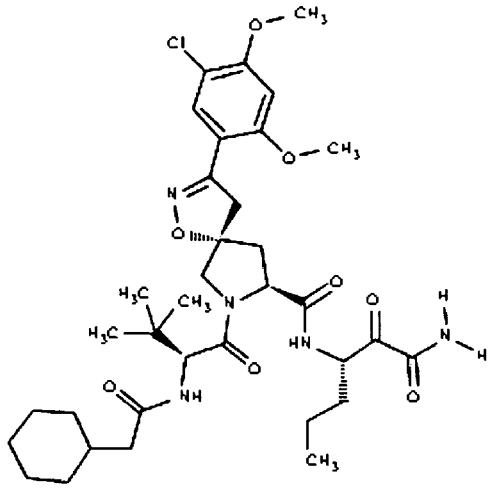
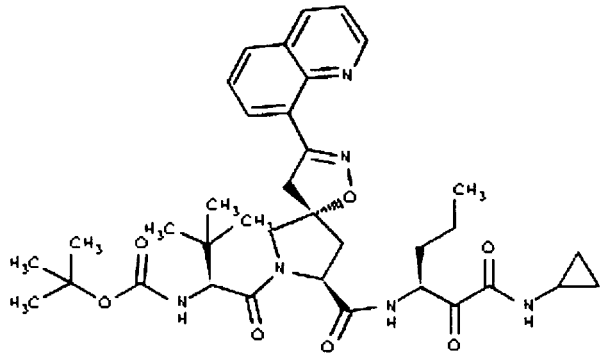
	602
	603
	604

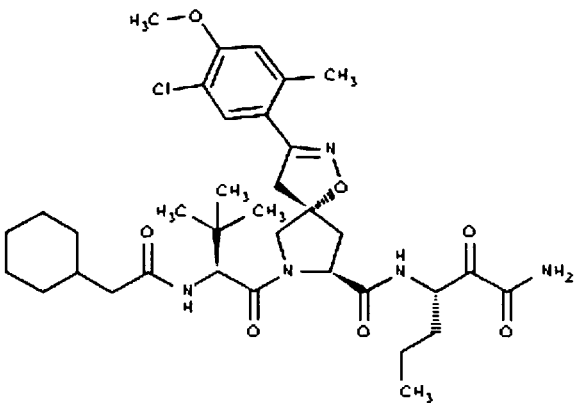
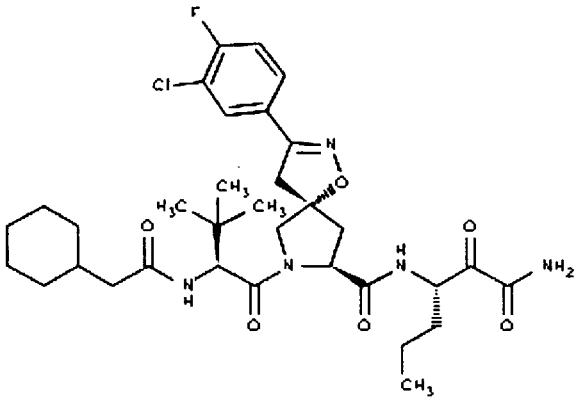
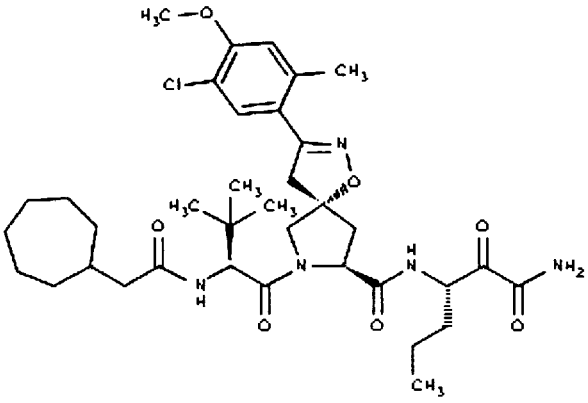
	605
	606
	607

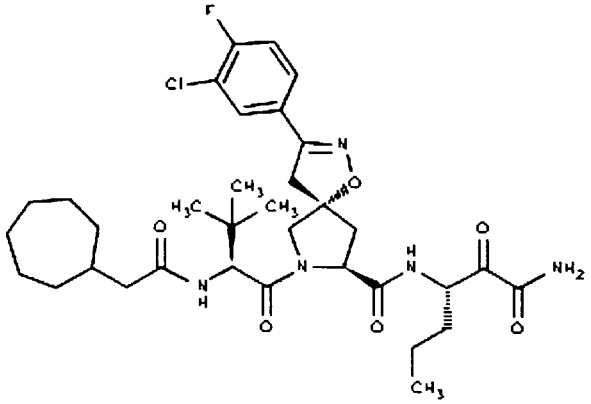
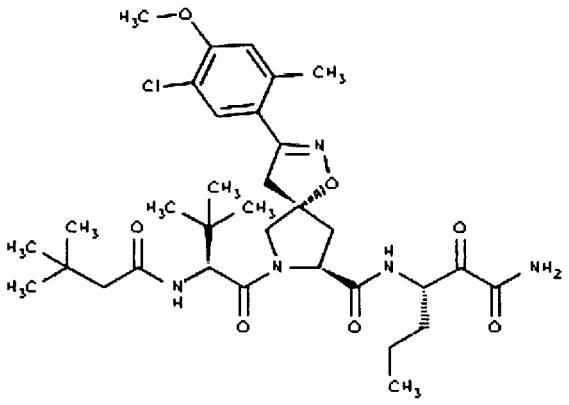
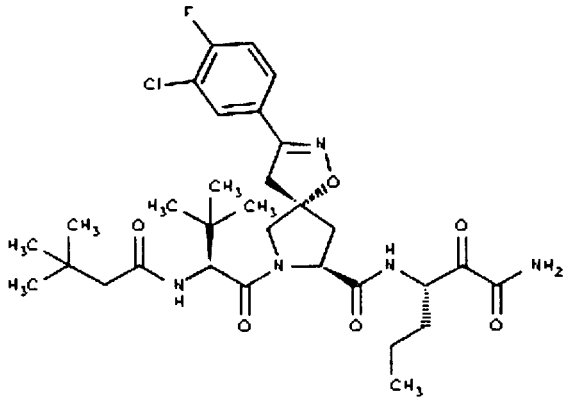
	608
	609
	610

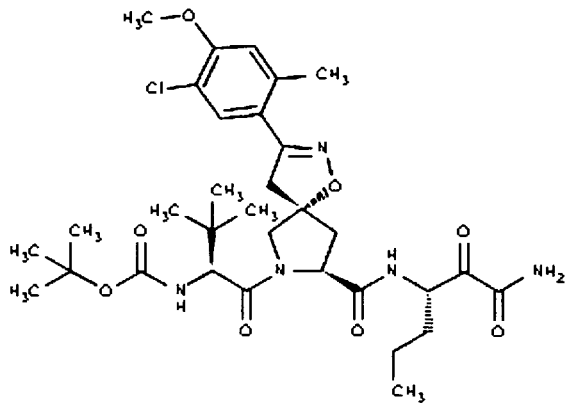
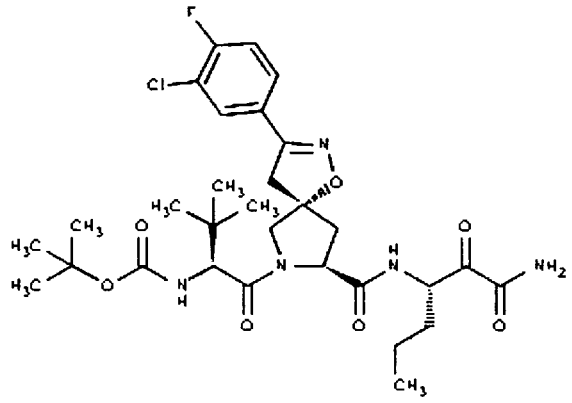
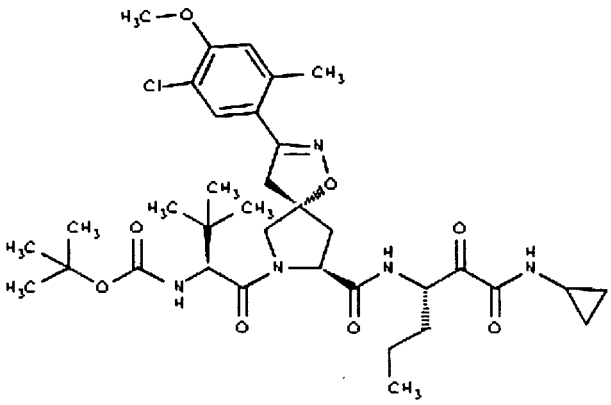
	611
	612
	613

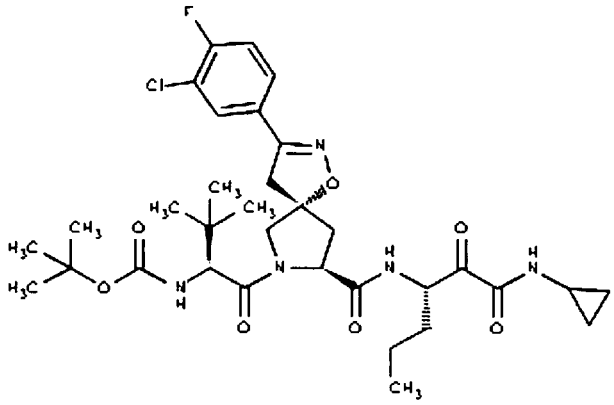
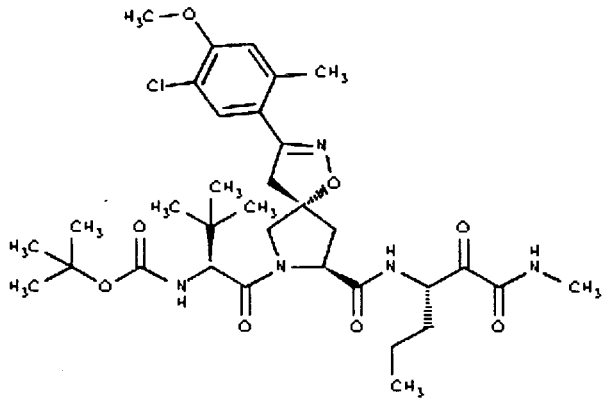
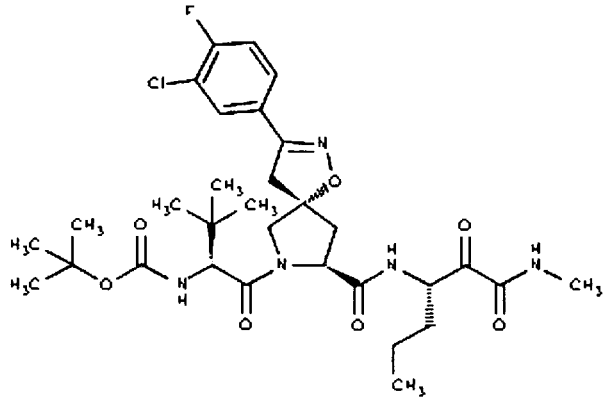
	614
	615
	616

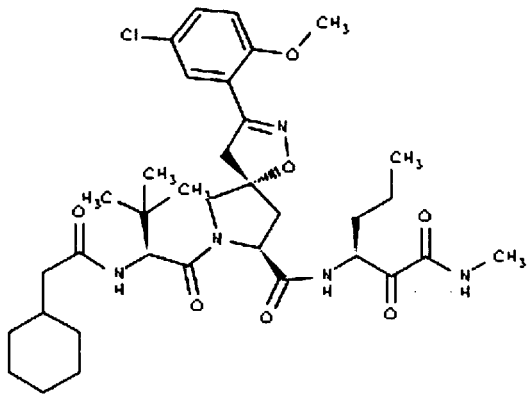
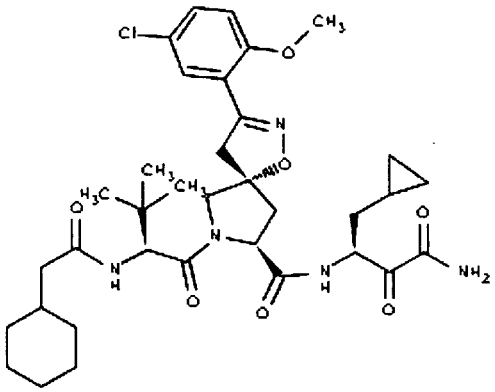
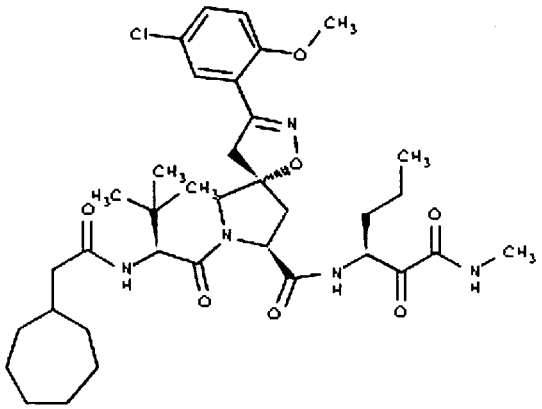
	617
	618
	619

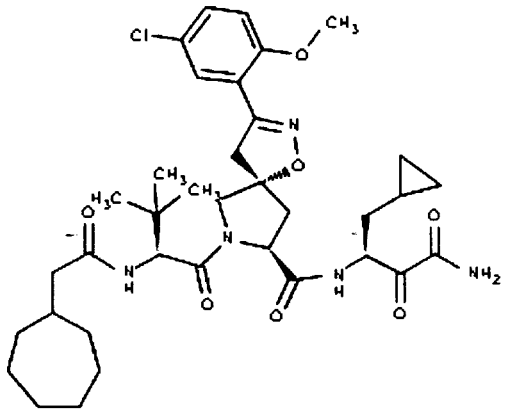
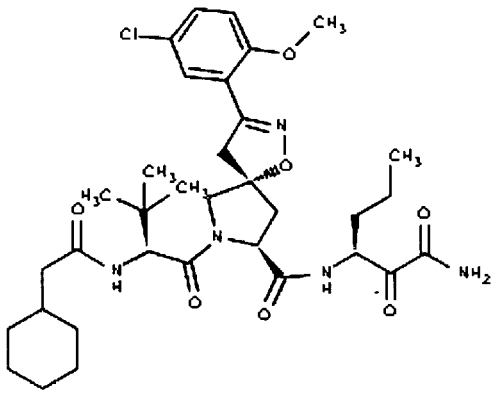
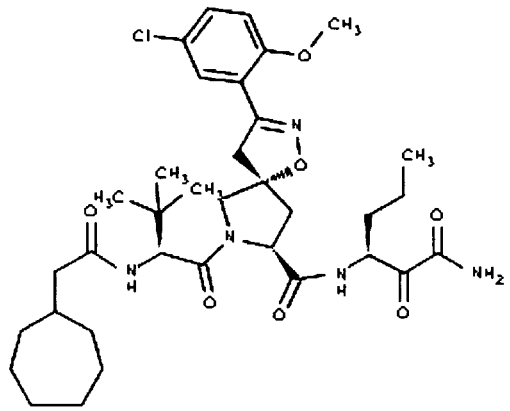
	620
	621
	622

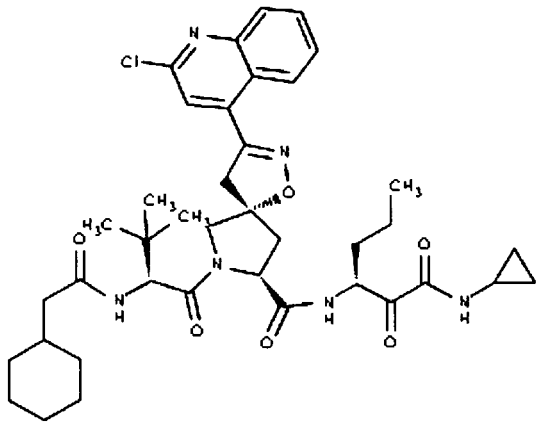
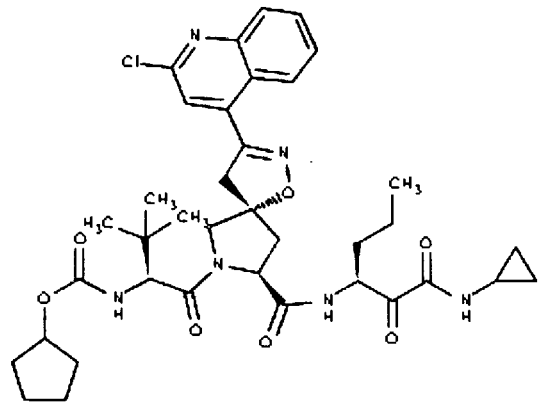
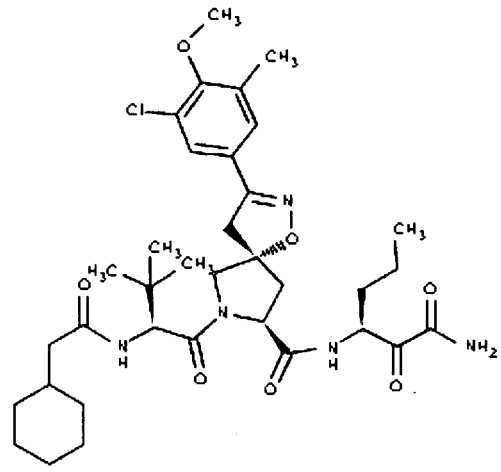
	623
	624
	625

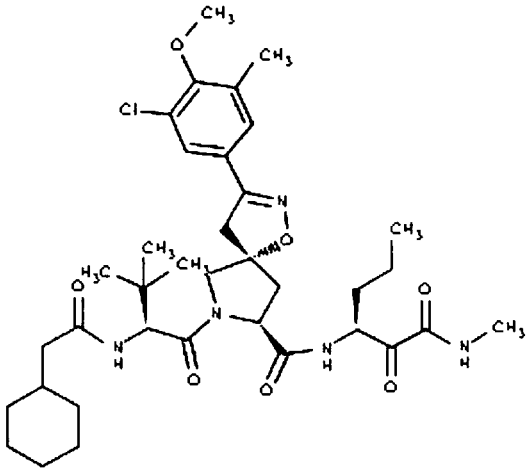
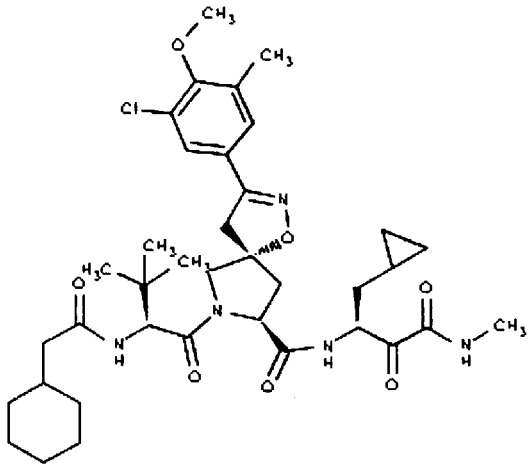
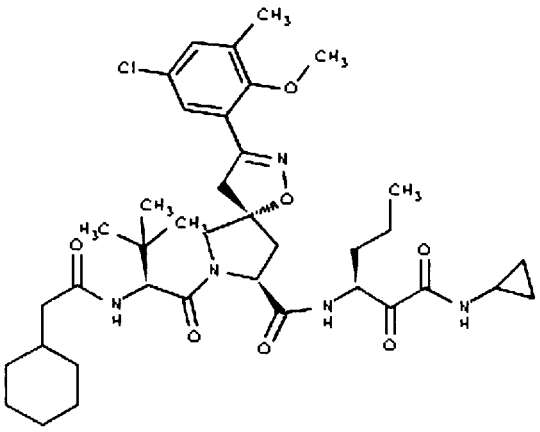
	626
	627
	628

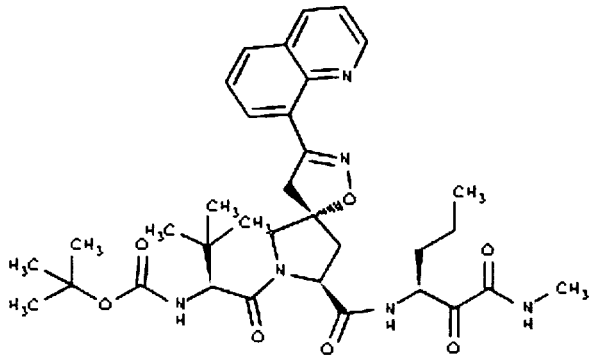
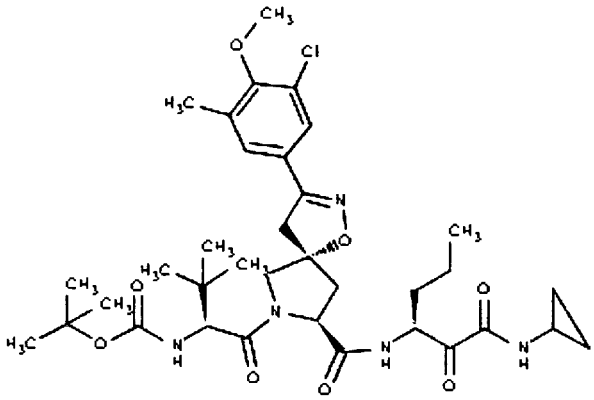
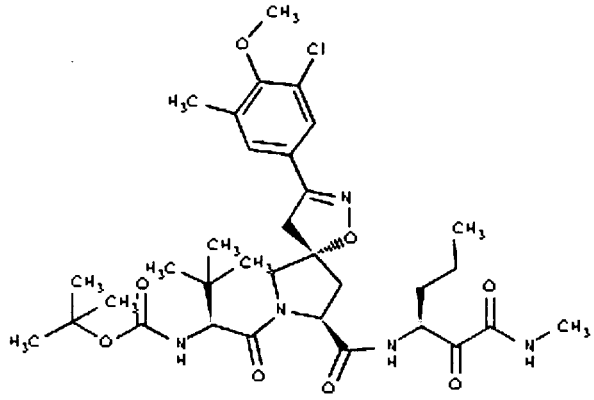
	629
	630
	631

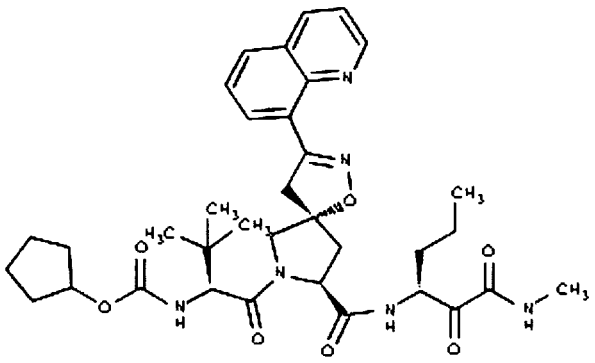
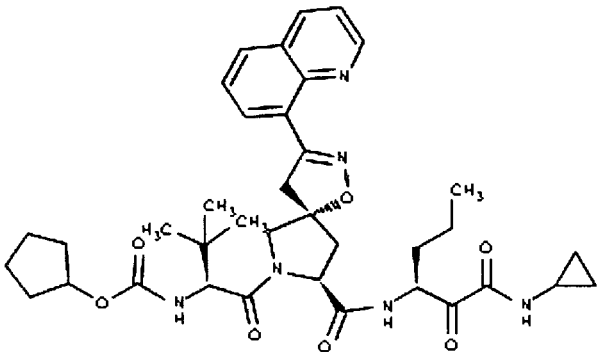
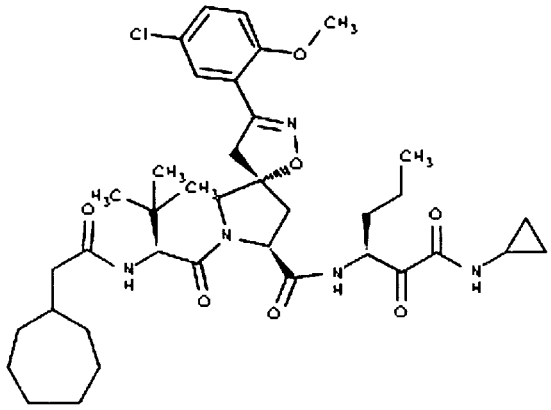
	632
	633
	634

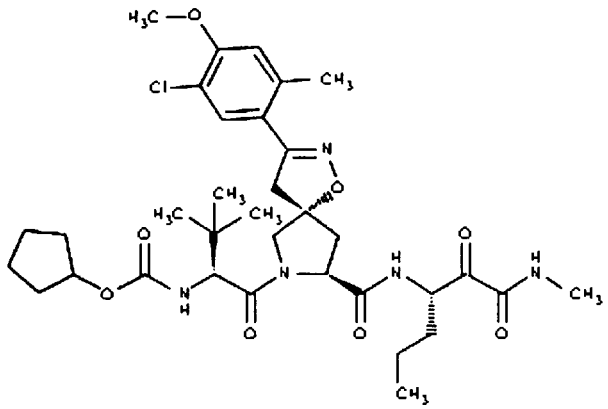
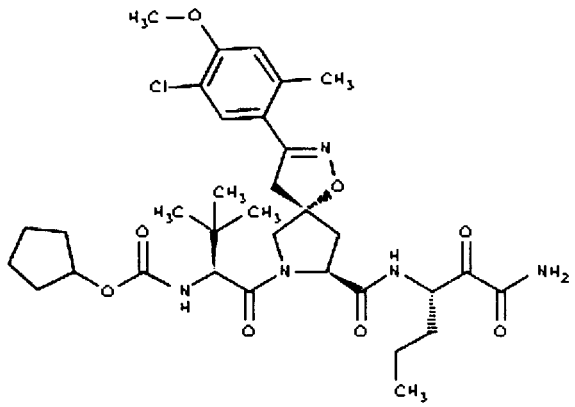
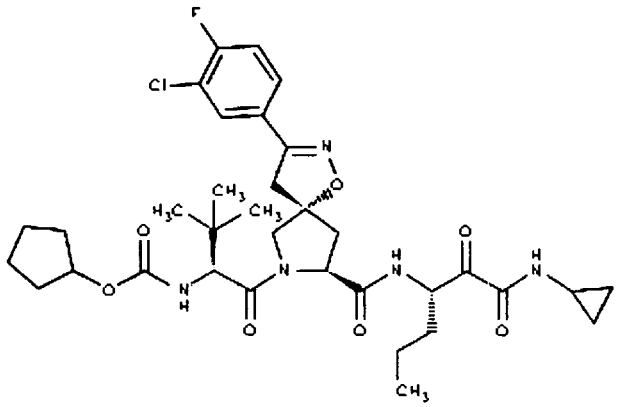
	635
	636
	637

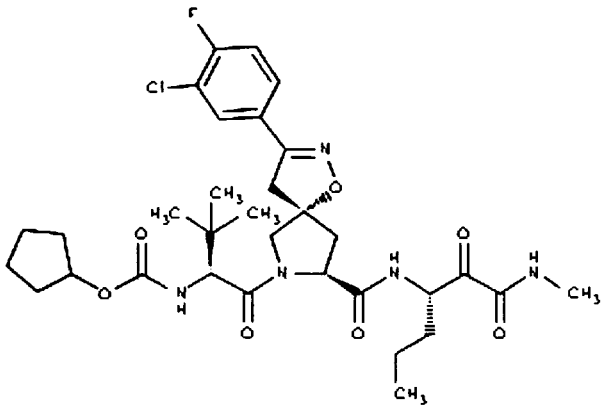
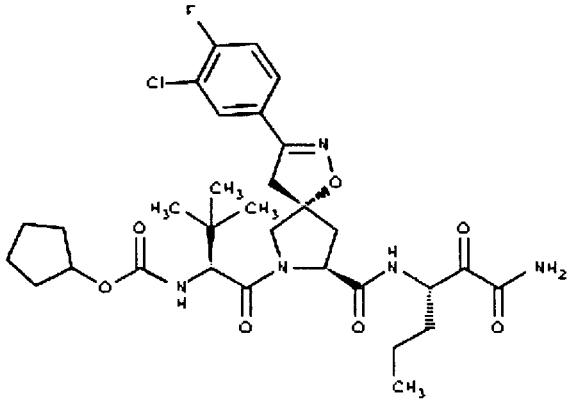
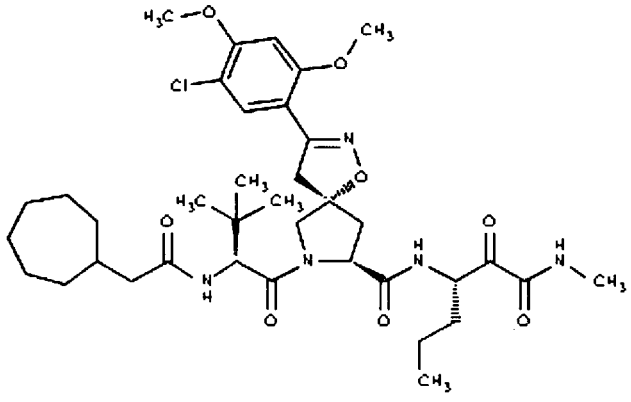
	638
	639
	640

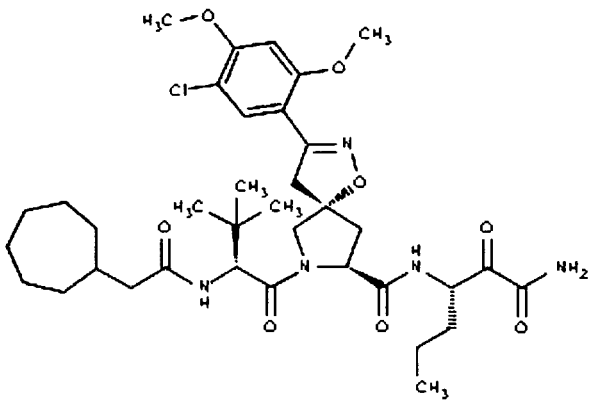
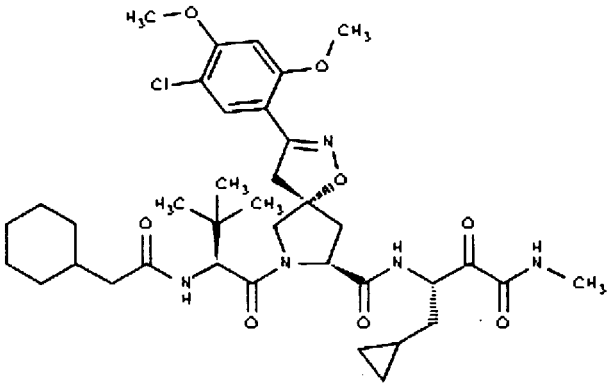
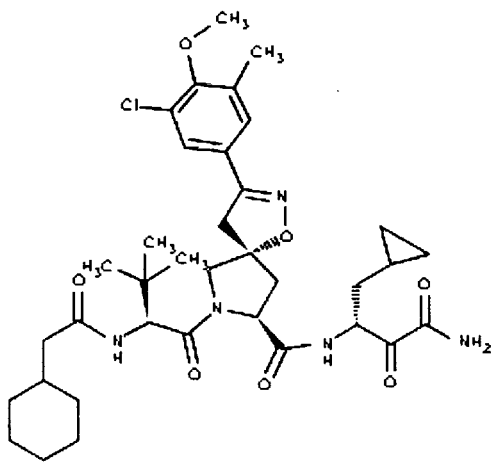
	641
	642
	643

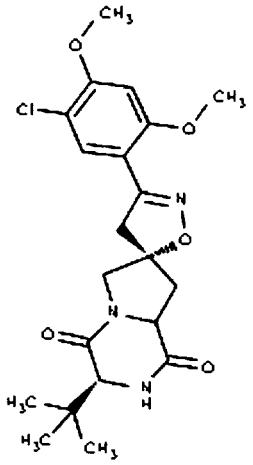
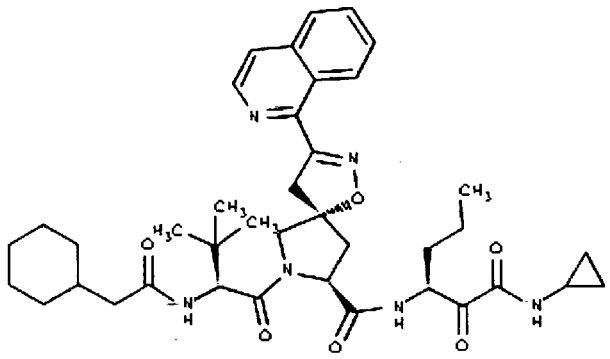
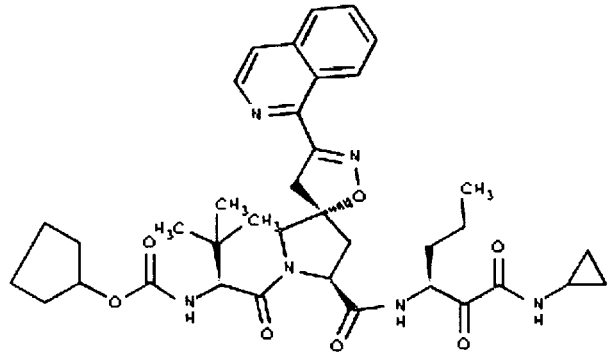
	644
	645
	646

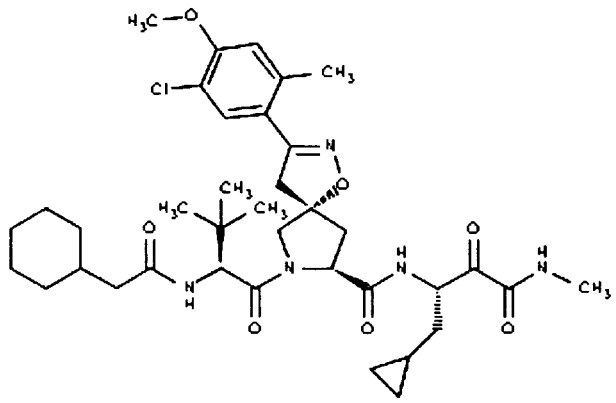
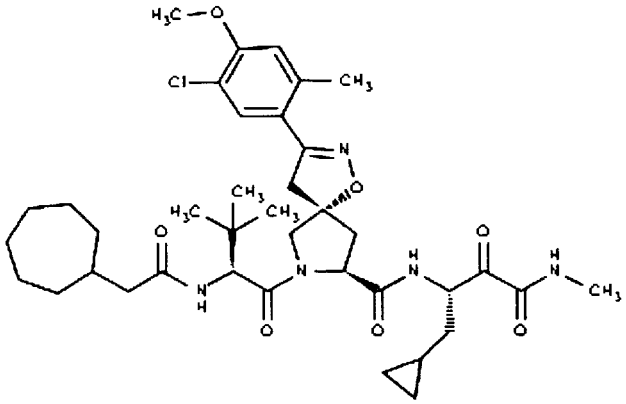
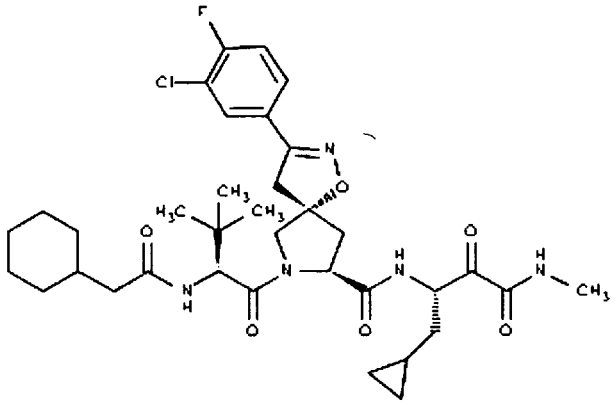
	647
	648
	649

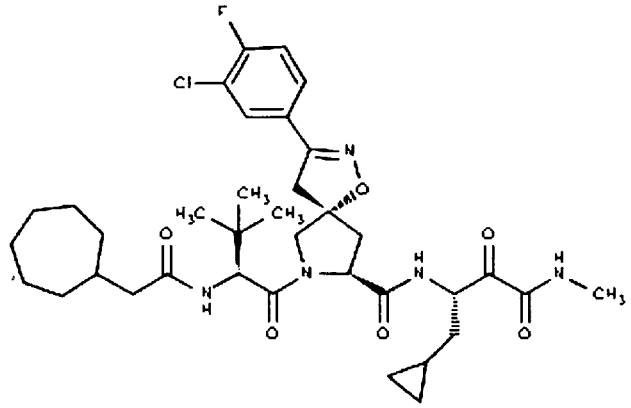
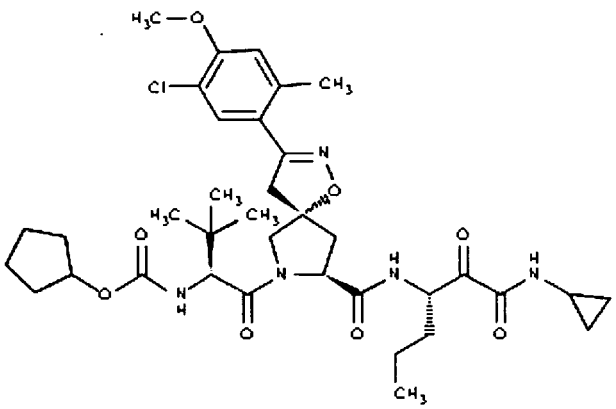
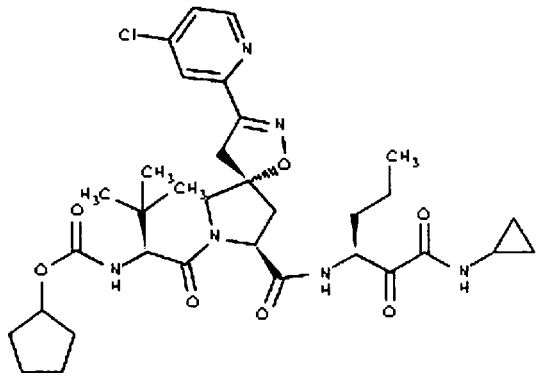
	650
	651
	652

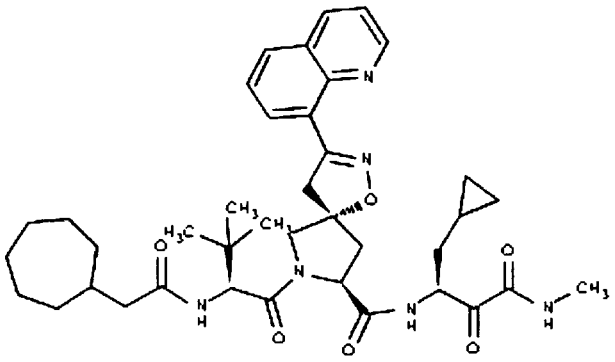
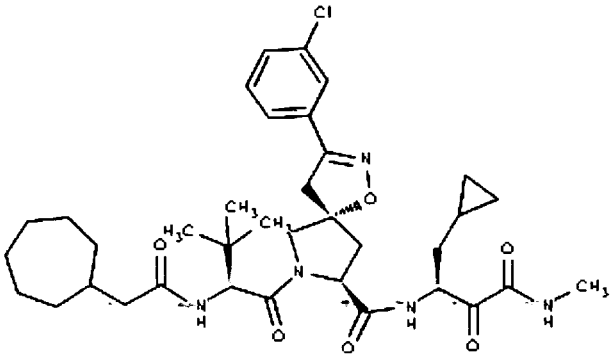
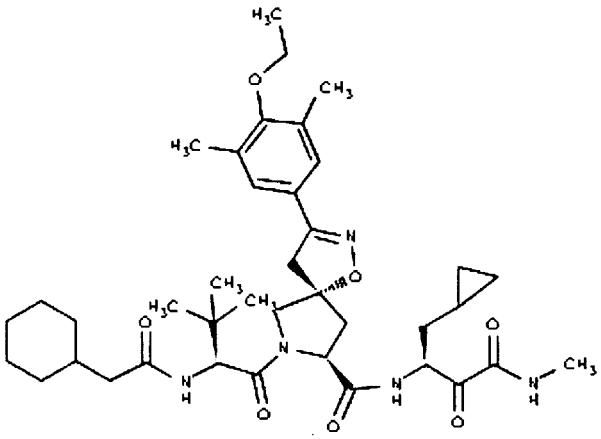
	653
	654
	655

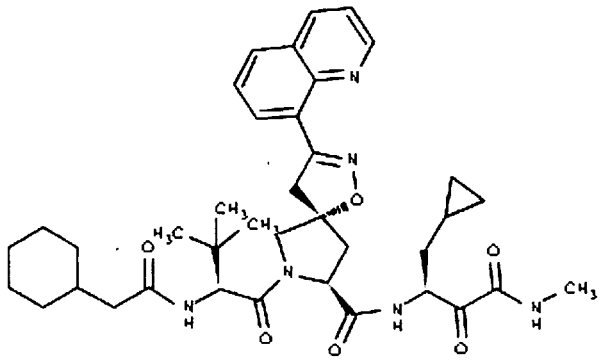
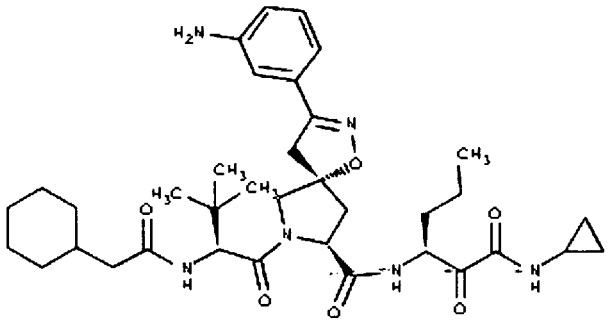
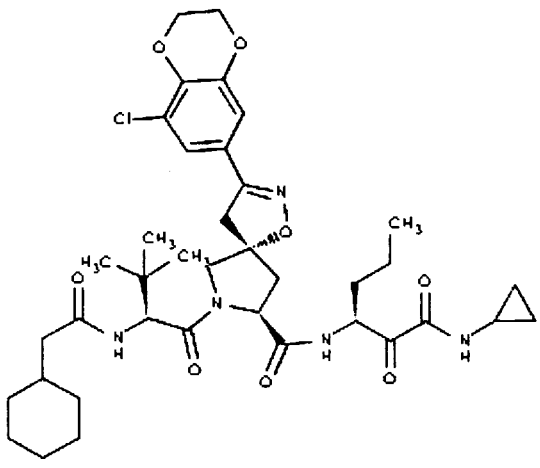
	656
	657
	658

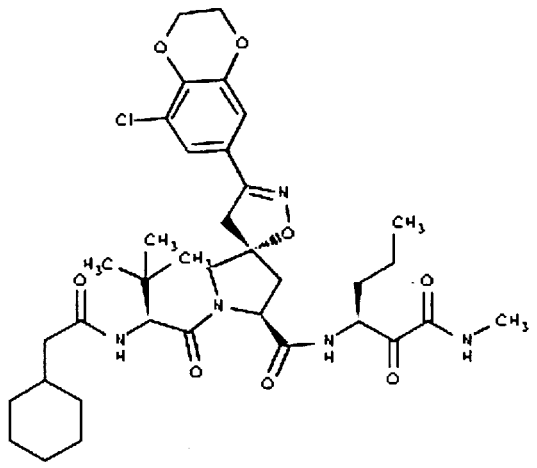
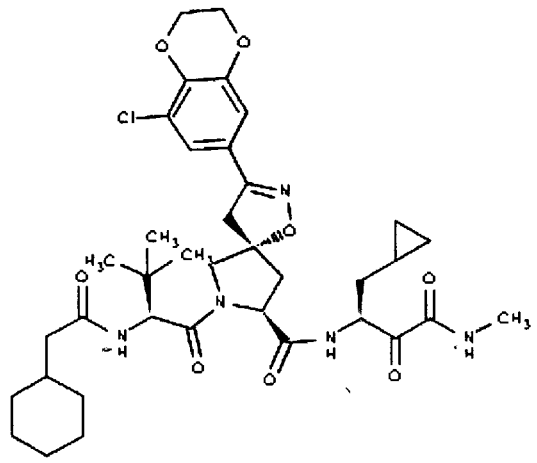
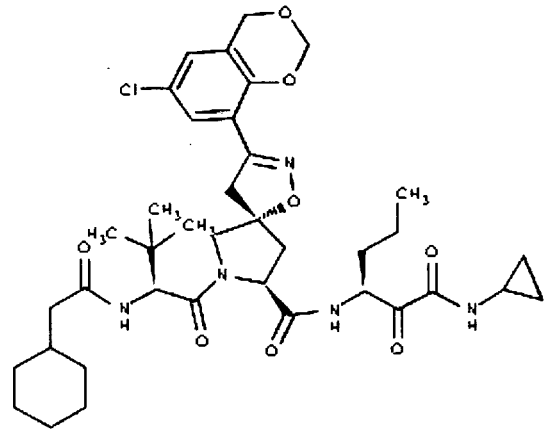
	659
	660
	661

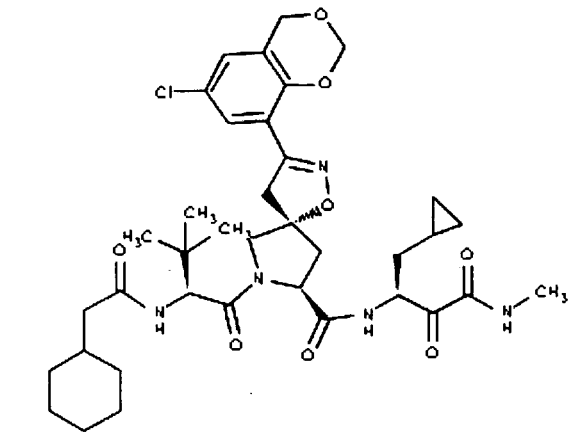
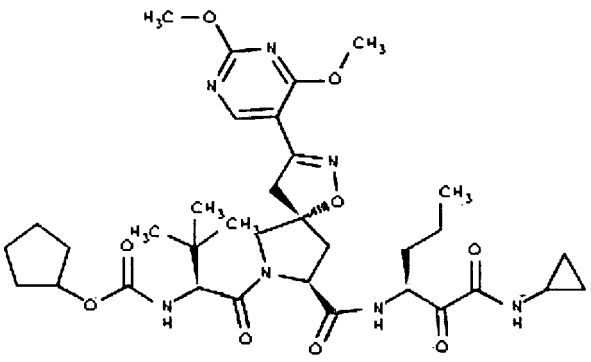
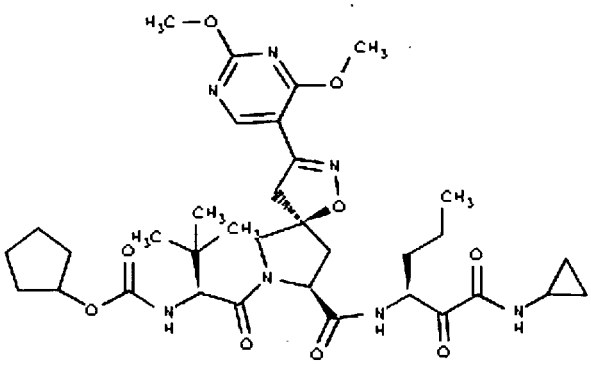
	662
	663
	664

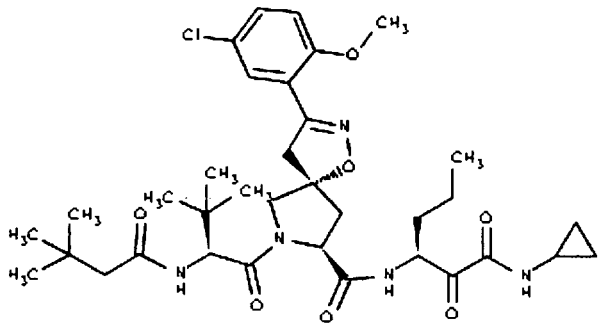
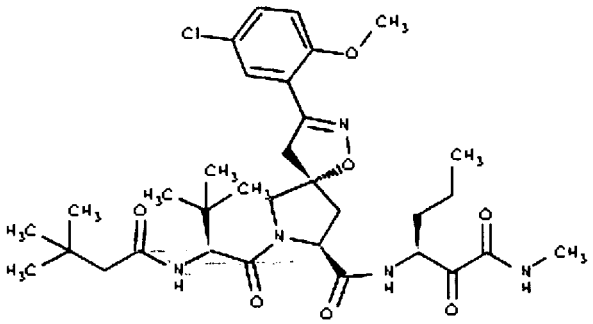
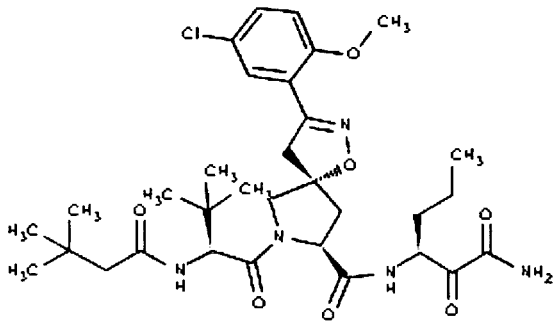
	665
	666
	667

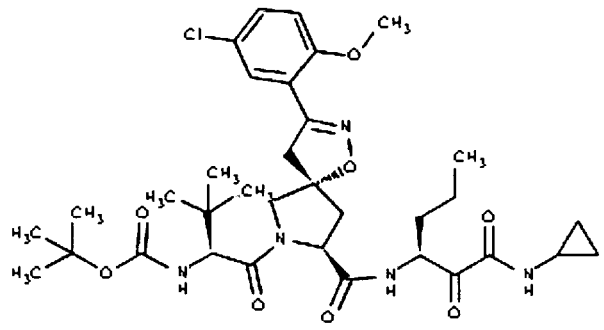
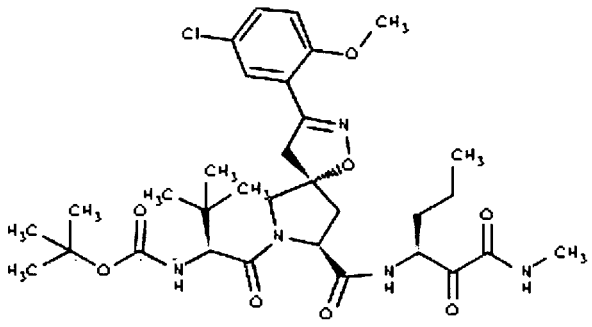
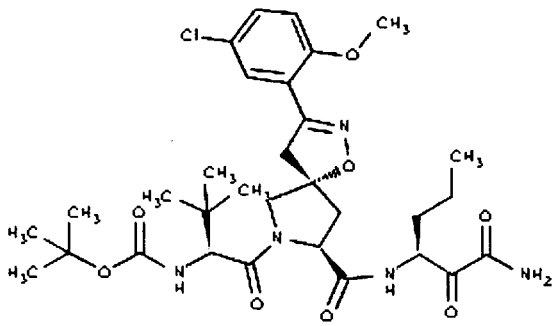
	668
	669
	670

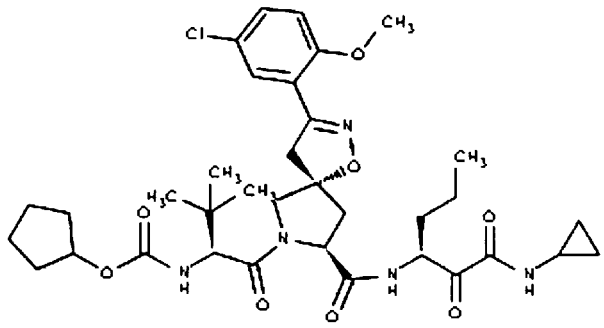
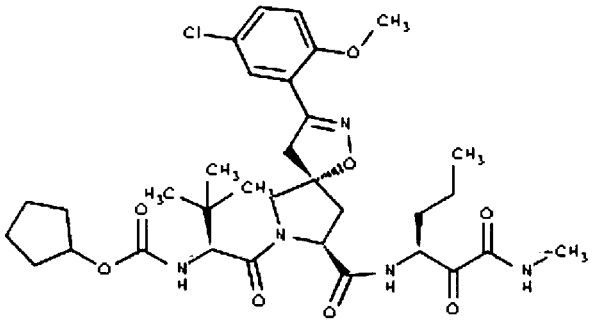
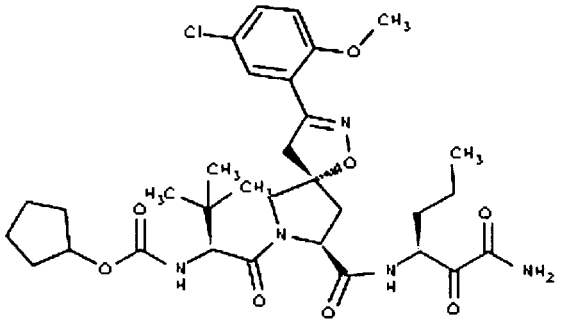
	671
	672
	673

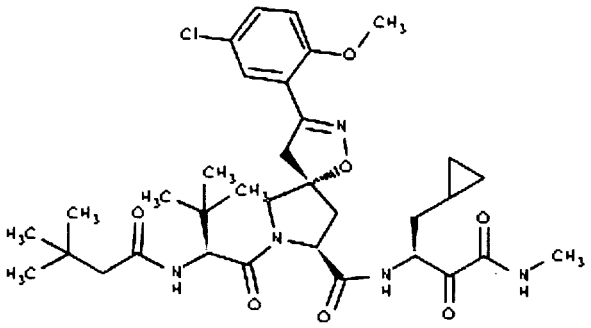
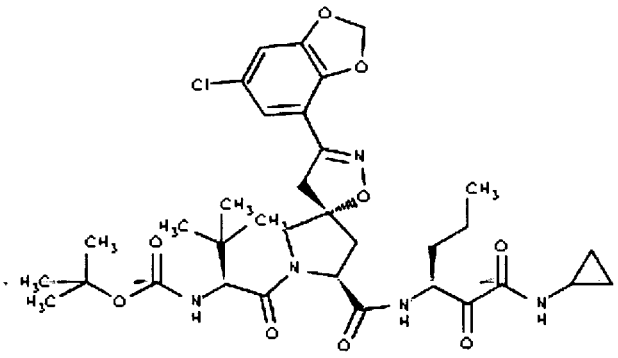
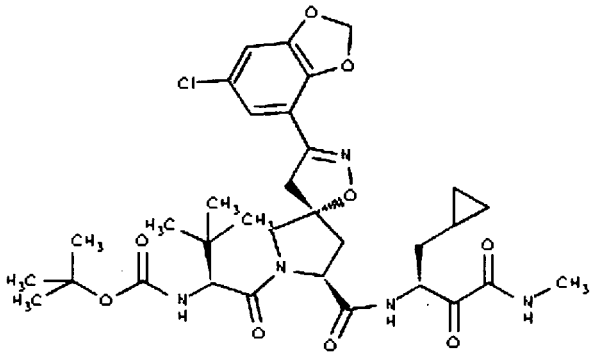
	674
	675
	676

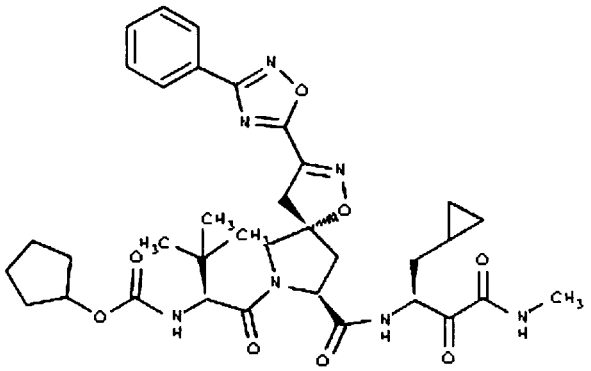
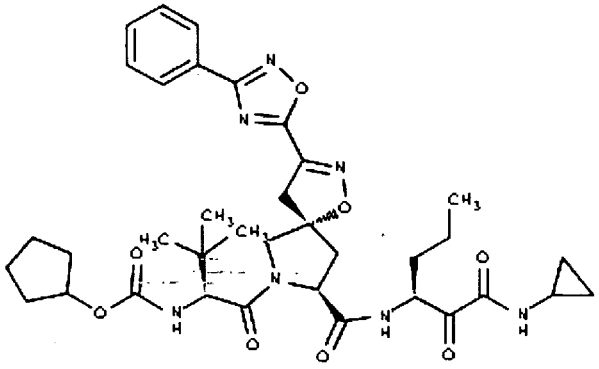
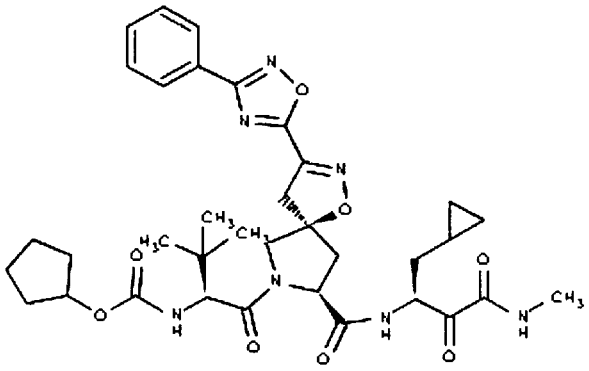
	677
	678
	679

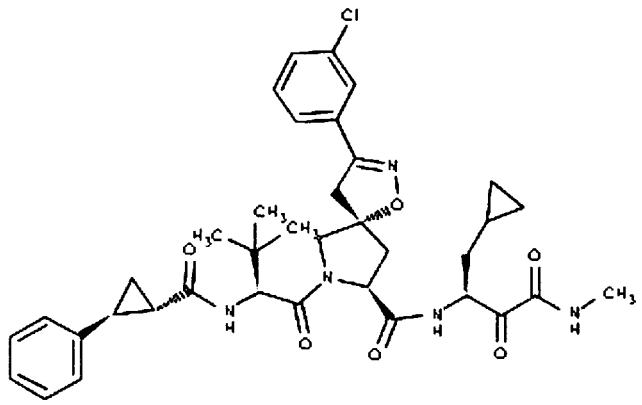
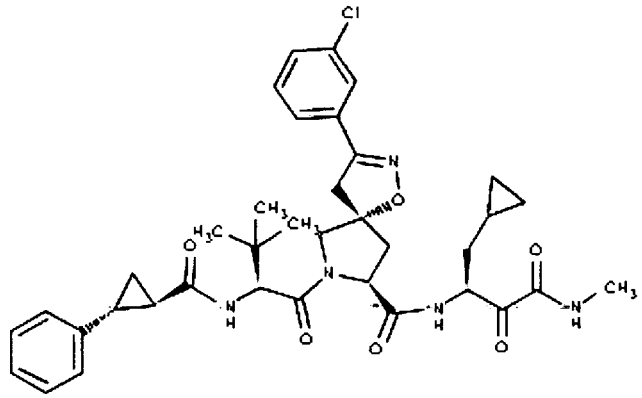
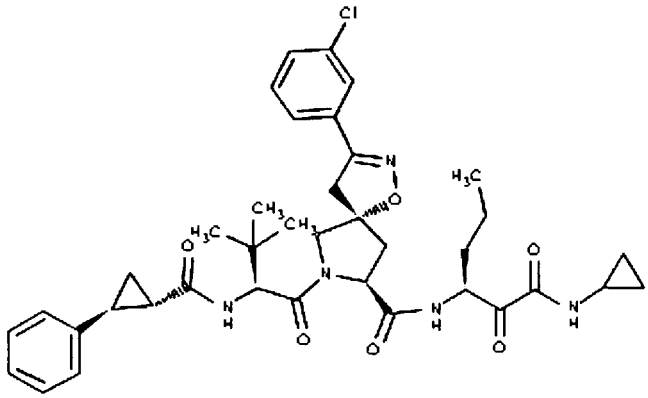
	680
	681
	682

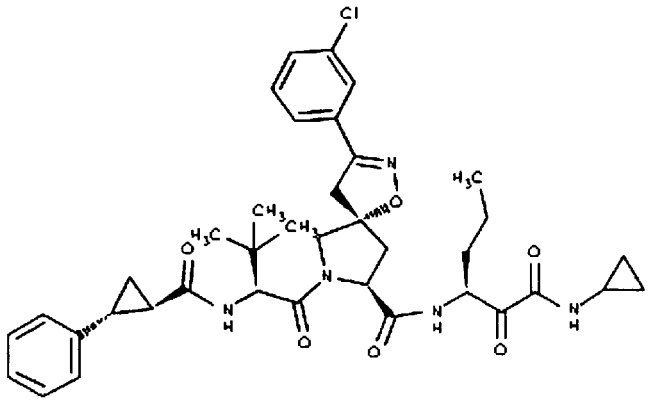
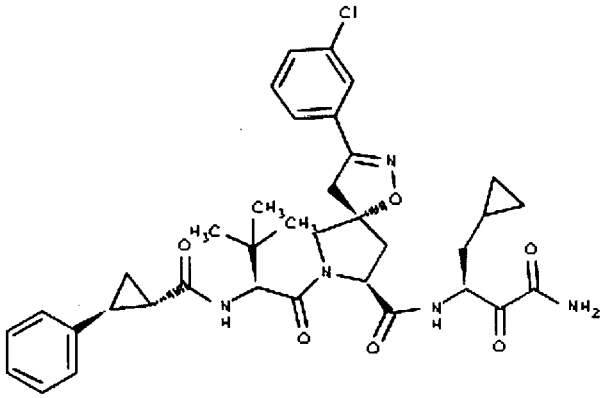
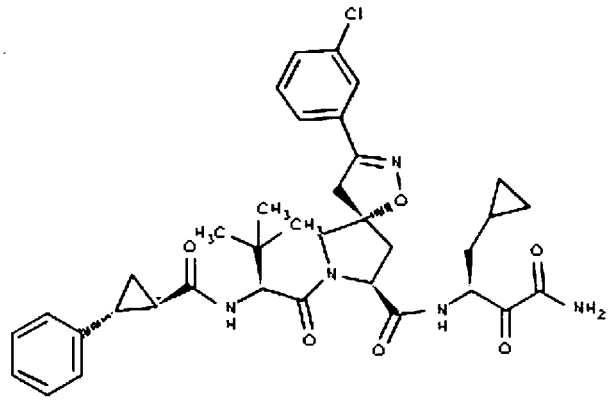
	683
	684
	685

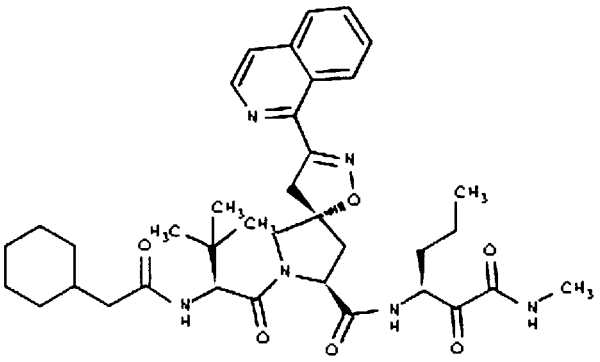
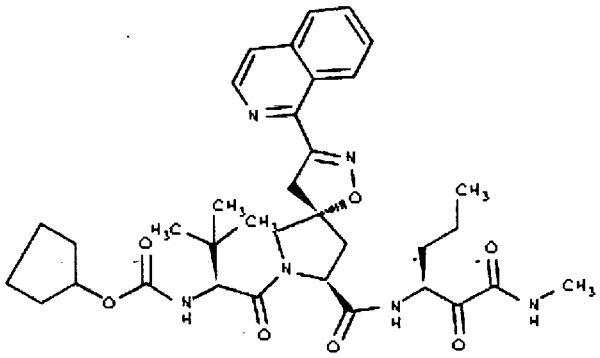
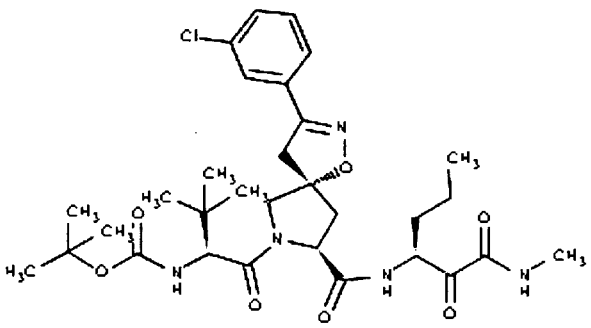
	686
	687
	688

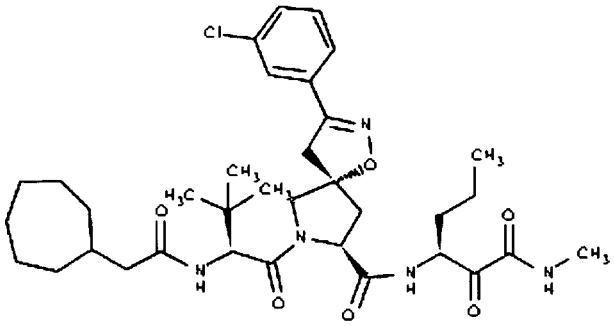
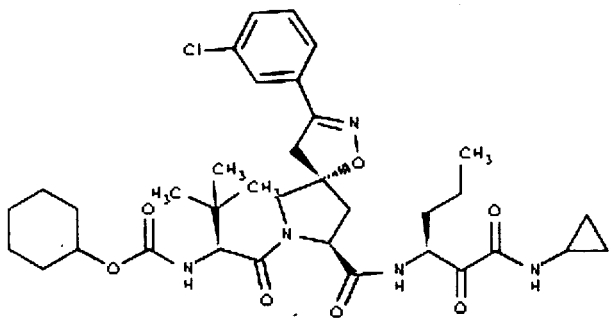
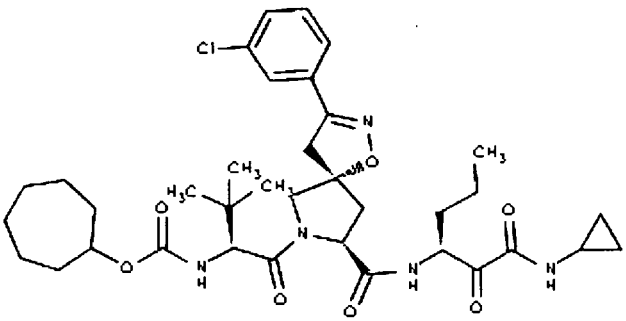
	689
	690
	691

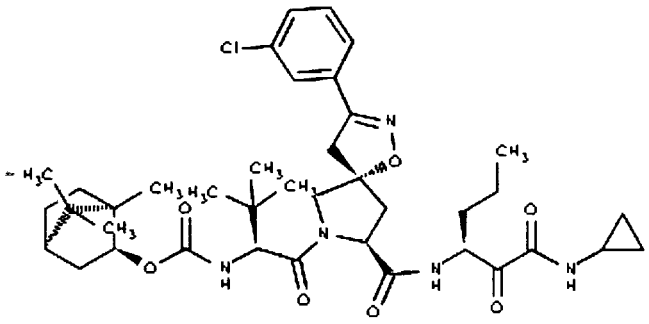
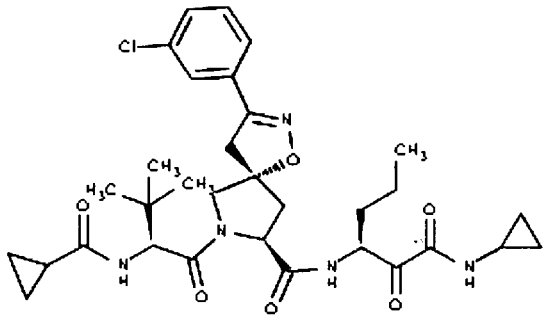
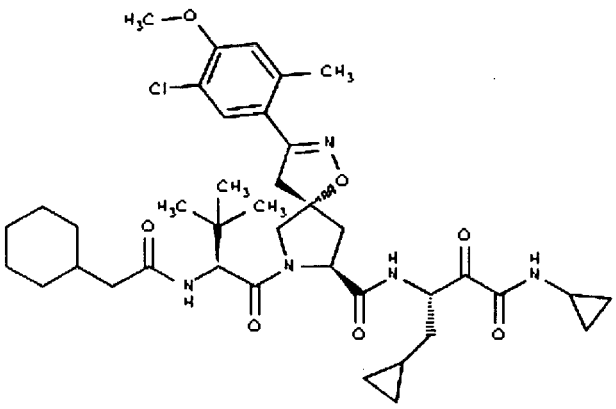
	692
	693
	694

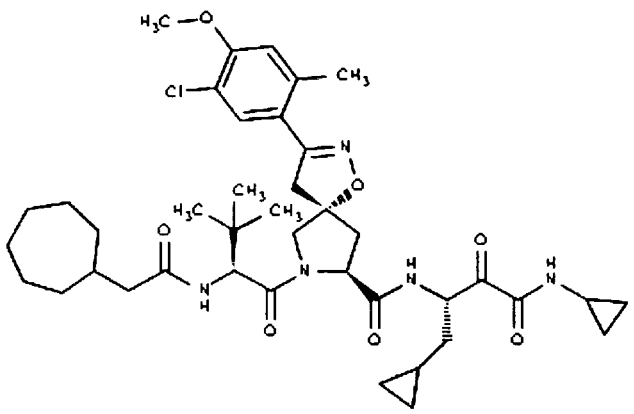
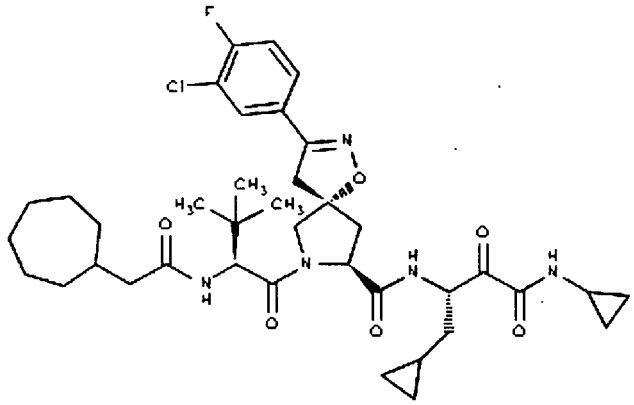
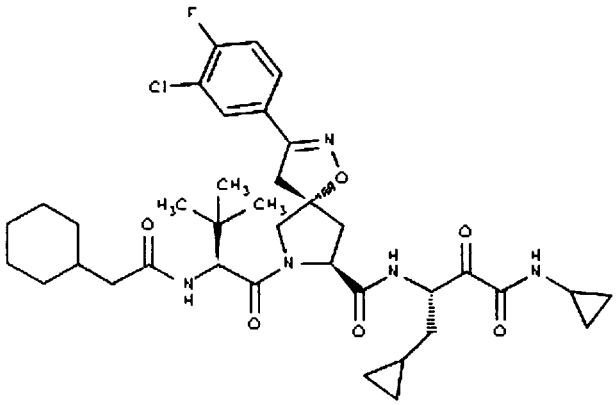
	695
	696
	697

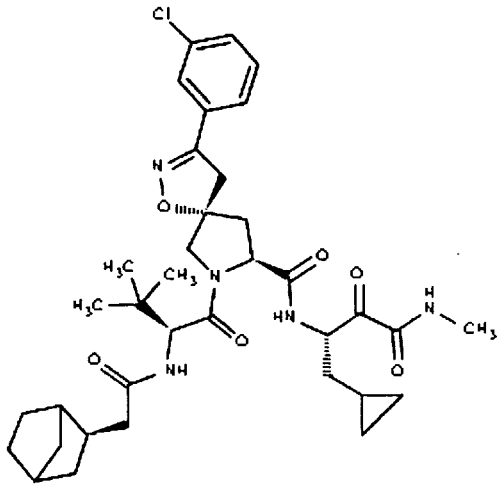
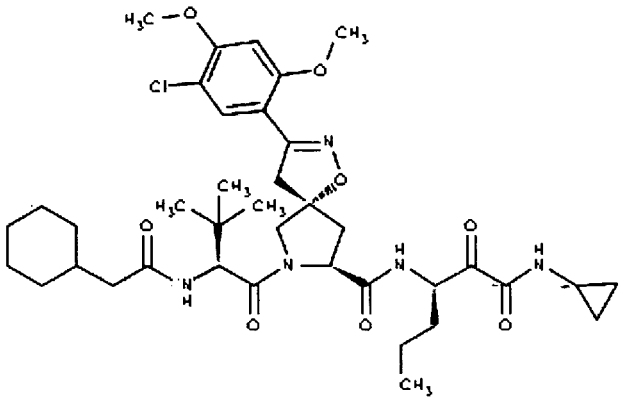
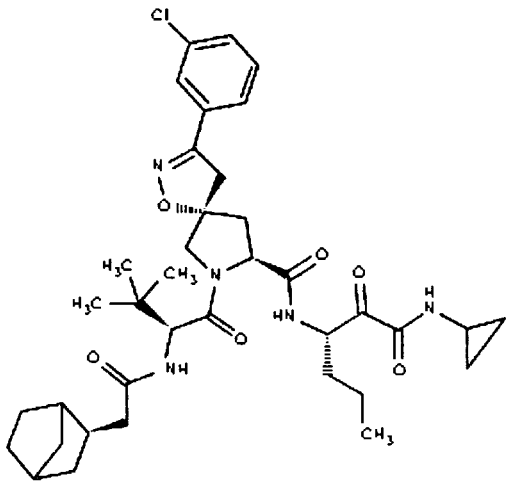
	698
	699
	700

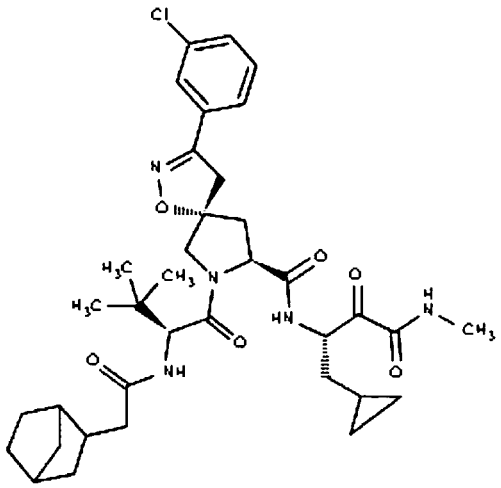
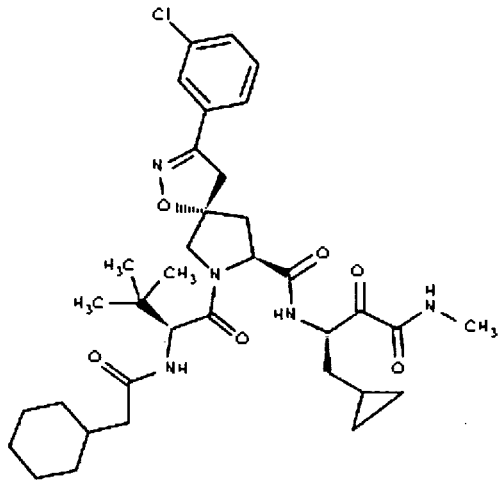
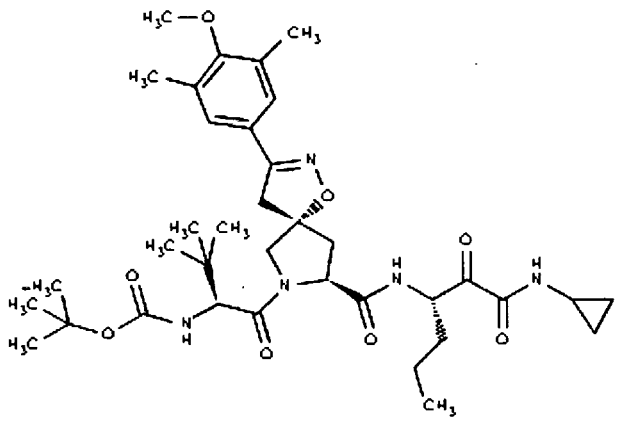
	701
	702
	703

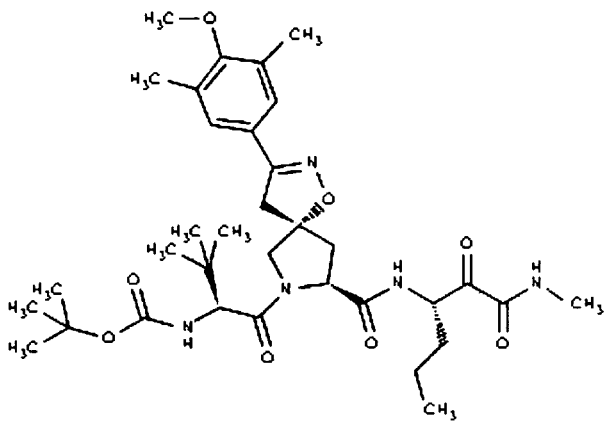
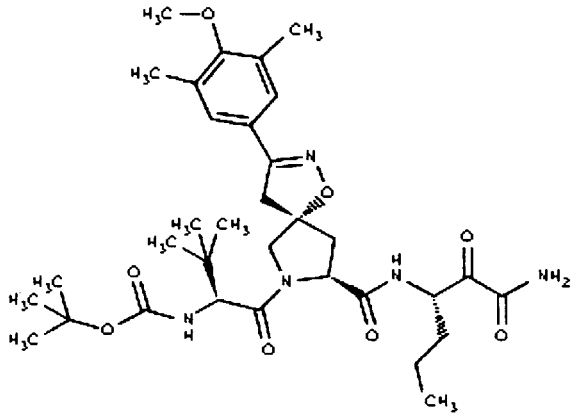
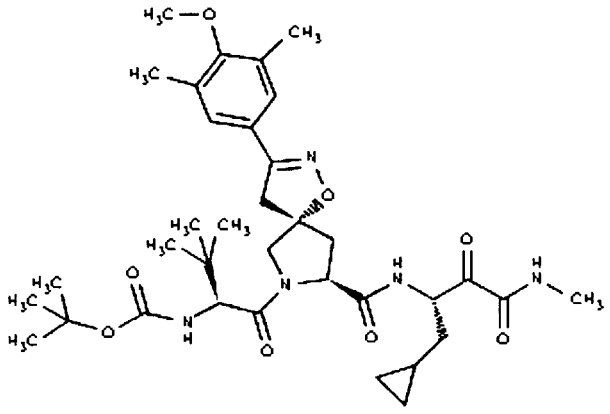
	704
	705
	706

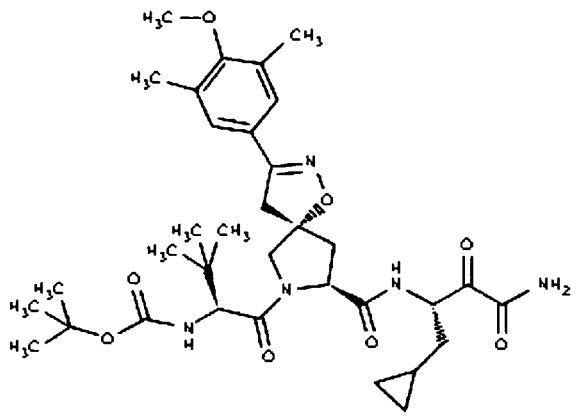
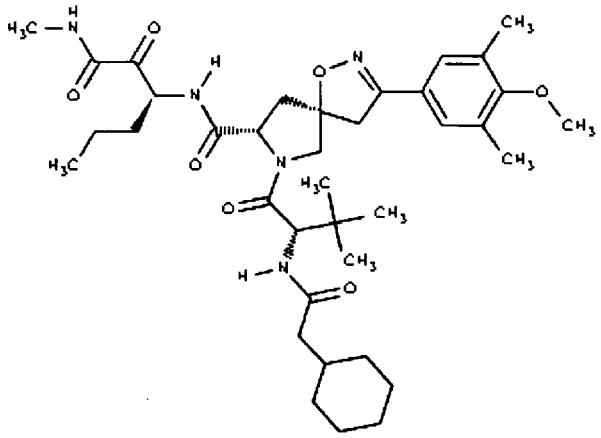
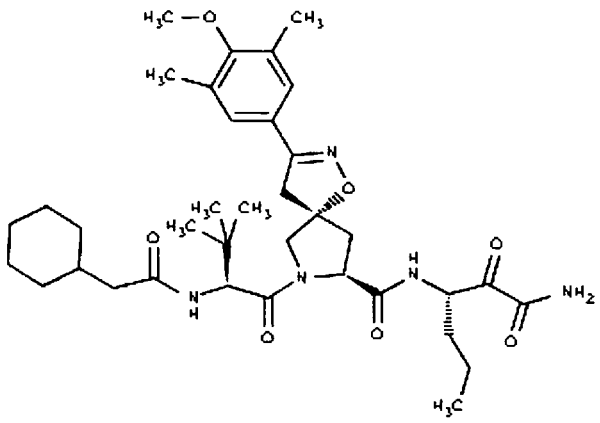
	707
	708
	709

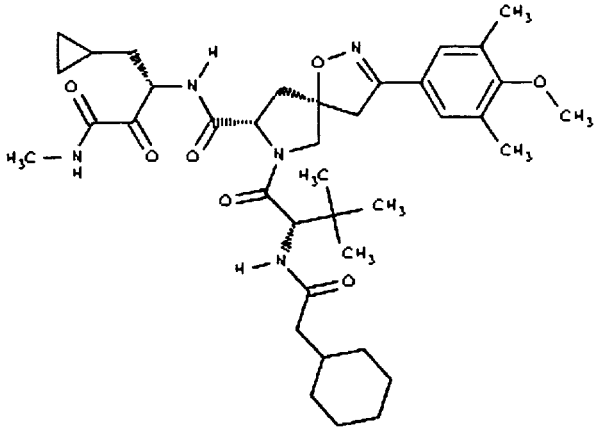
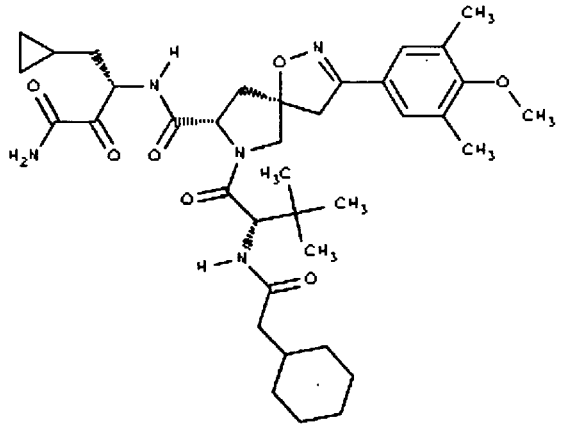
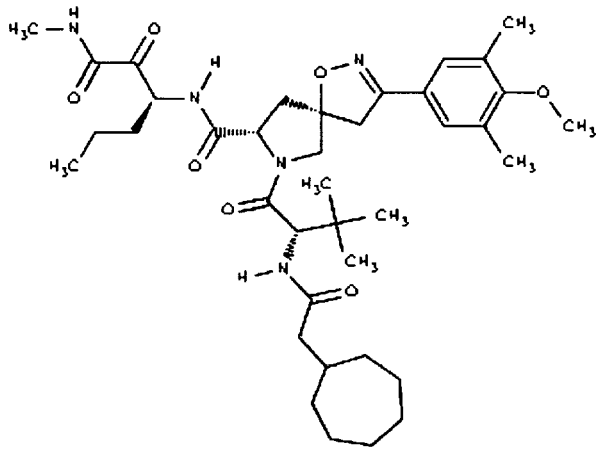
	710
	711
	712

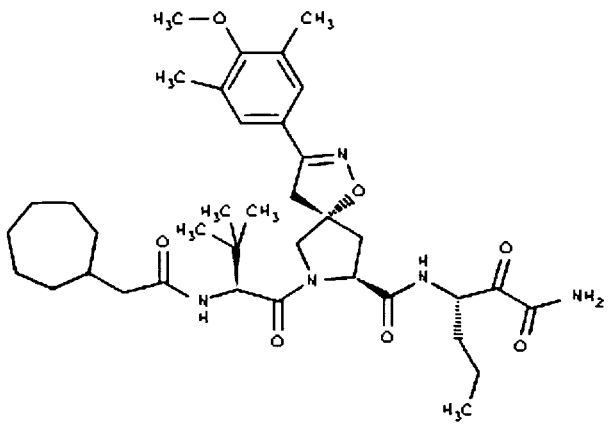
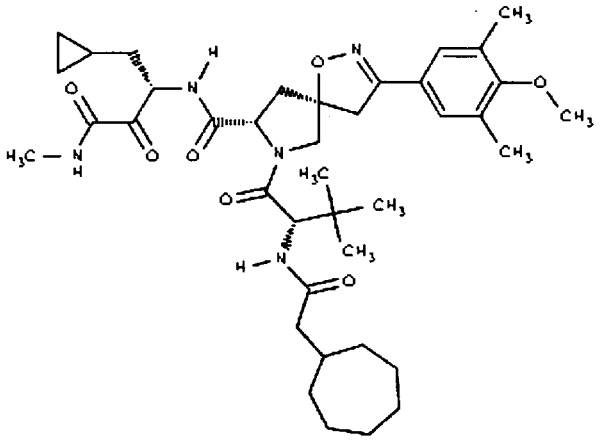
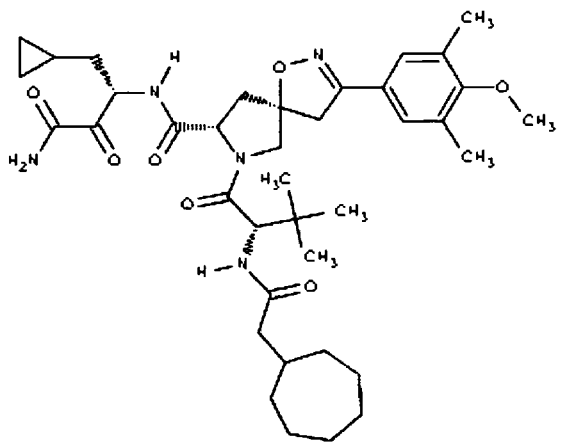
	713
	714
	715

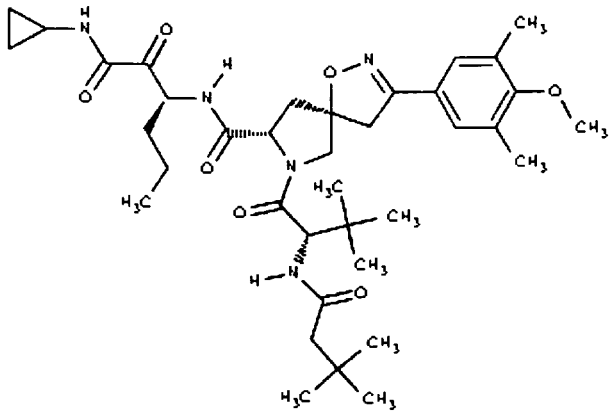
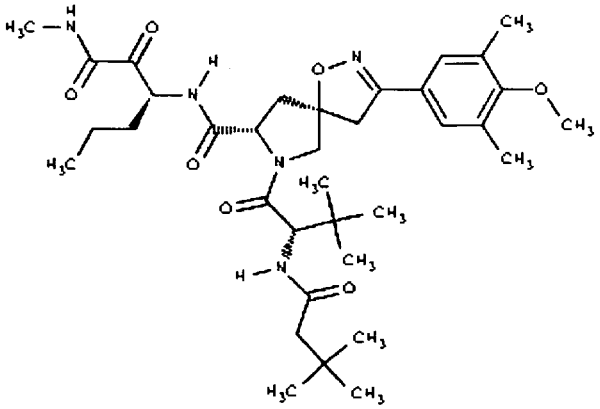
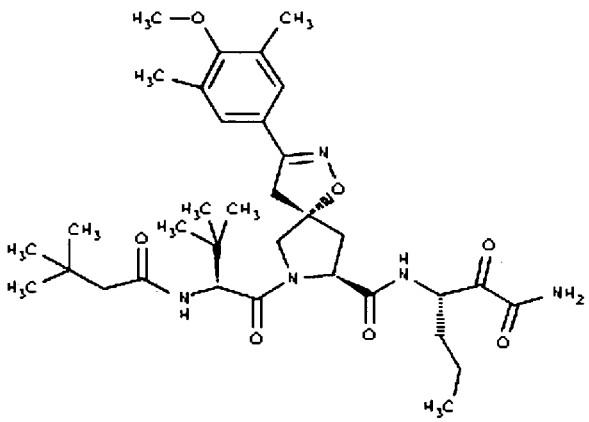
	716
	717
	718

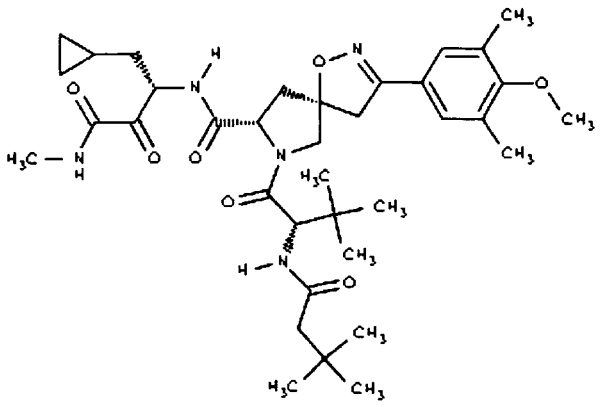
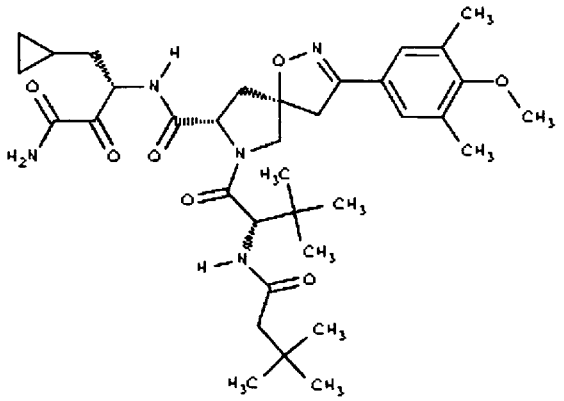
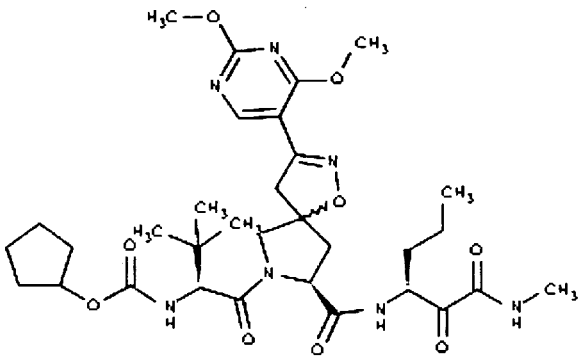
	719
	720
	721

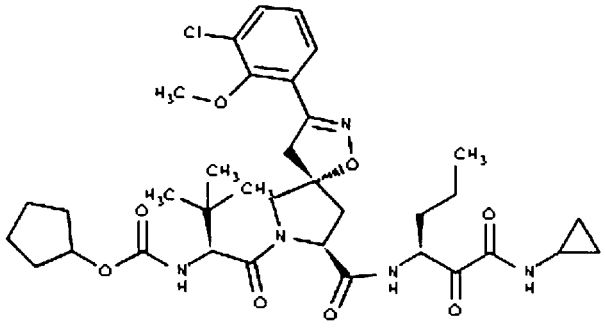
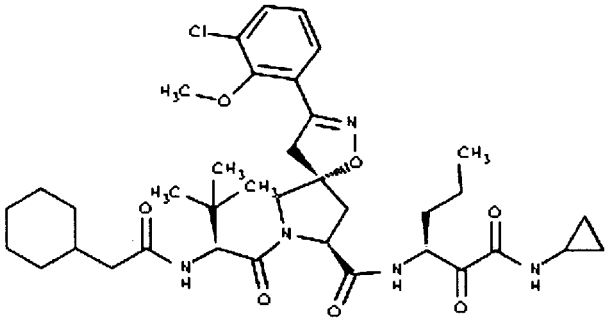
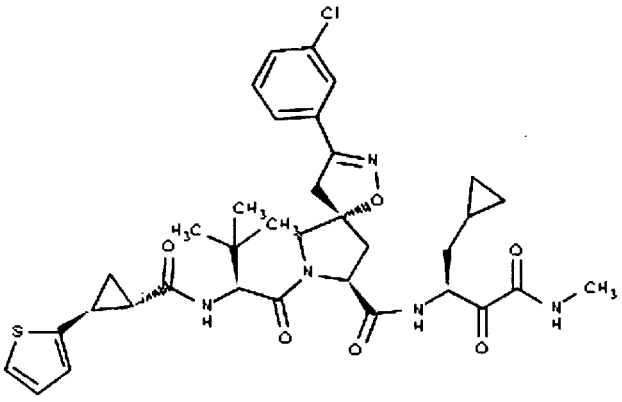
	722
	723
	724

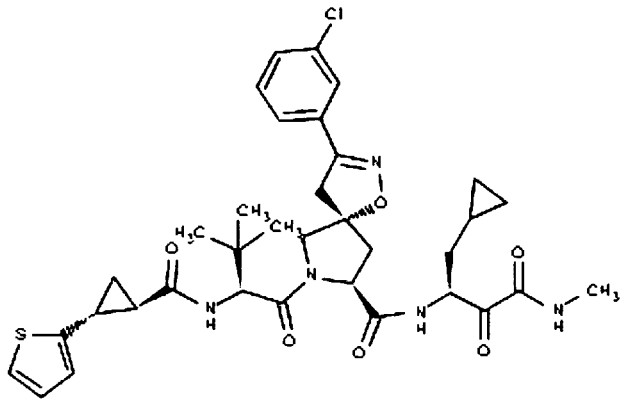
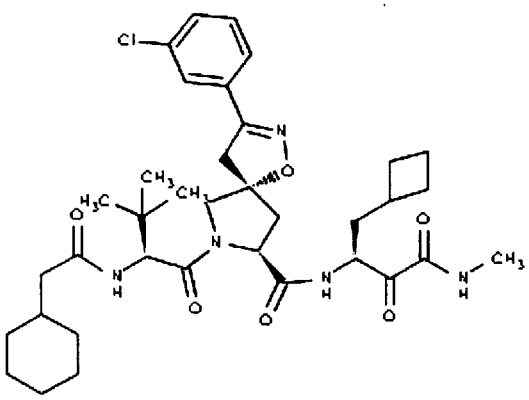
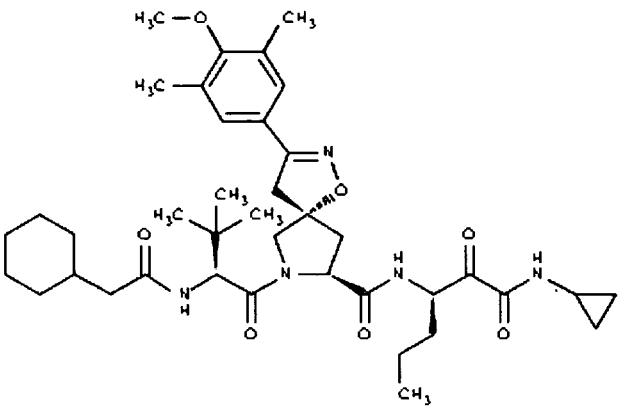
	725
	726
	727

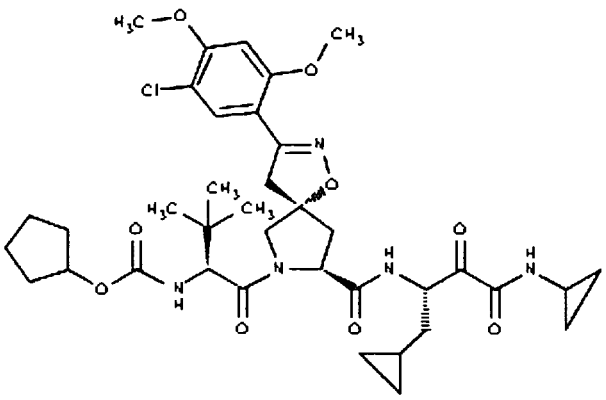
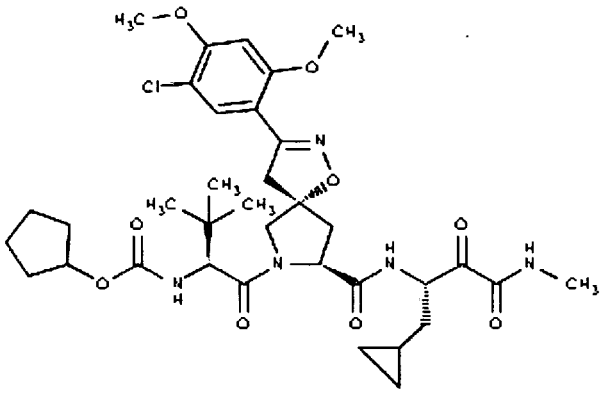
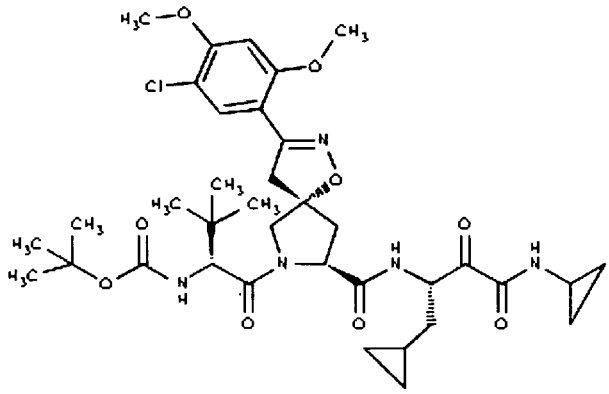
	728
	729
	730

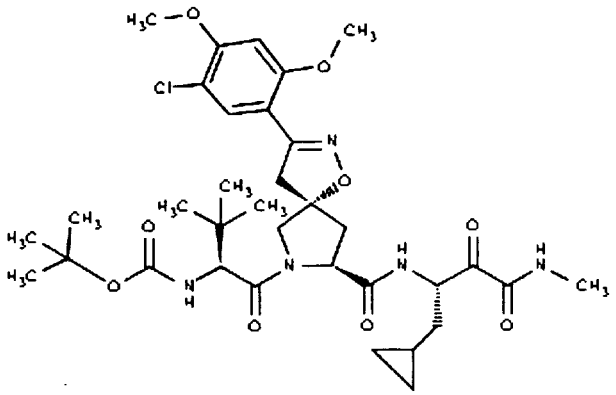
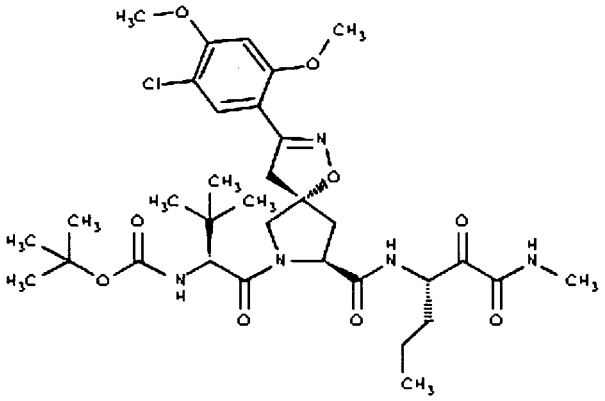
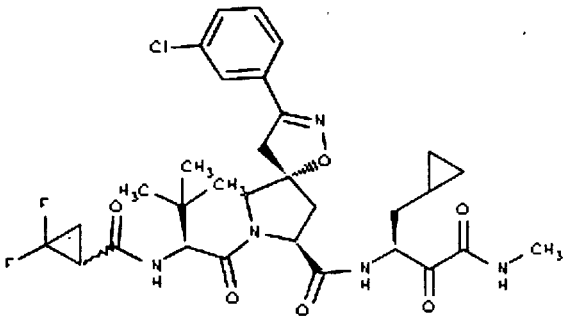
	731
	732
	733

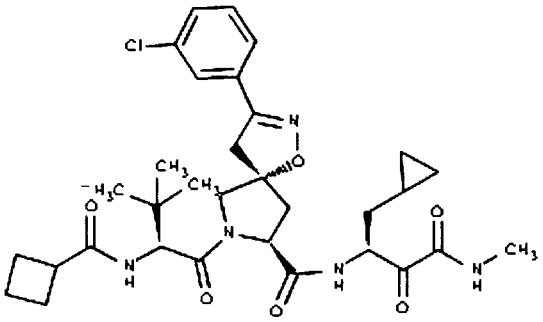
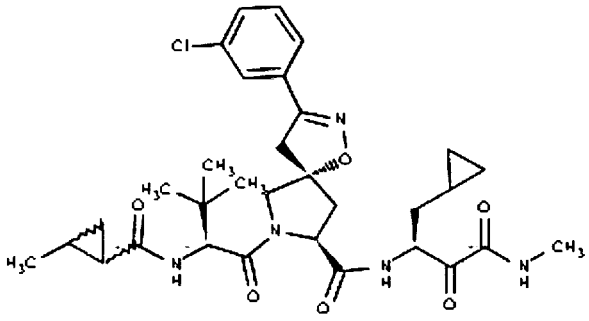
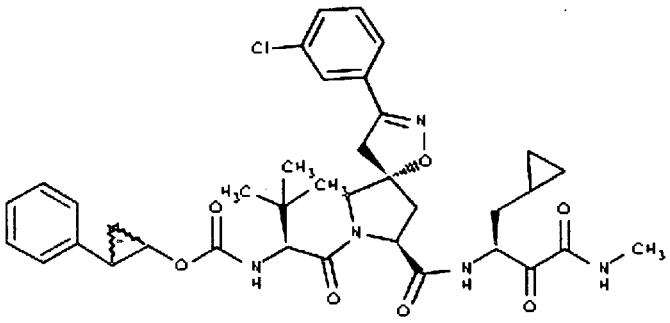
	734
	735
	736

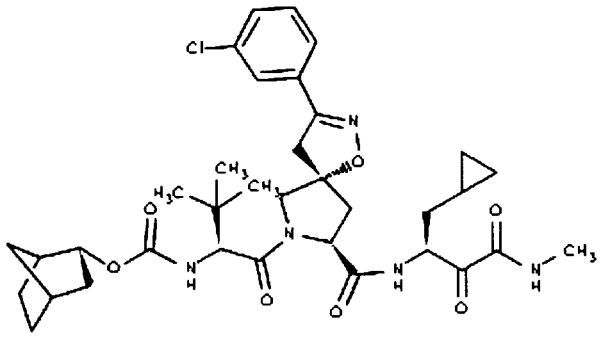
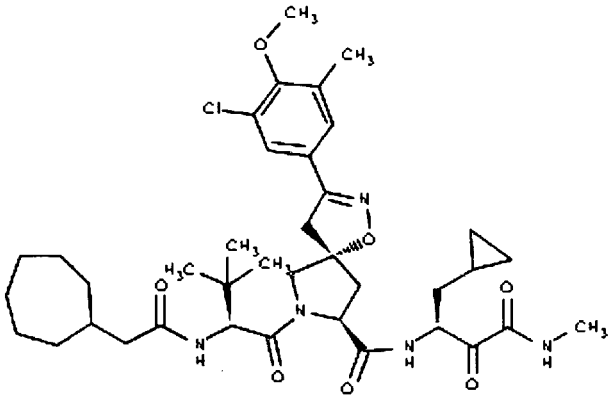
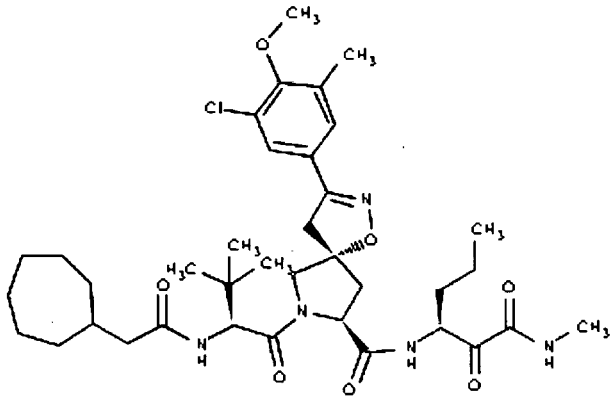
	737
	738
	739

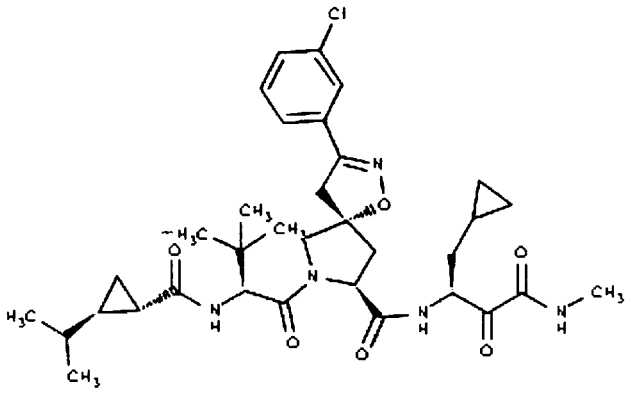
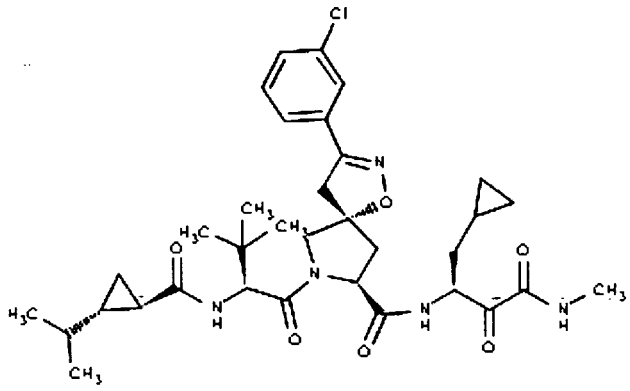
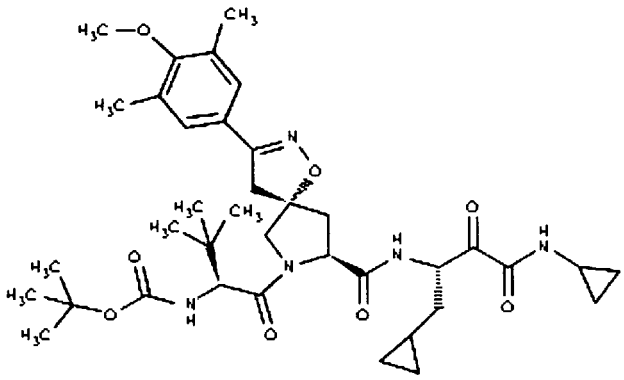
	740
	741
	742

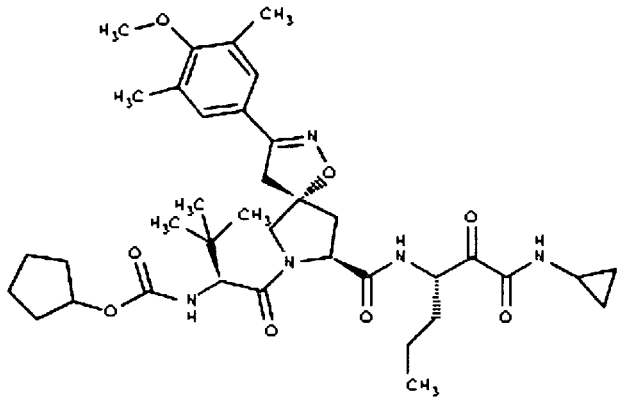
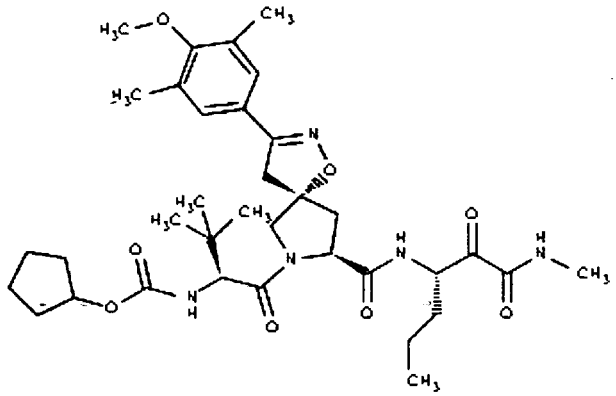
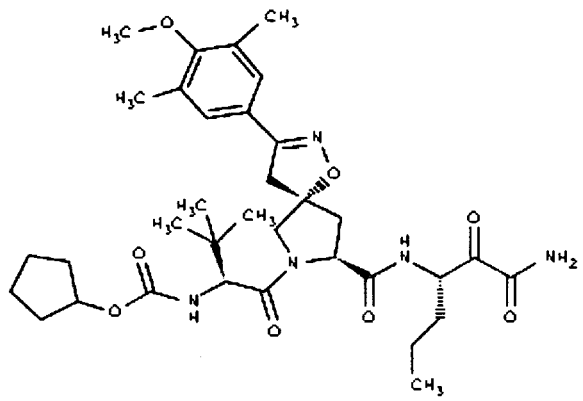
	743
	744
	745

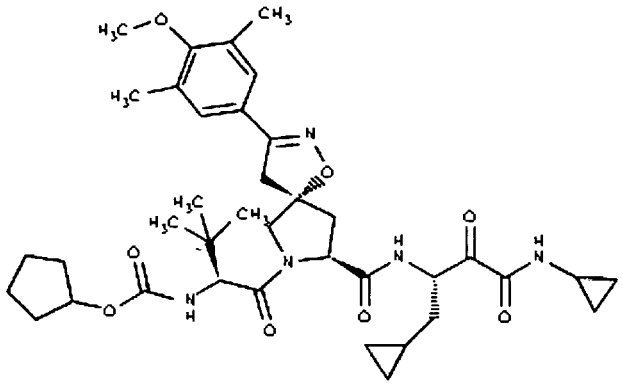
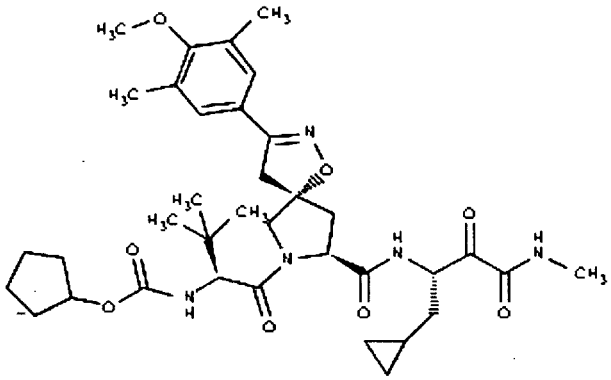
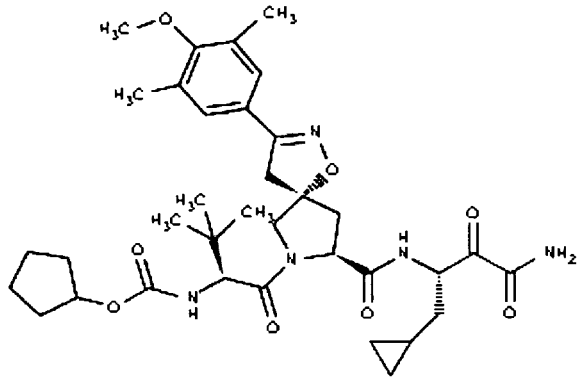
	746
	747
	748

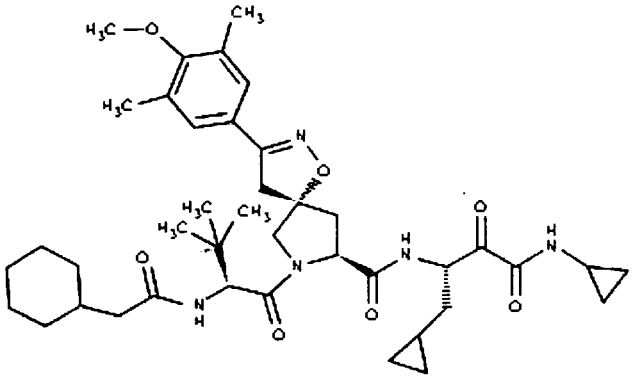
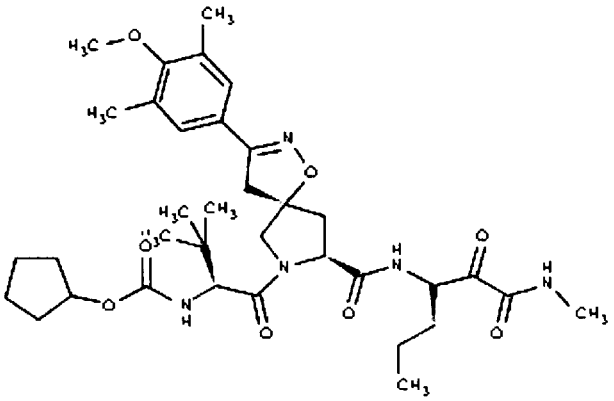
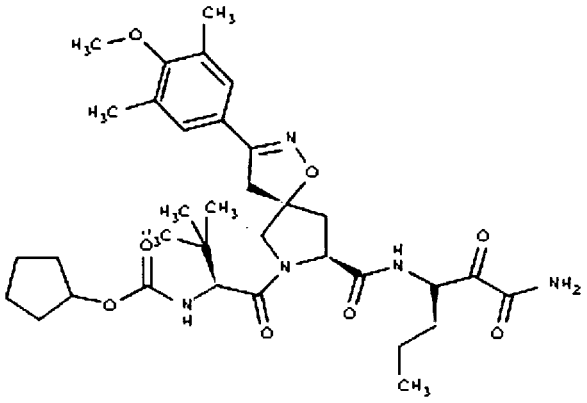
	749
	750
	751

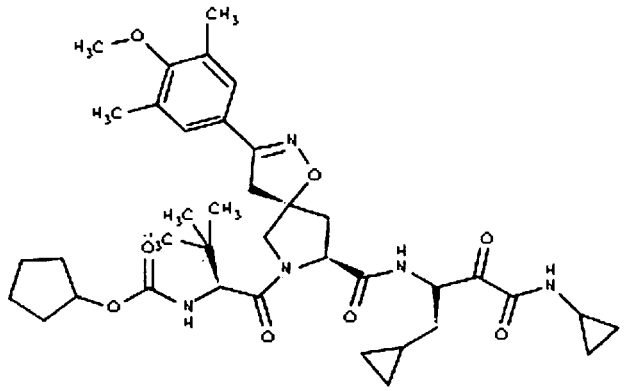
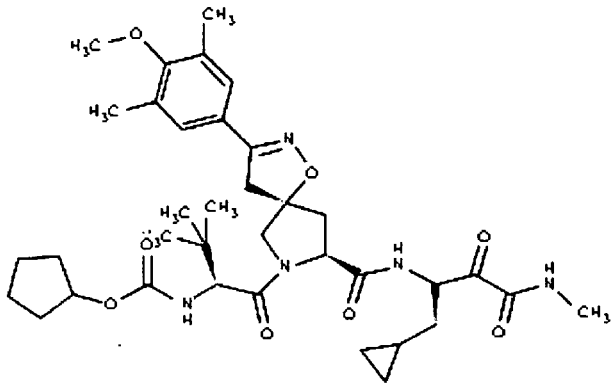
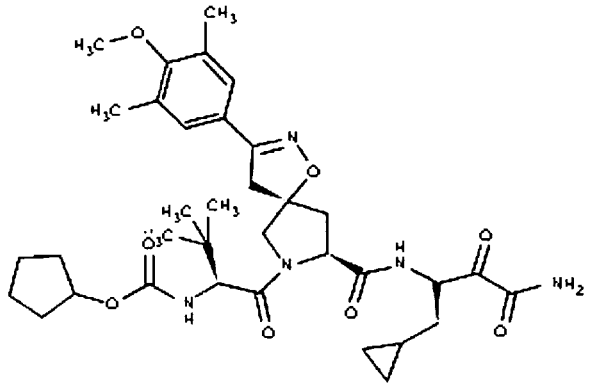
	752
	753
	754

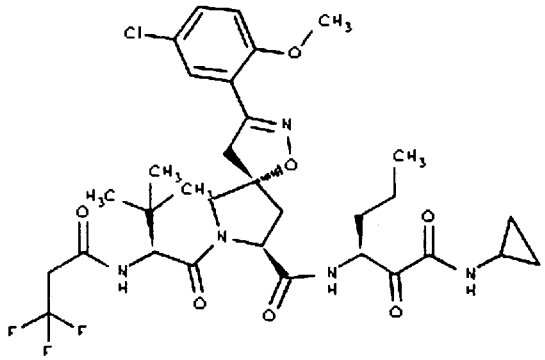
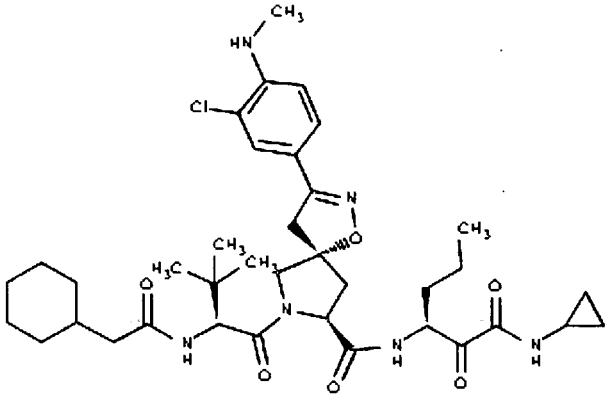
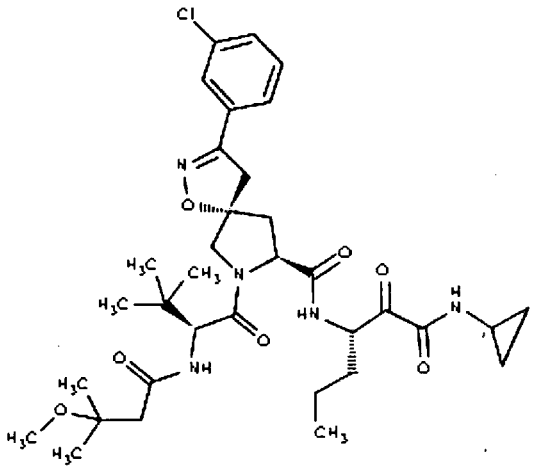
	755
	756
	757

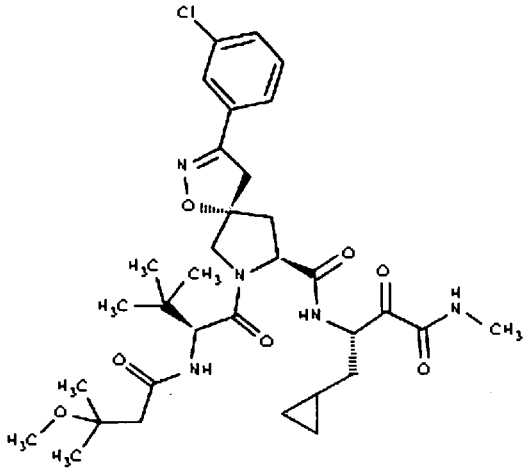
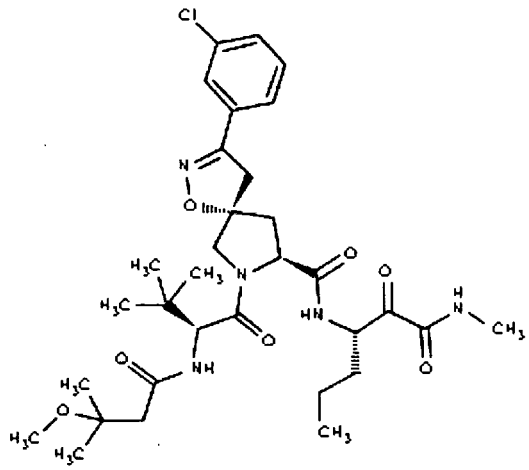
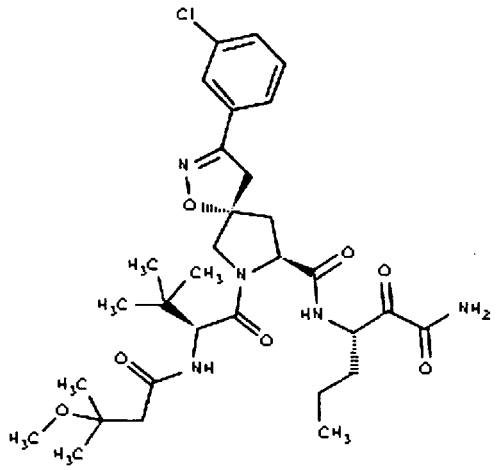
	758
	759
	760

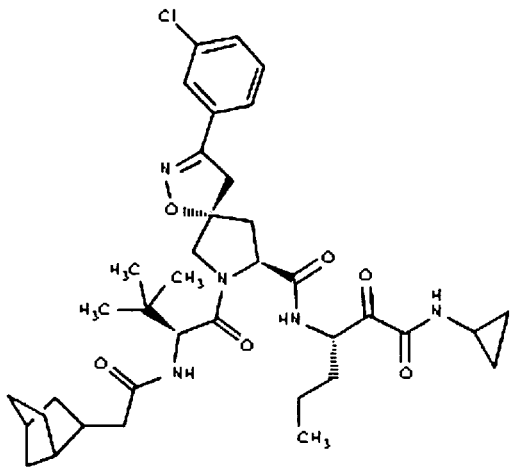
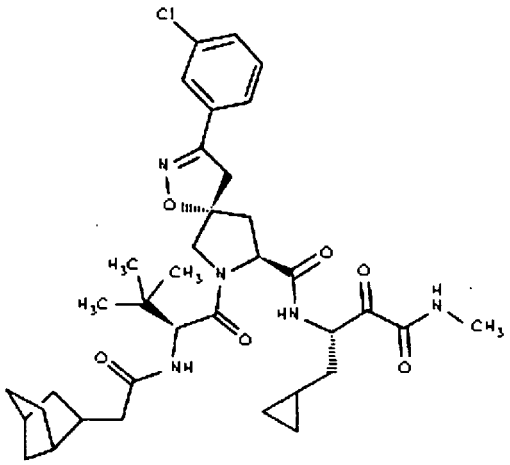
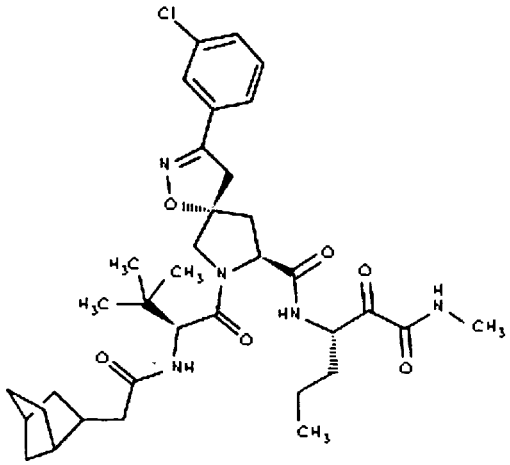
	761
	762
	763

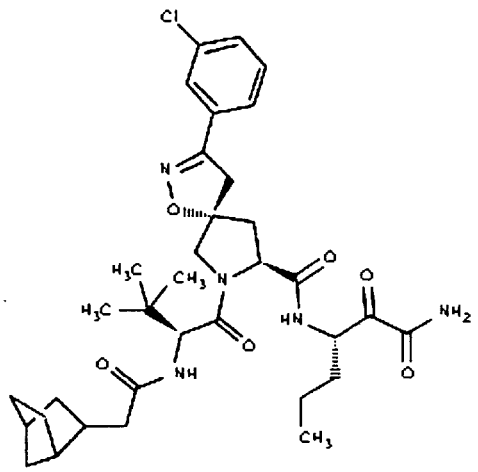
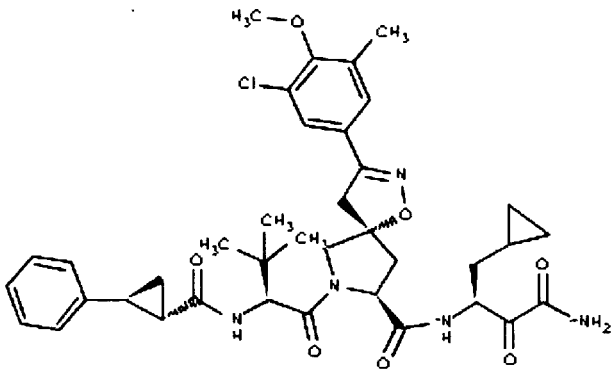
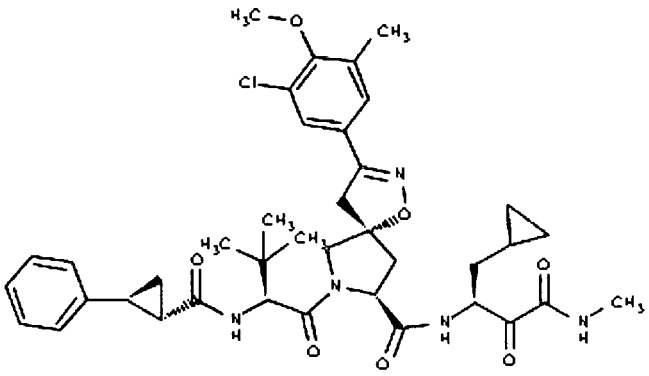
	764
	765
	766

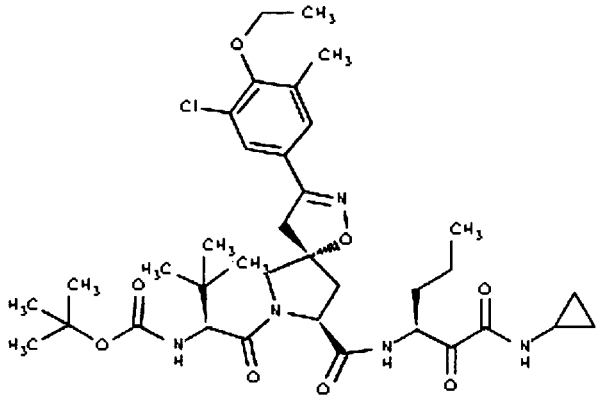
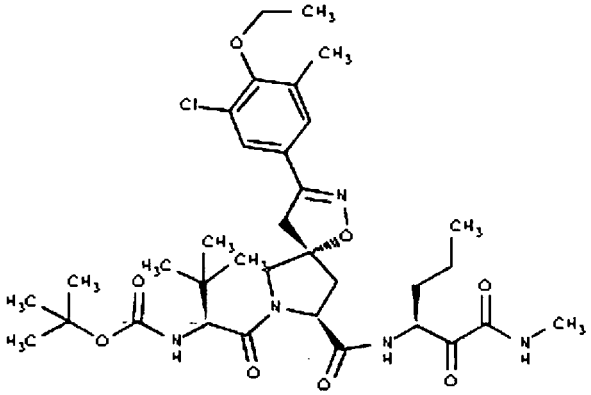
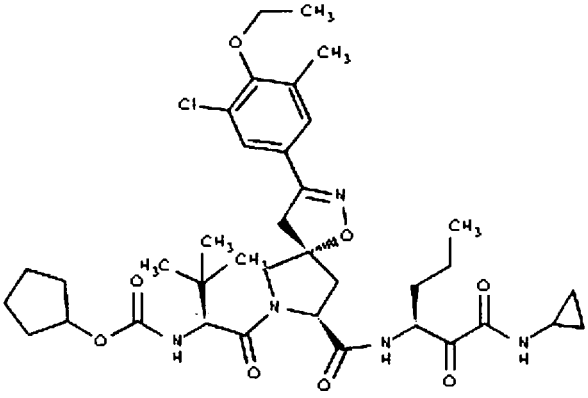
	767
	768
	769

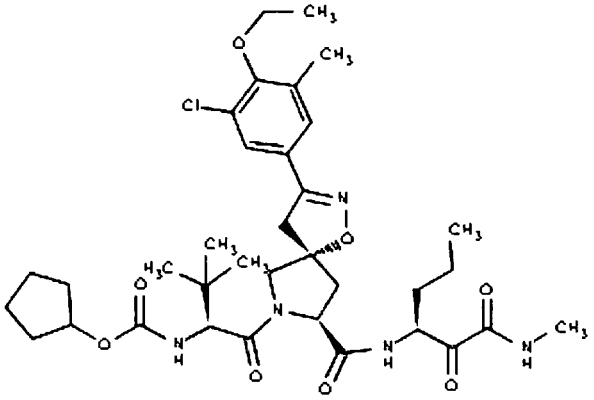
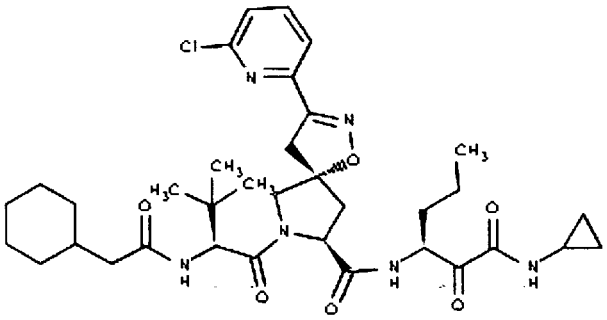
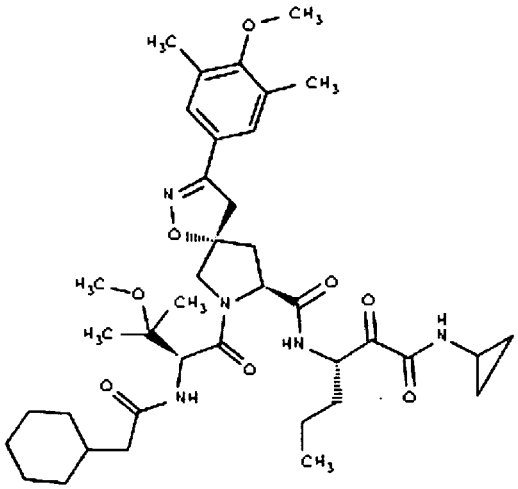
	770
	771
	772

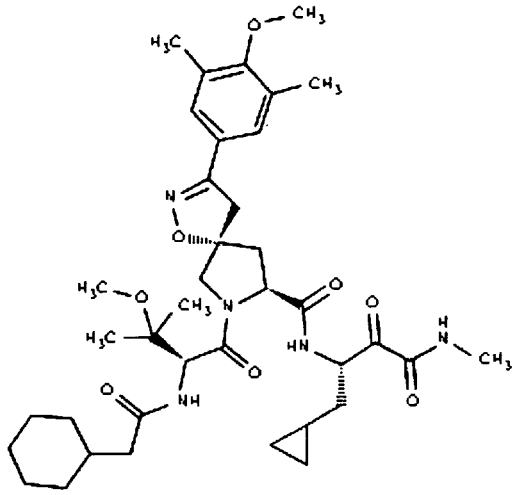
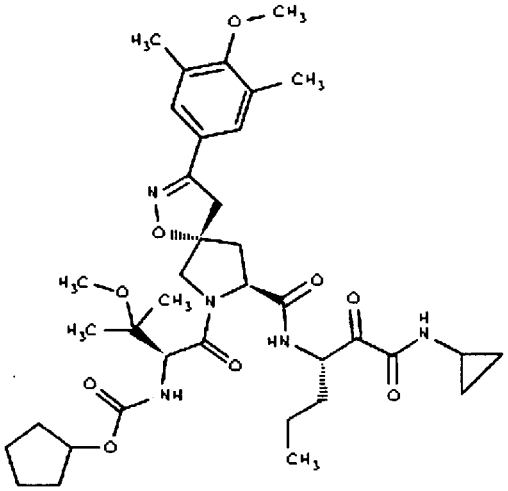
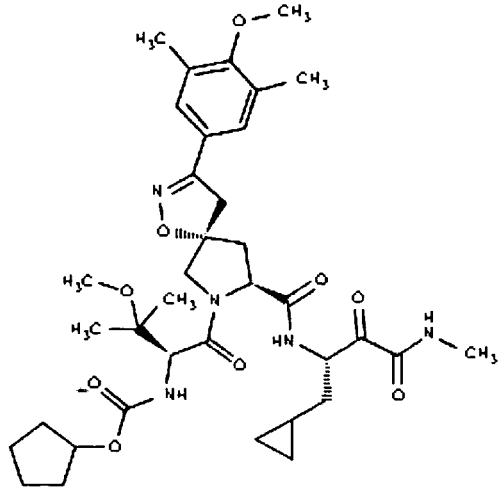
	773
	774
	775

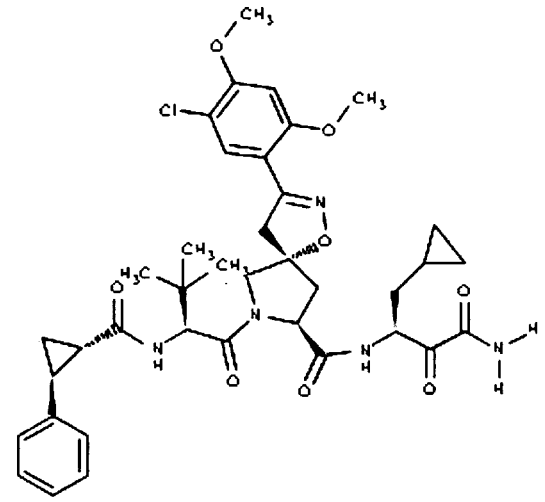
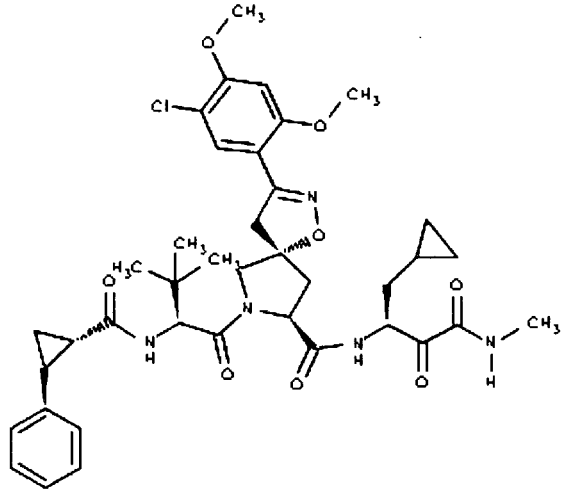
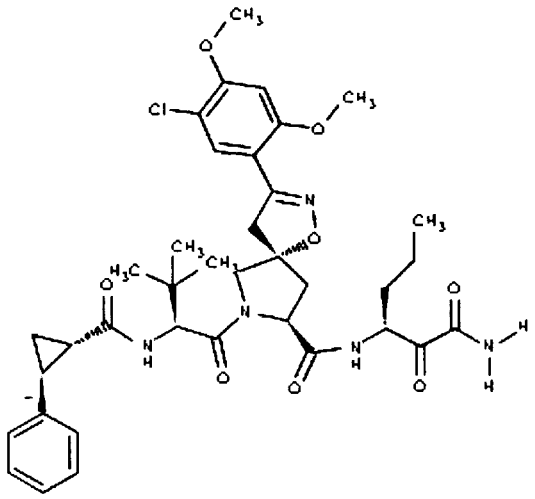
	776
	777
	778

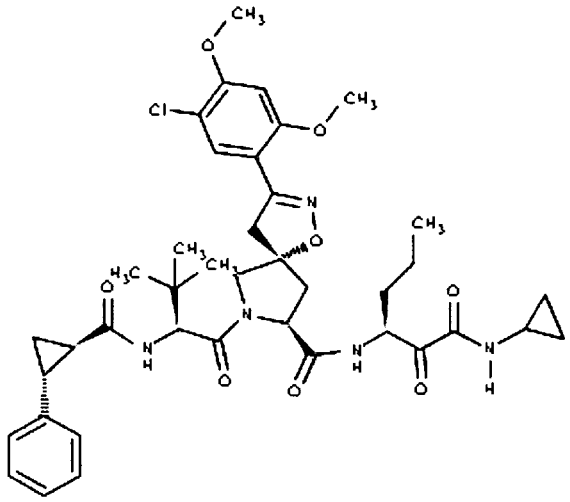
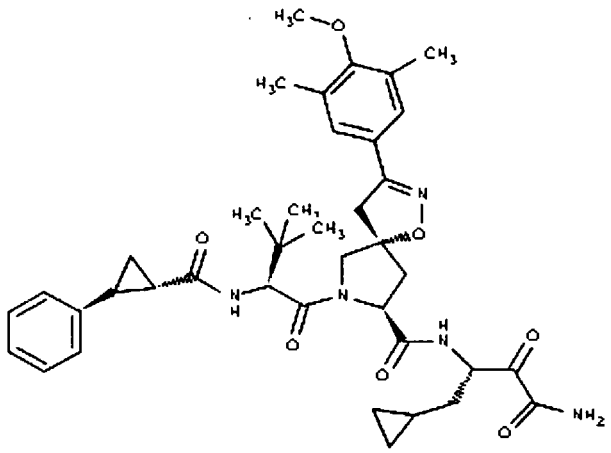
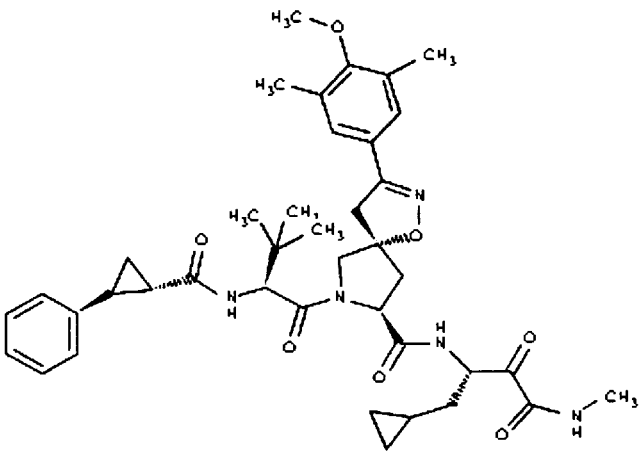
	779
	780
	781

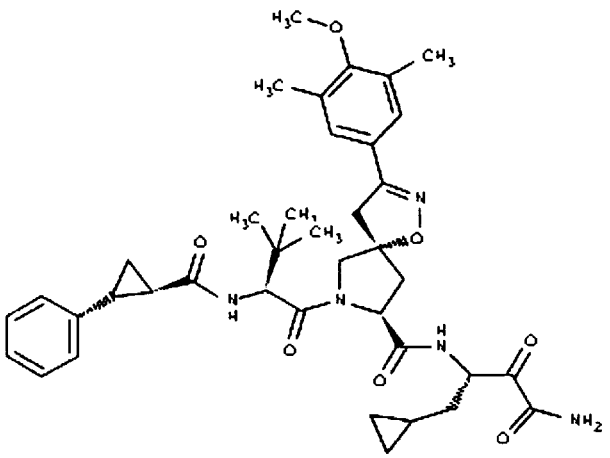
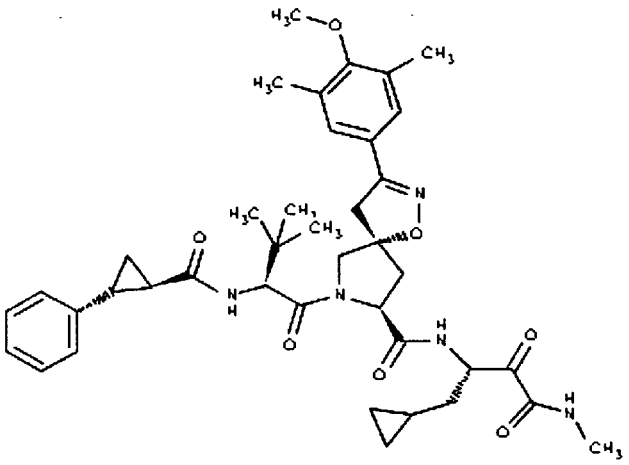
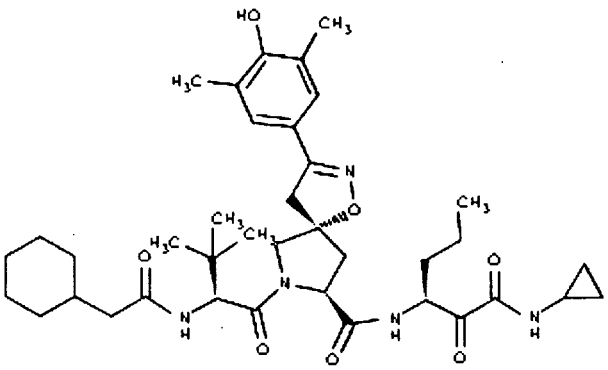
	782
	783
	784

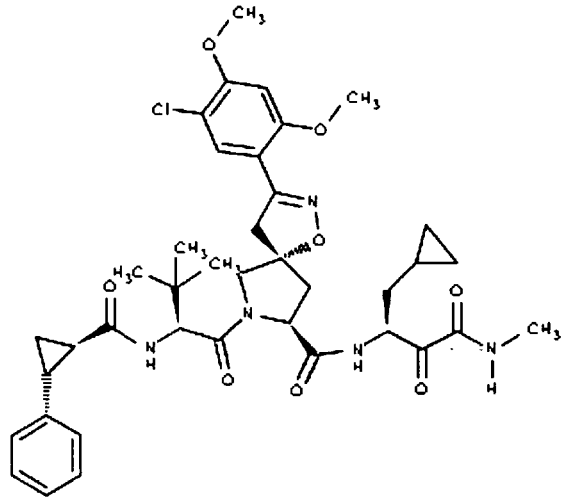
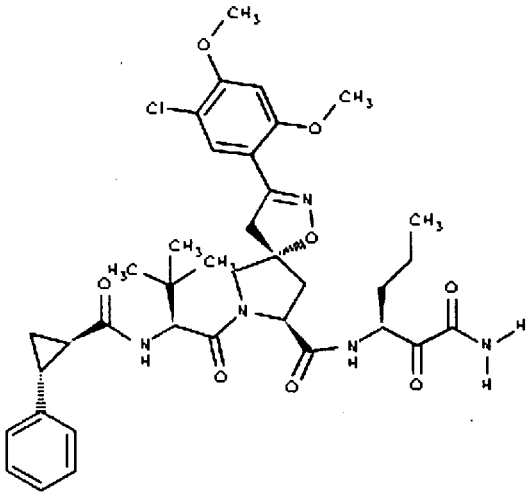
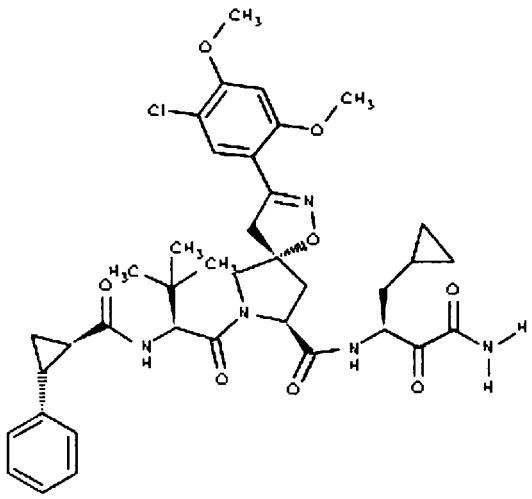
	785
	786
	787

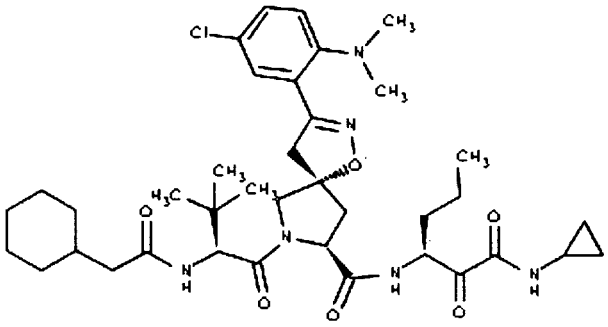
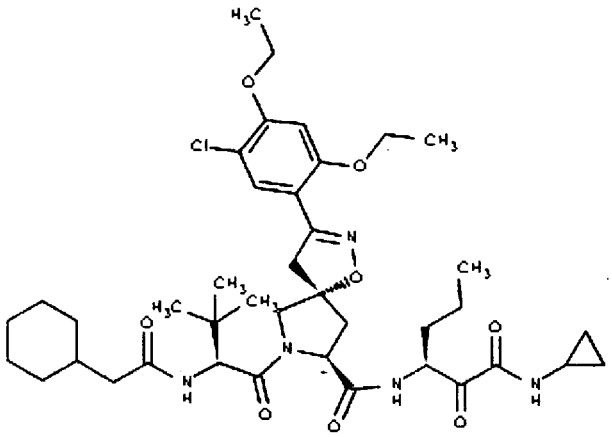
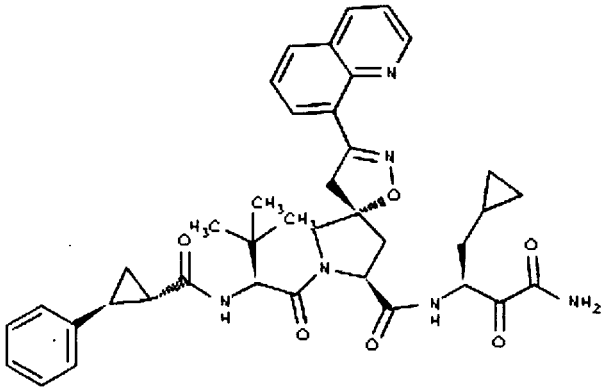
	788
	789
	790

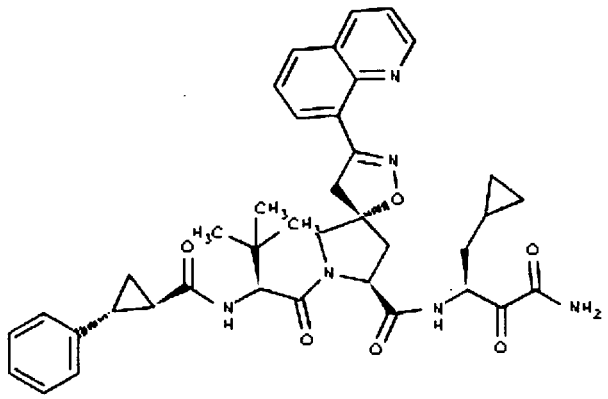
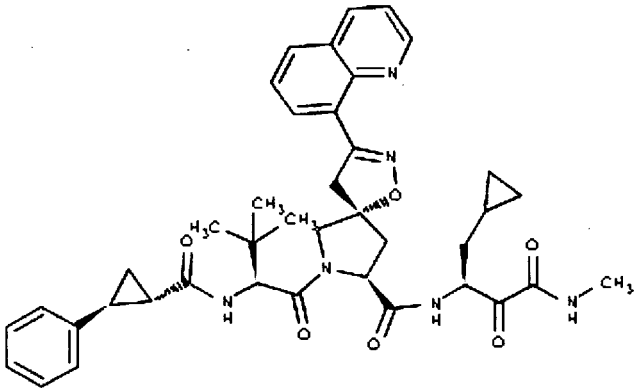
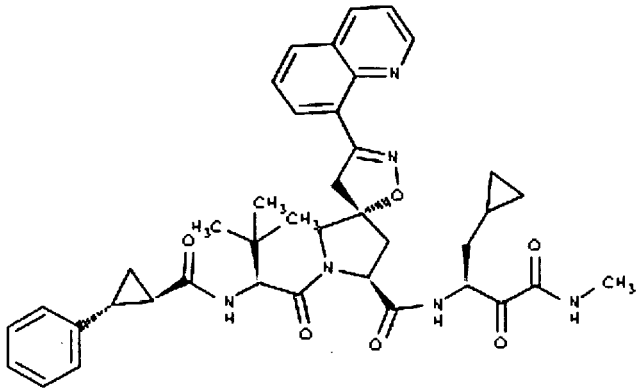
	791
	792
	793

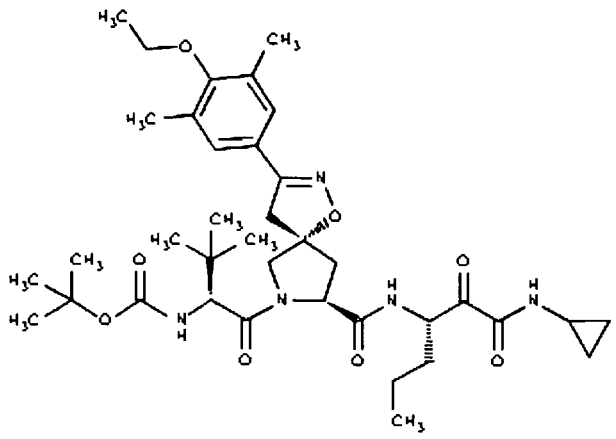
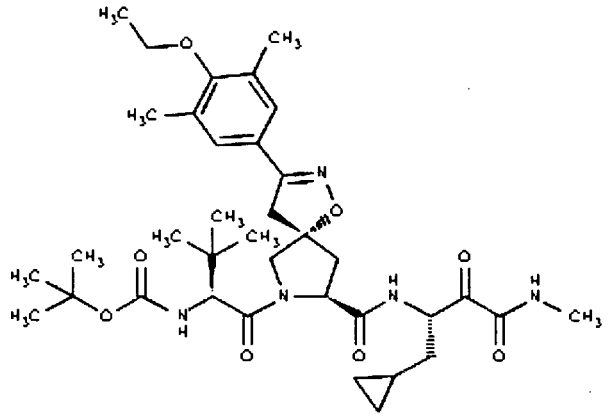
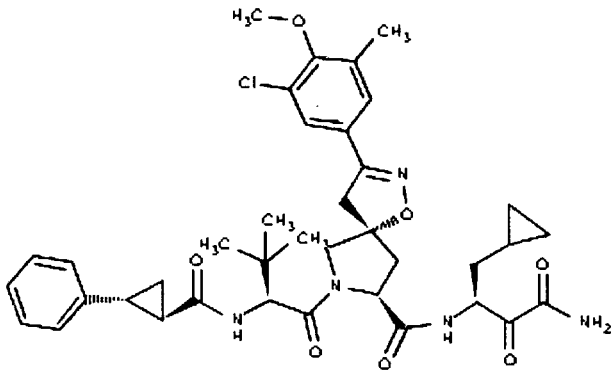
	794
	795
	796

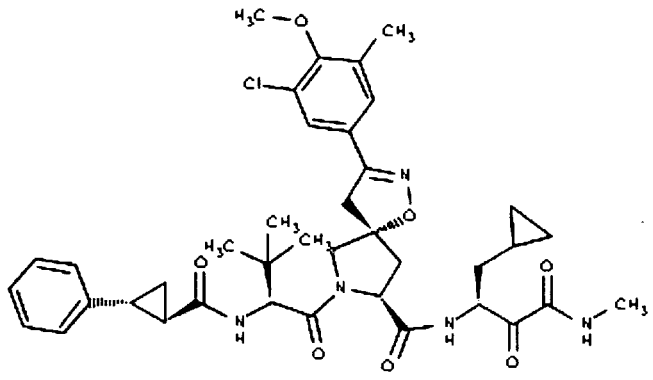
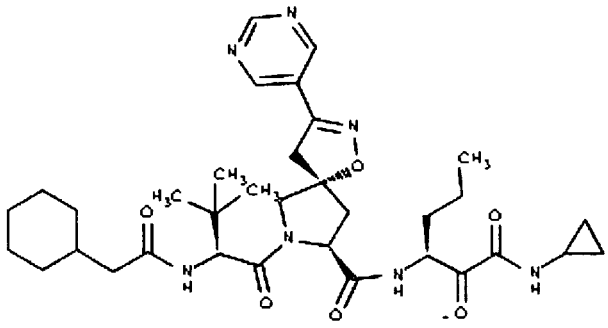
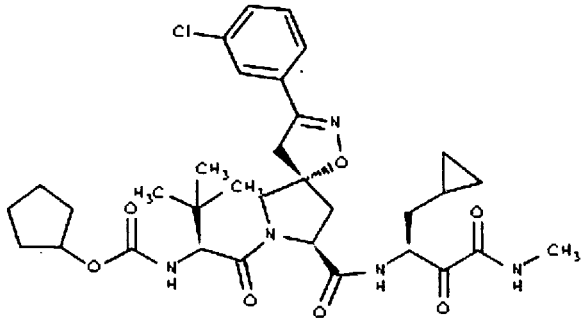
	797
	798
	799

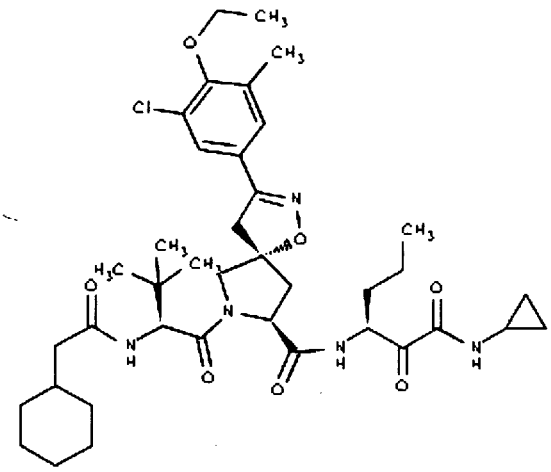
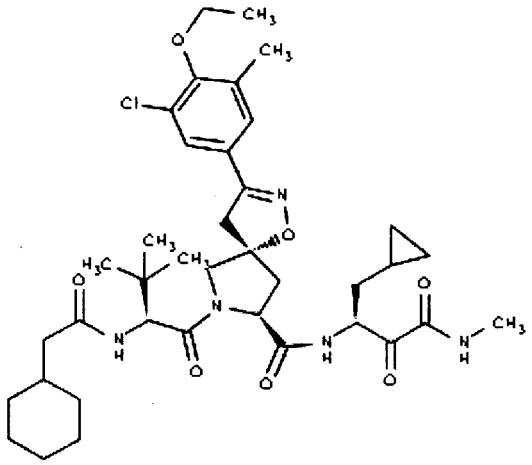
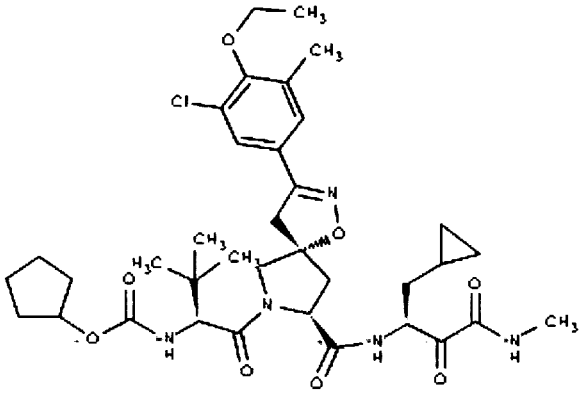
	800
	801
	802

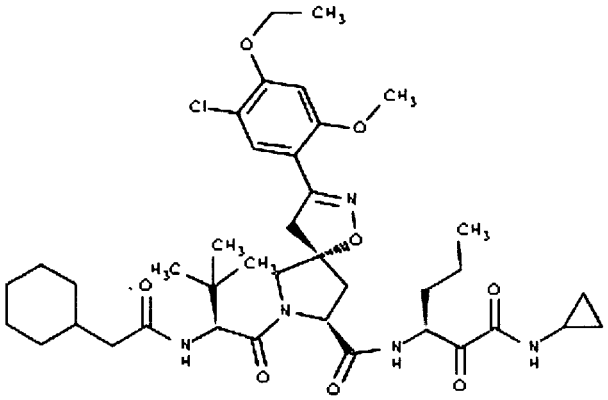
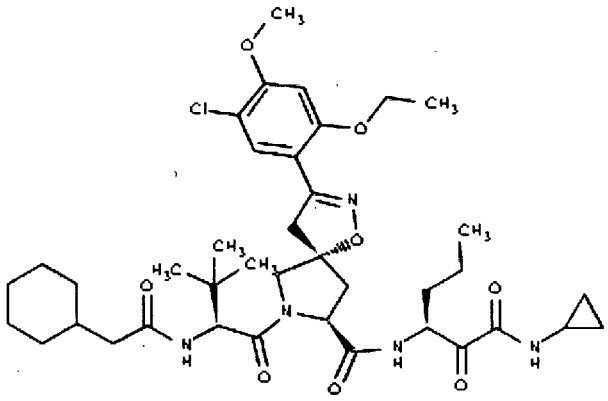
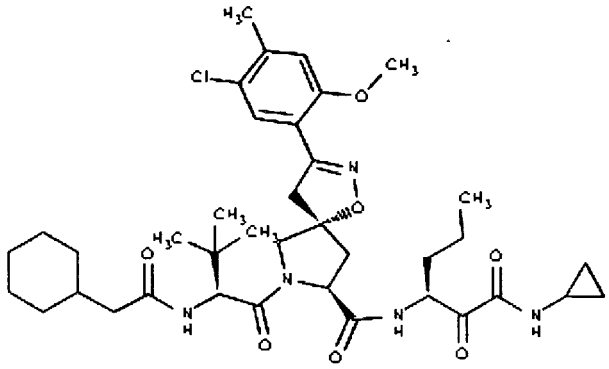
	803
	804
	805

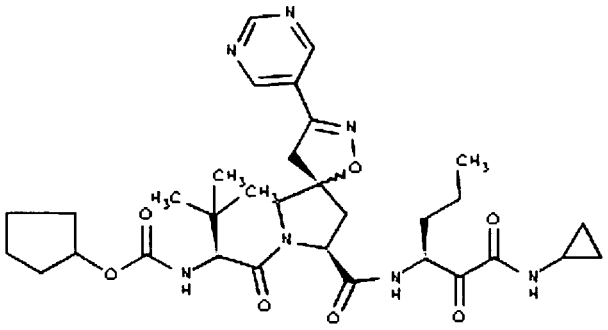
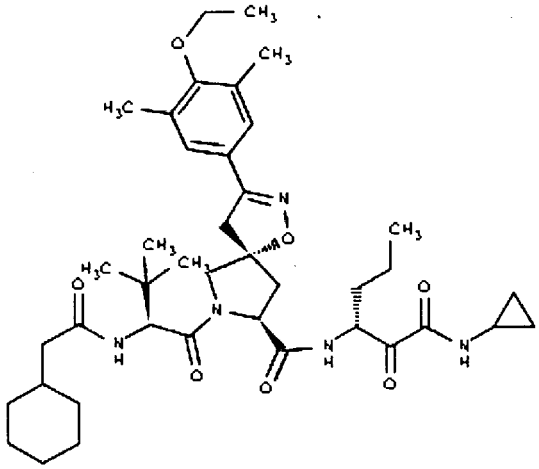
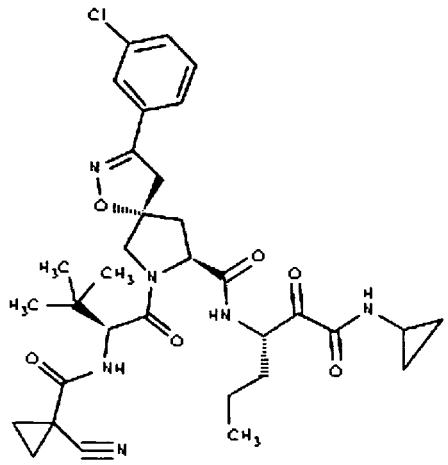
	806
	807
	808

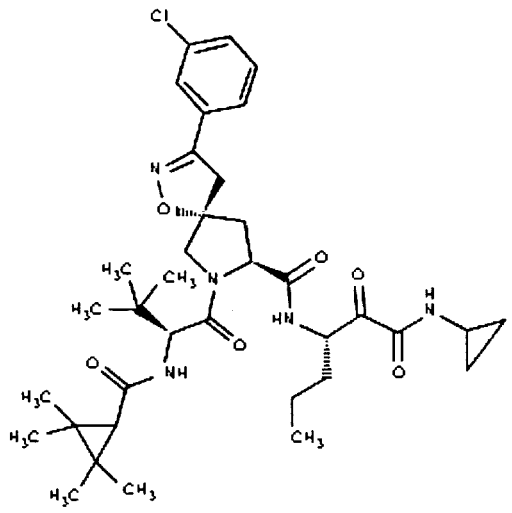
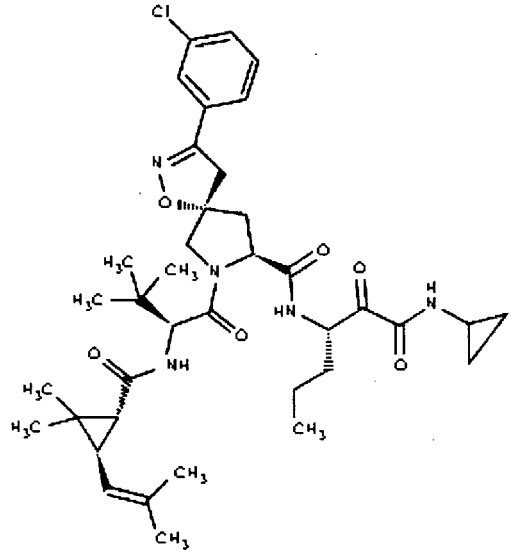
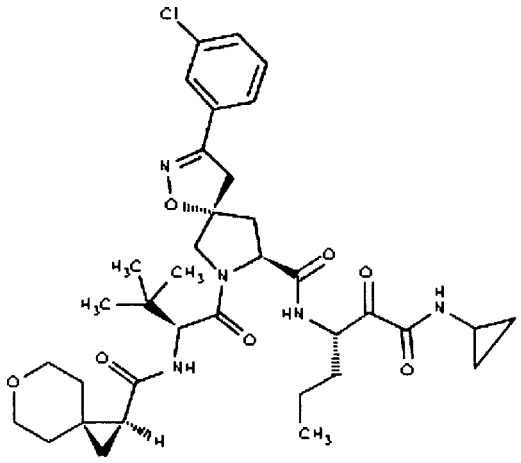
	809
	810
	811

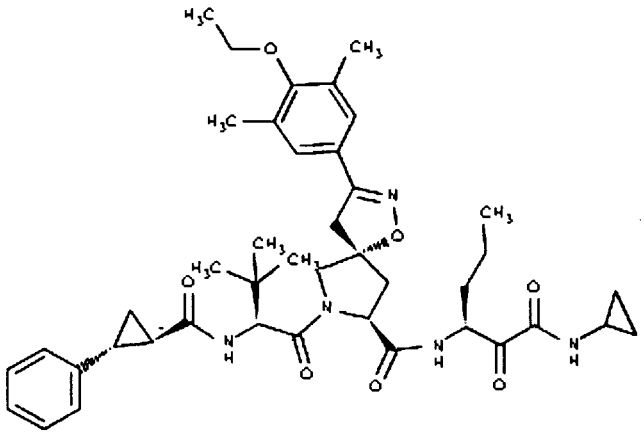
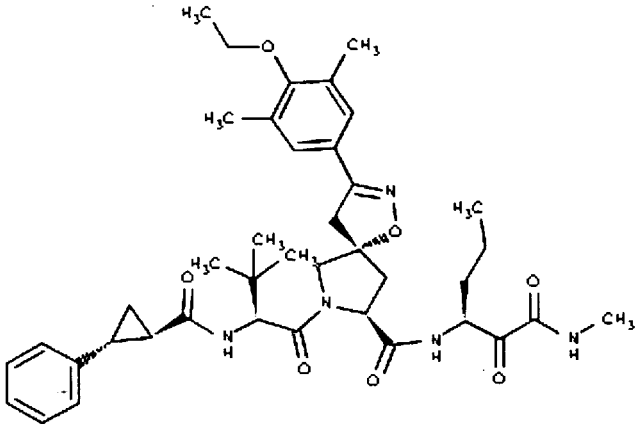
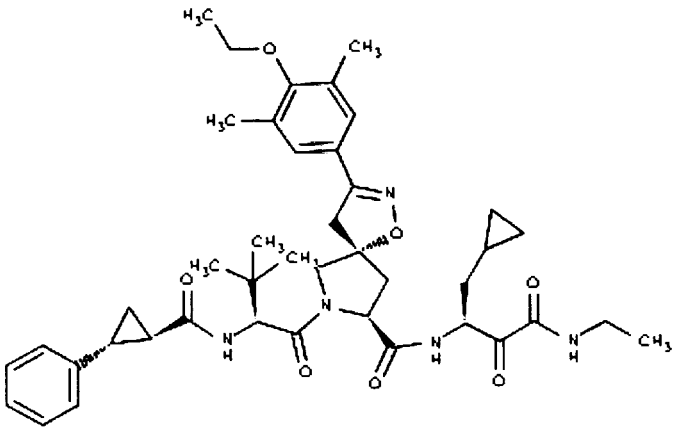
	812
	813
	814

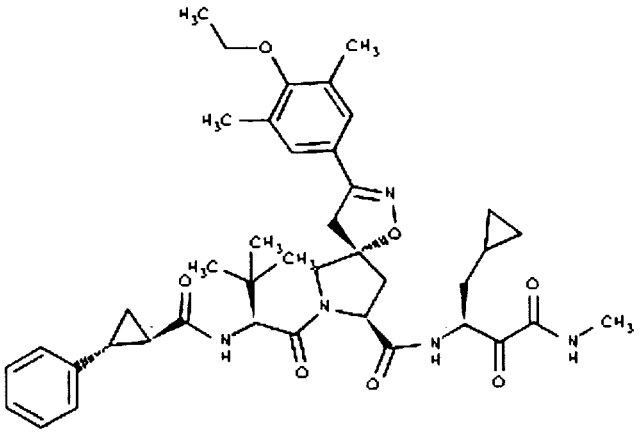
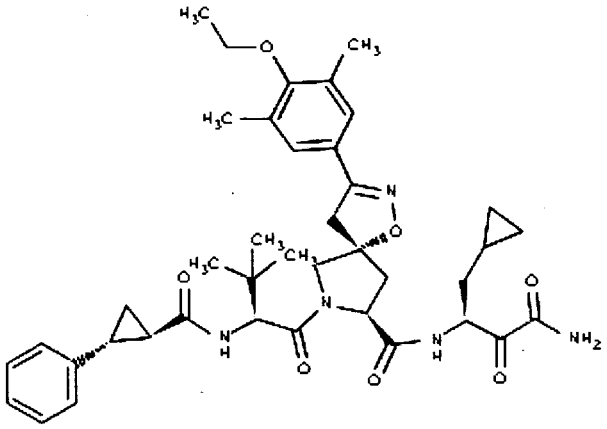
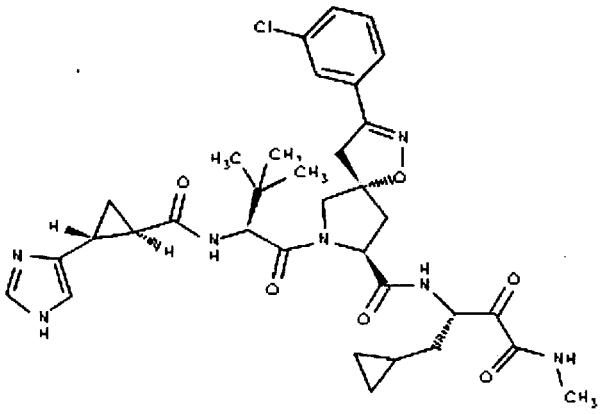
	815
	816
	817

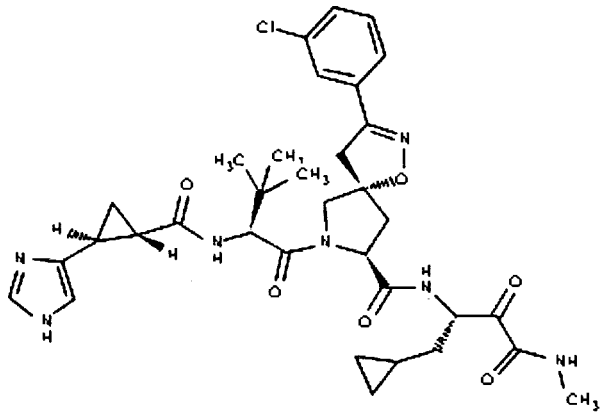
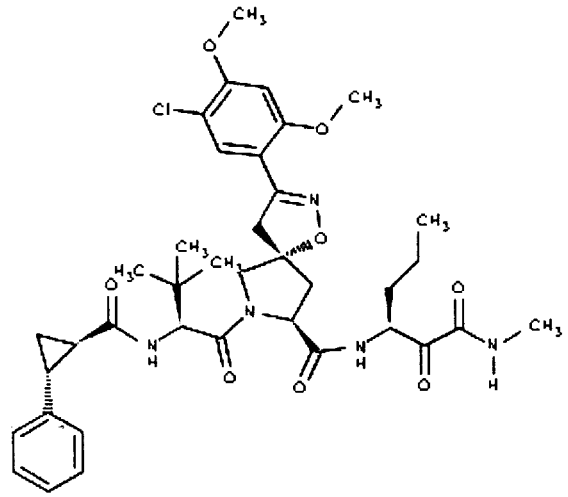
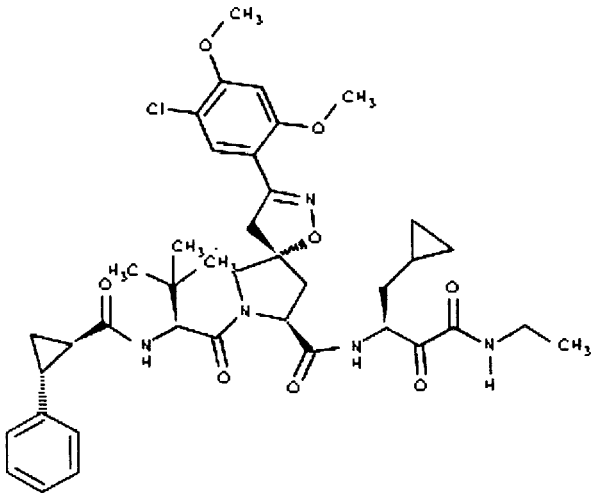
	818
	819
	820

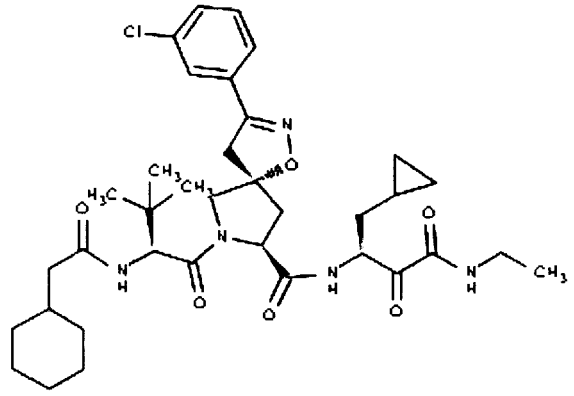
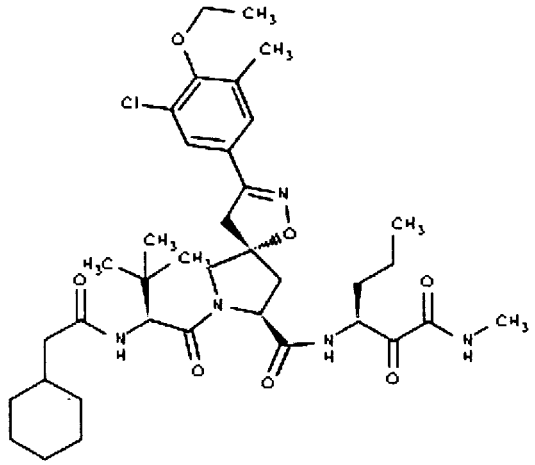
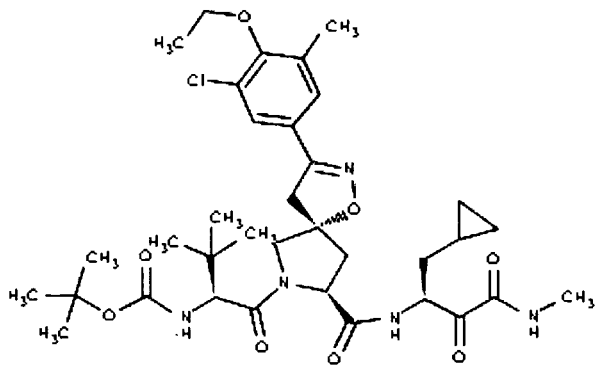
	821
	822
	823

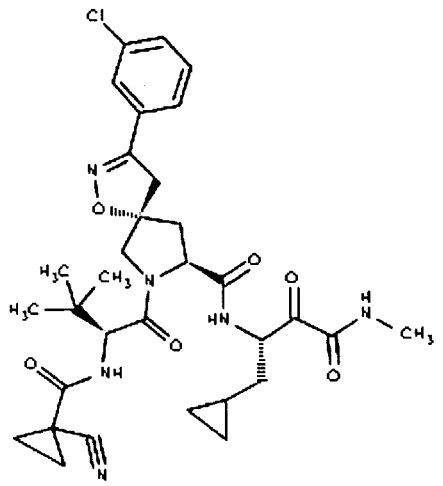
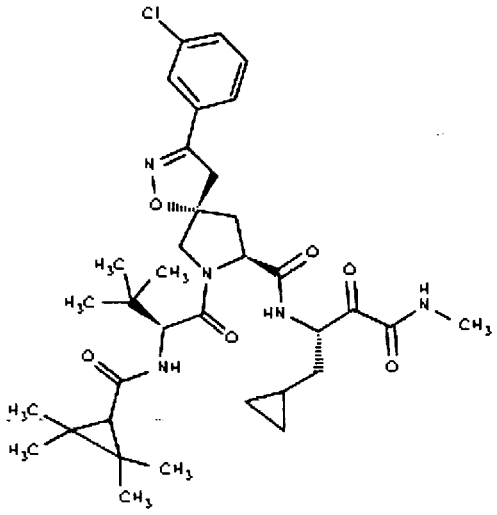
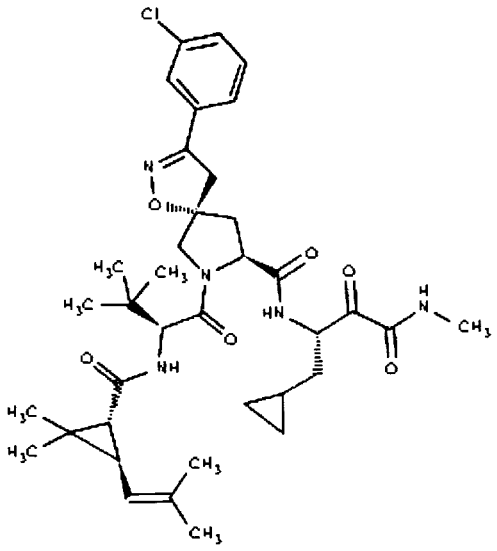
	824
	825
	826

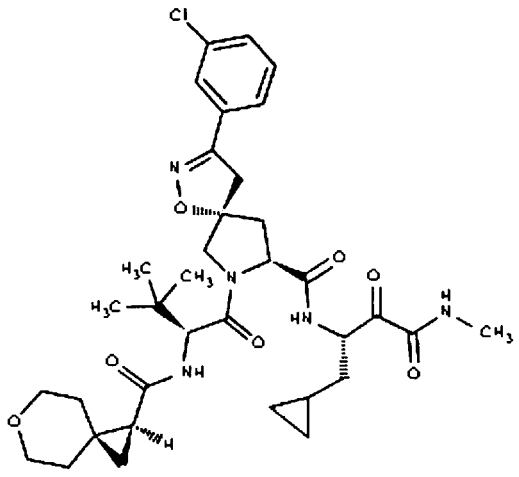
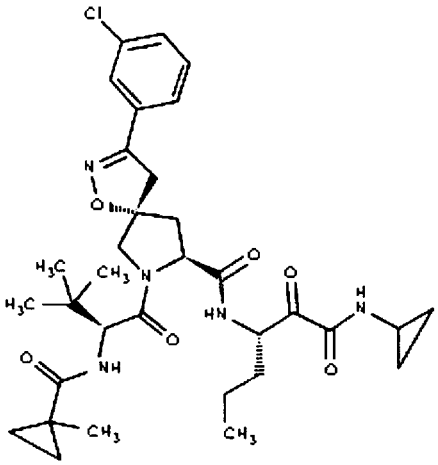
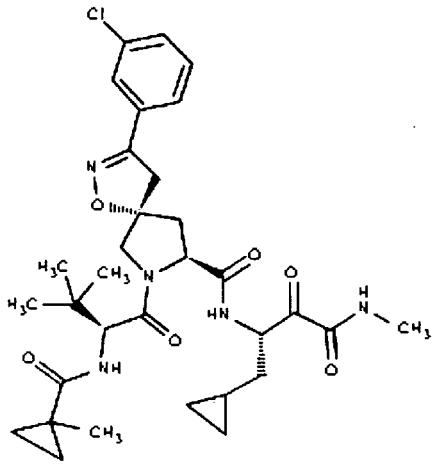
	827
	828
	829

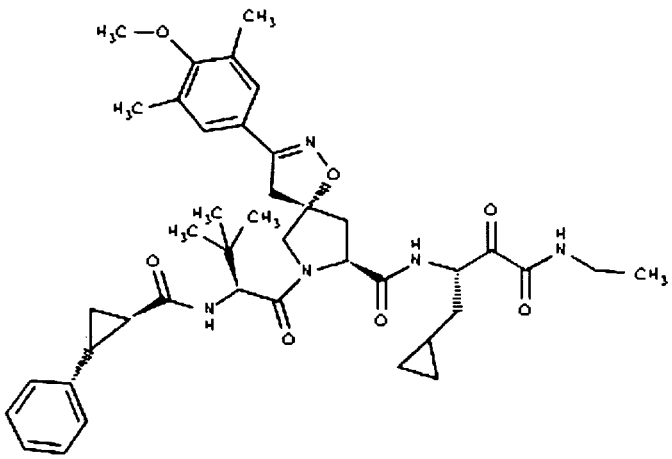
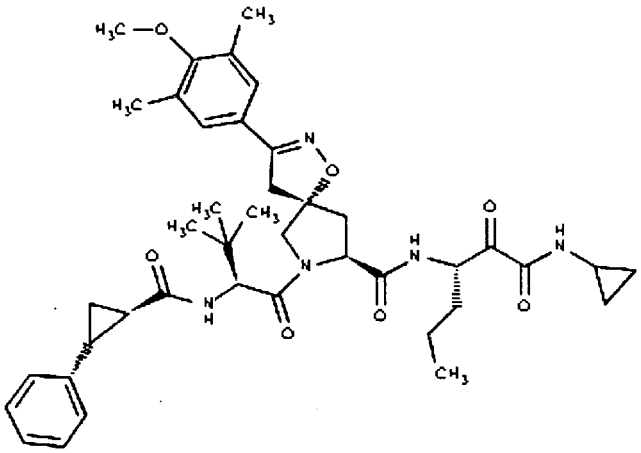
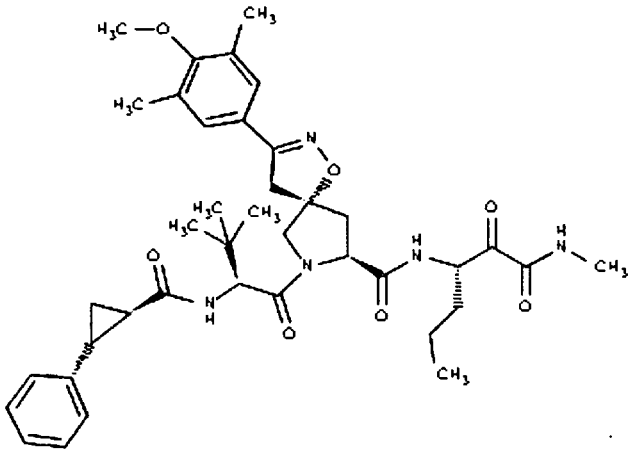
	830
	831
	832

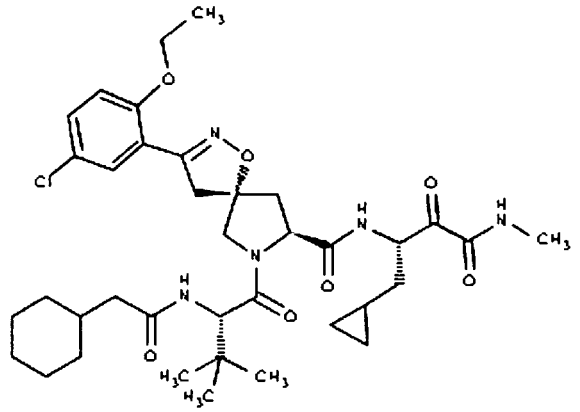
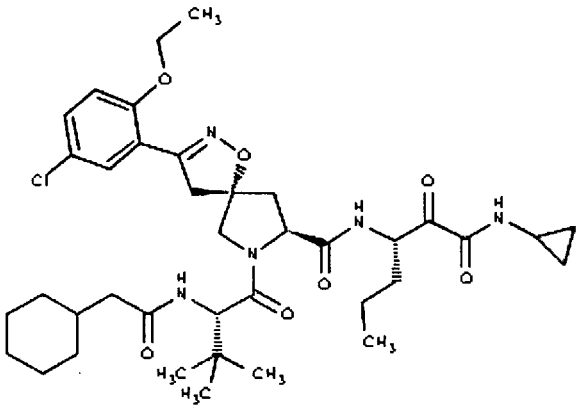
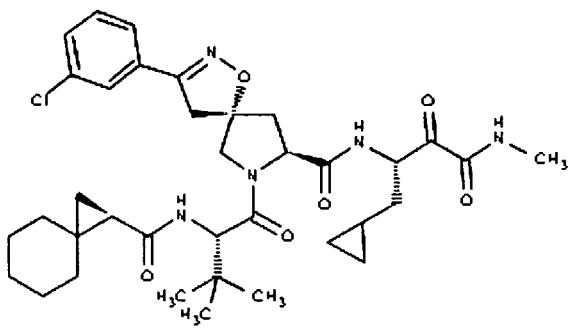
	833
	834
	835

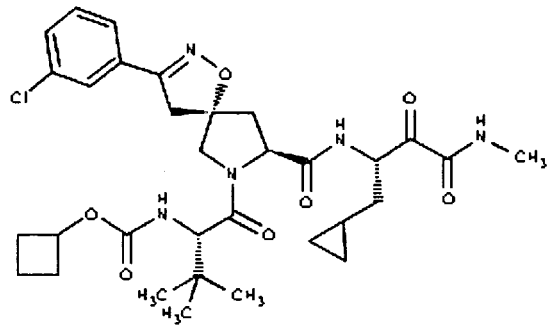
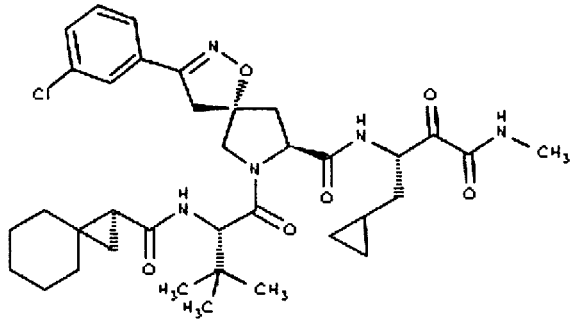
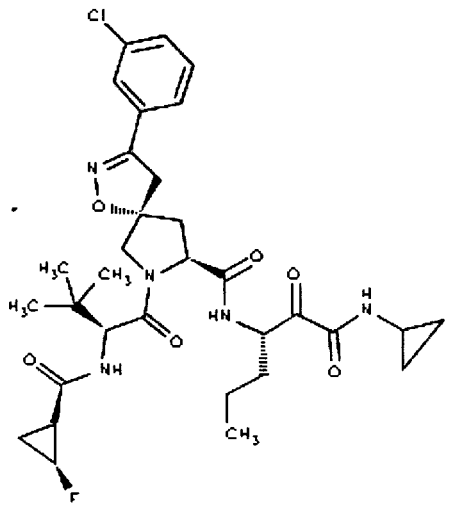
	836
	837
	838

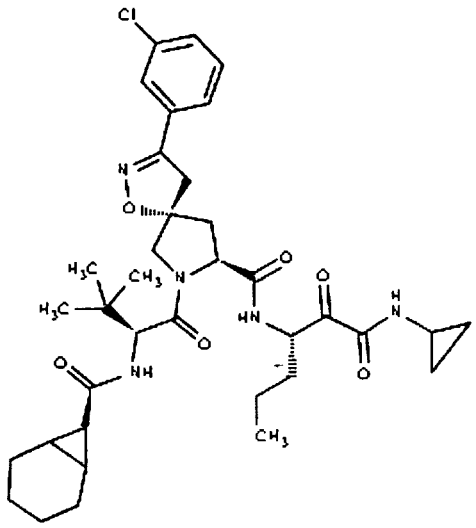
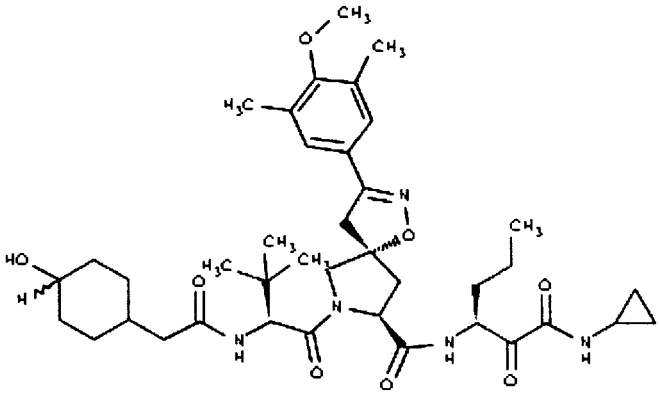
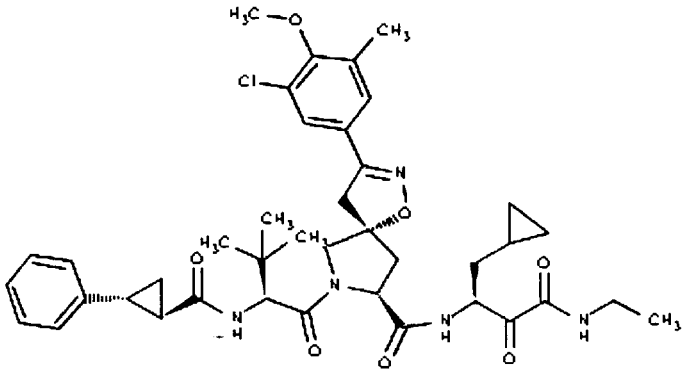
	839
	840
	841

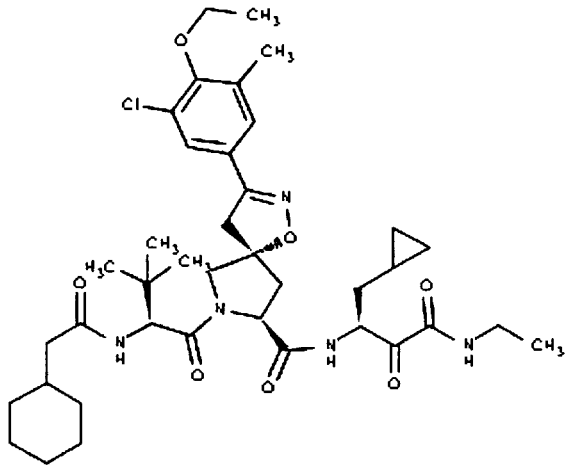
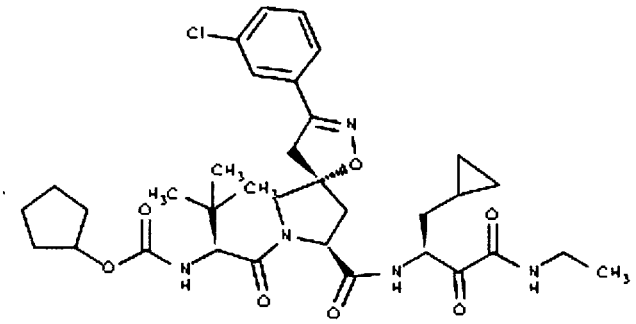
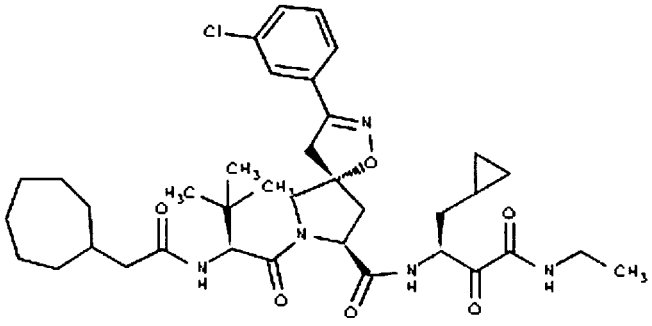
	842
	843
	844

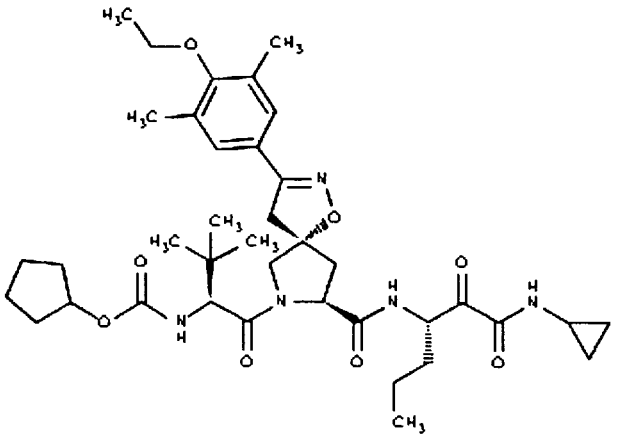
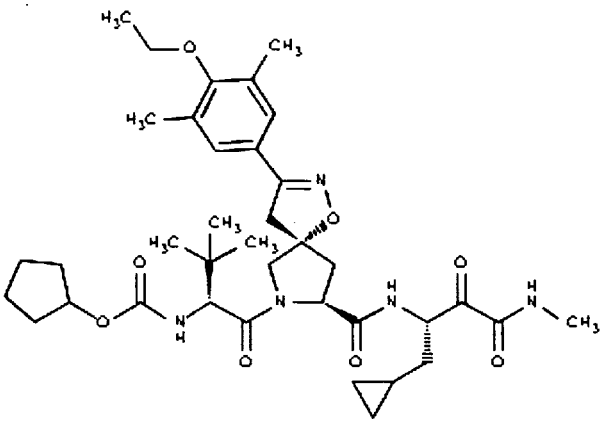
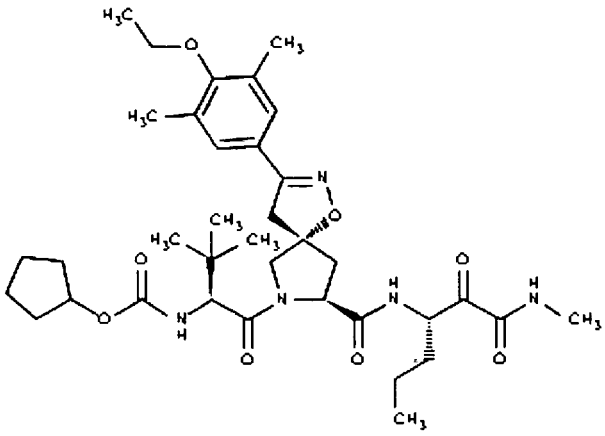
	845
	846
	847

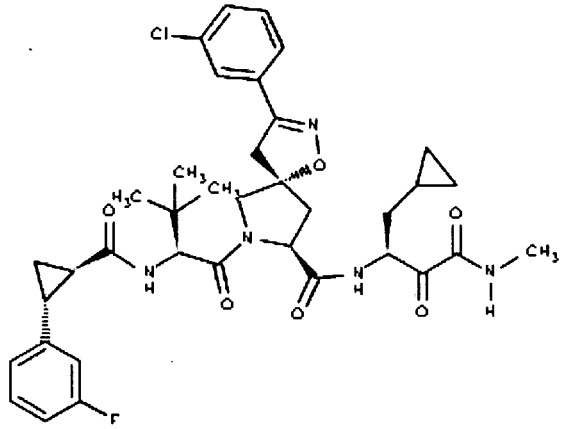
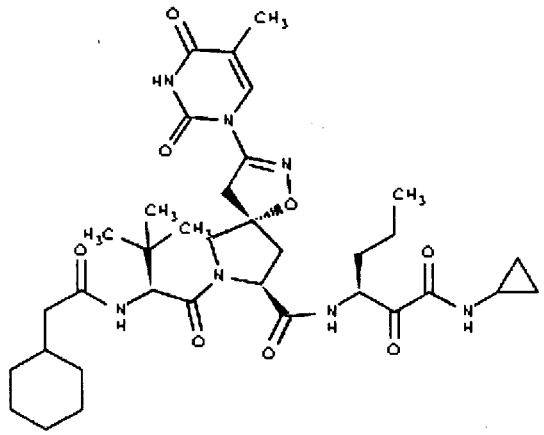
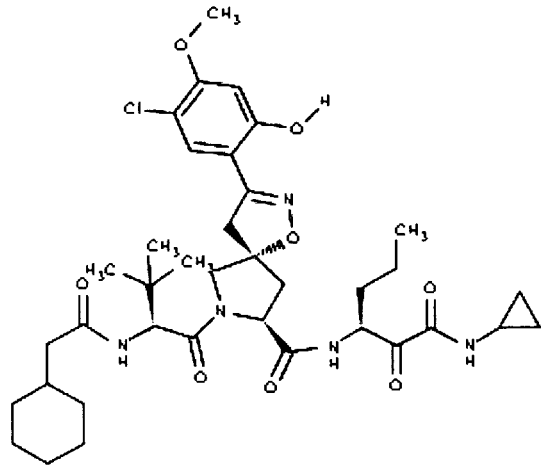
	848
	849
	850

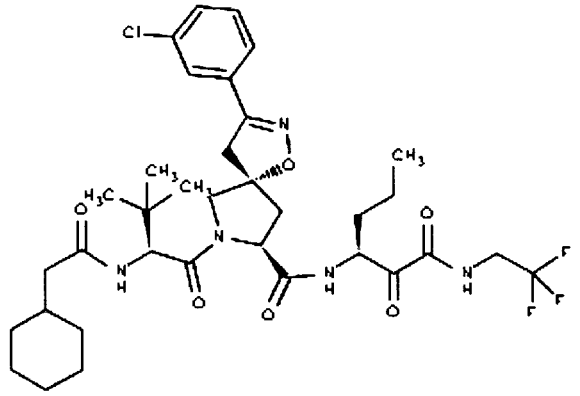
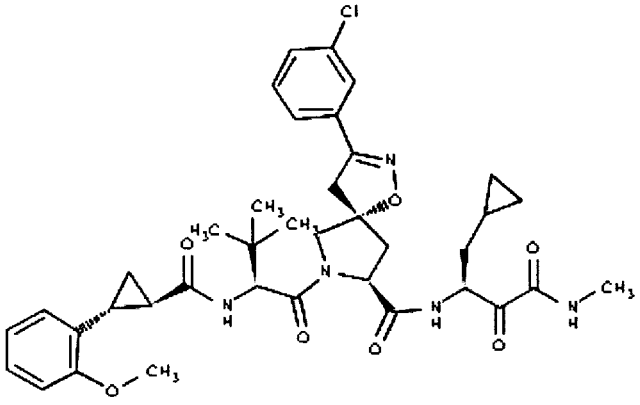
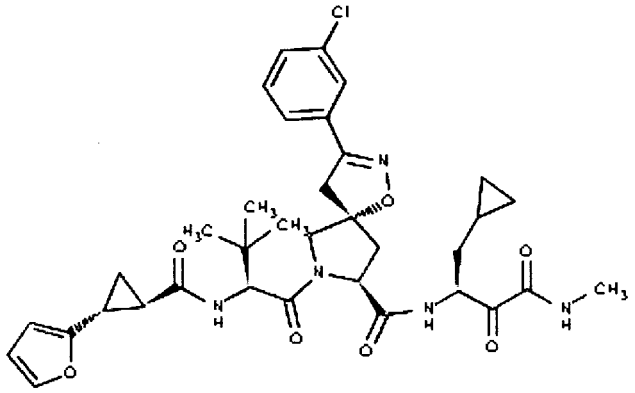
	851
	852
	853

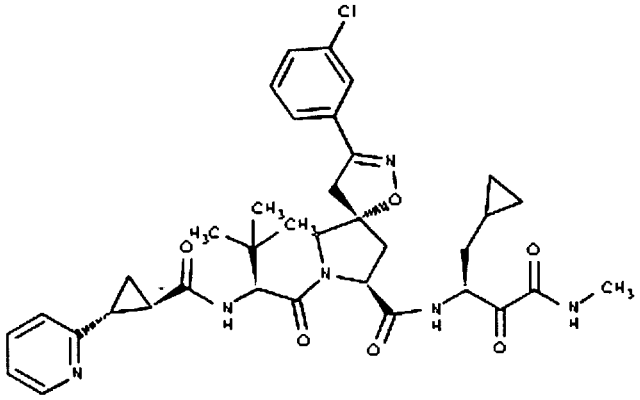
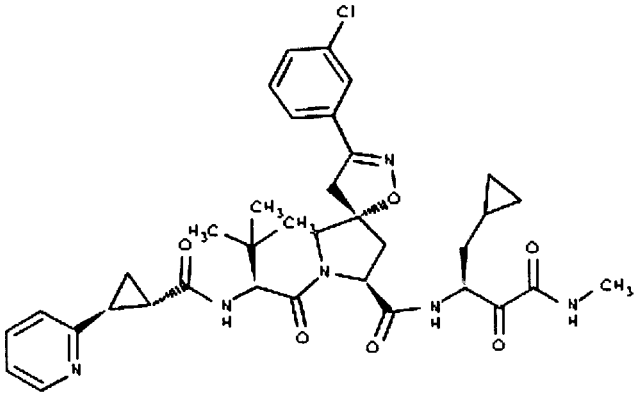
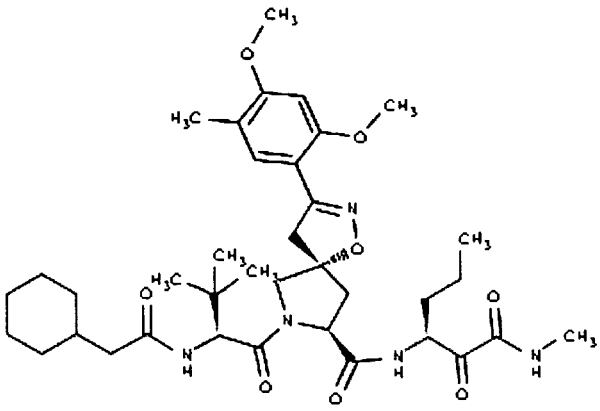
	854
	855
	856

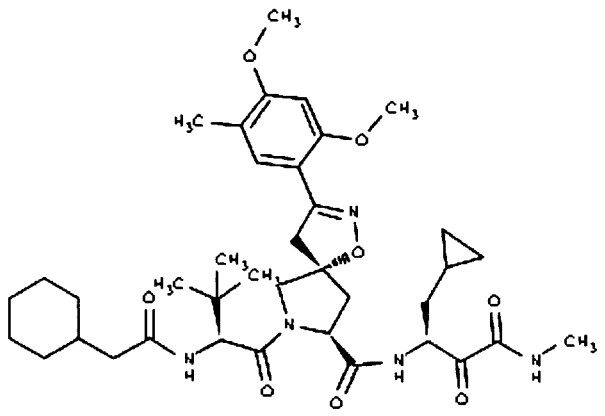
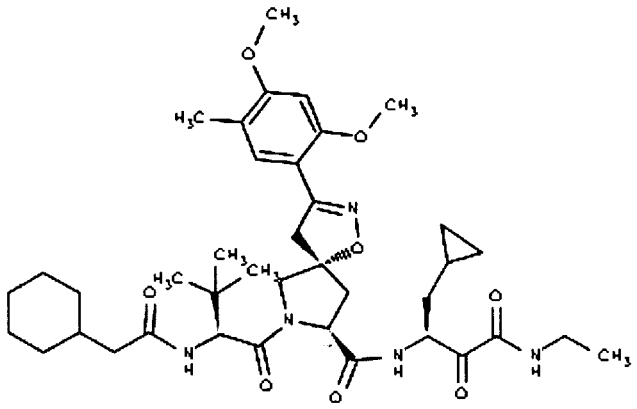
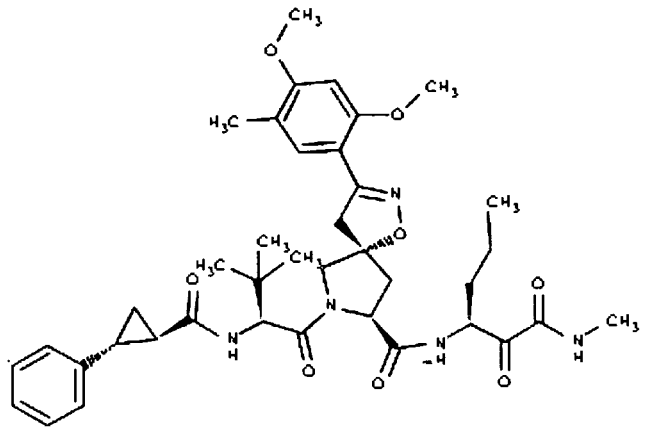
	857
	858
	859

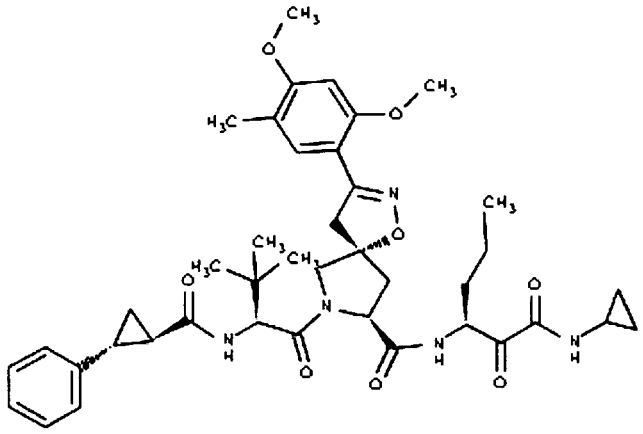
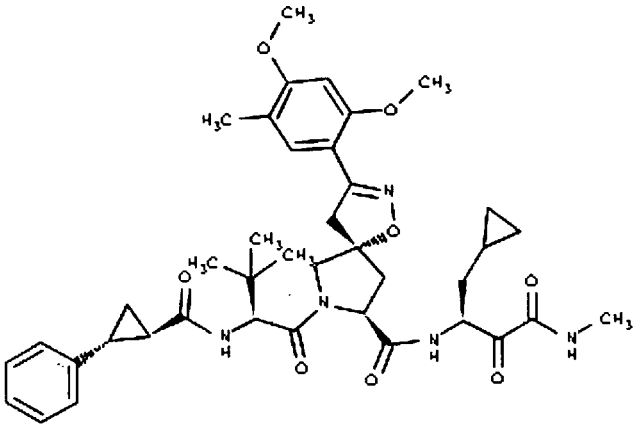
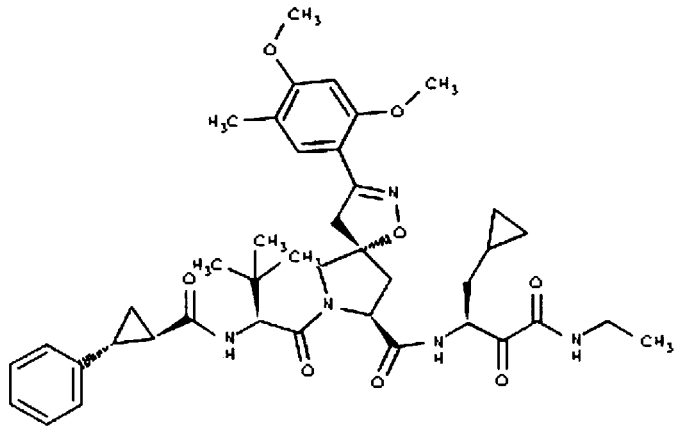
	860
	861
	862

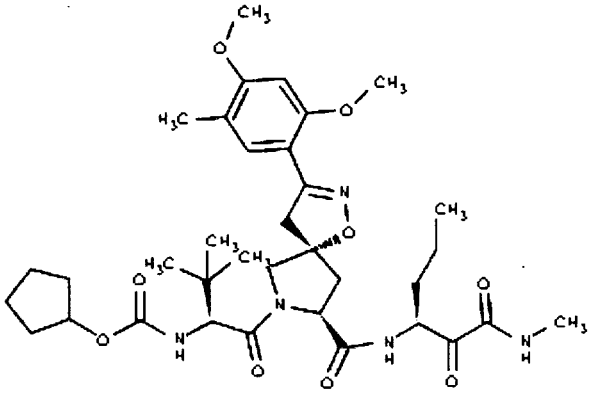
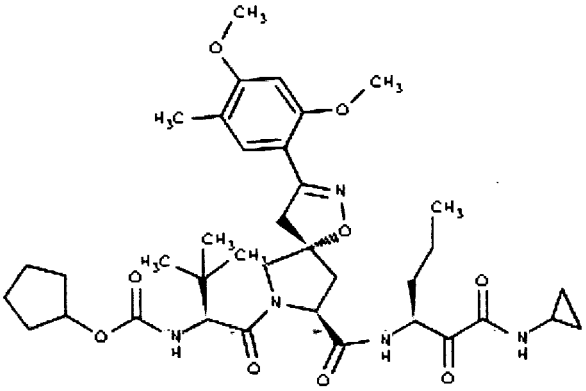
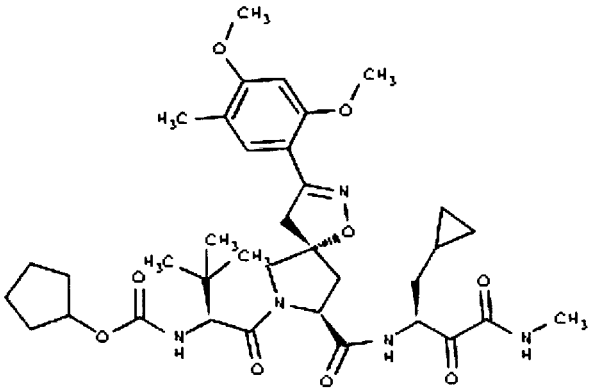
	863
	864
	865

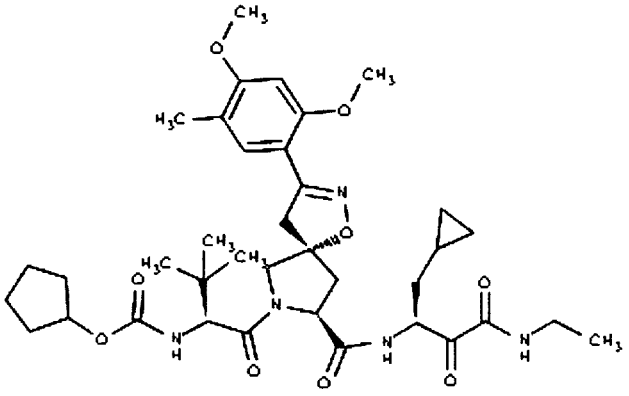
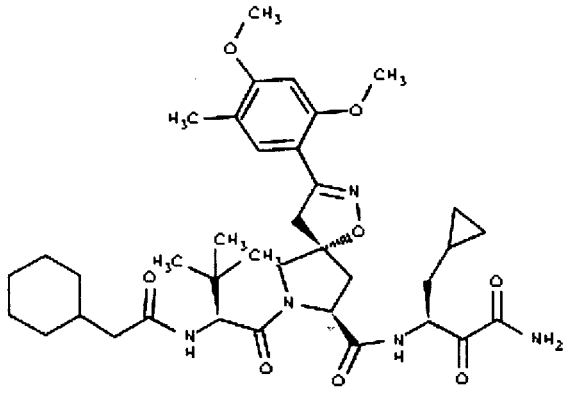
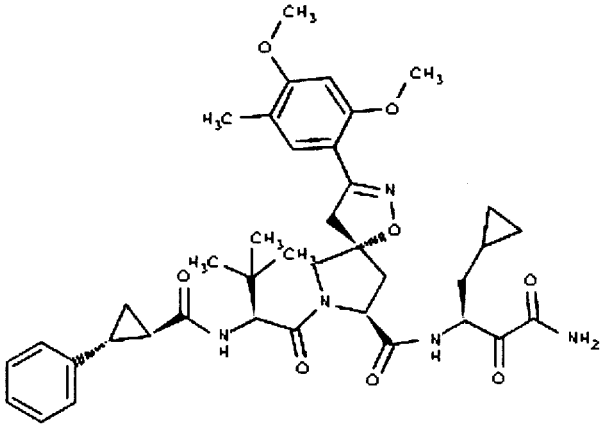
	866
	867
	868

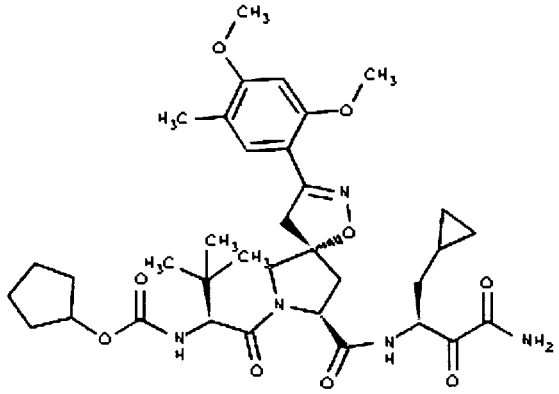
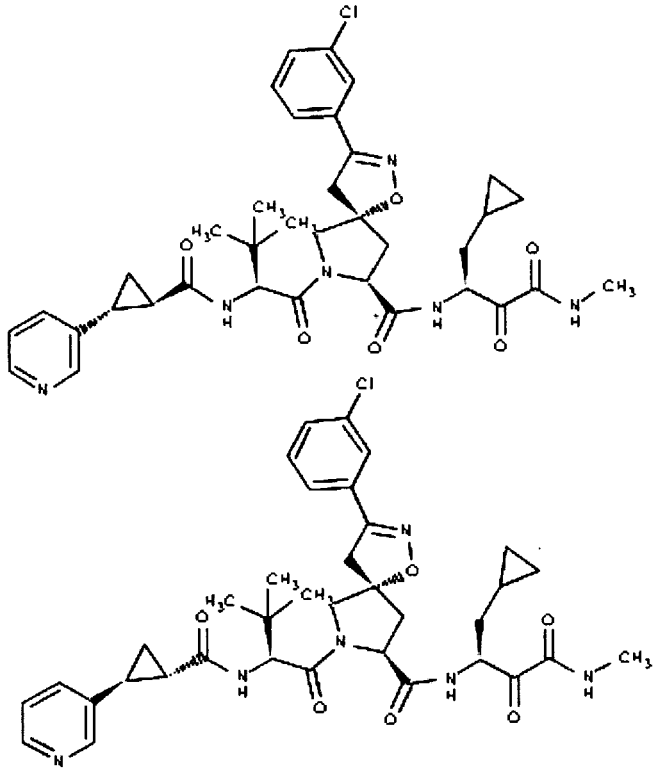
	869
	870
	871

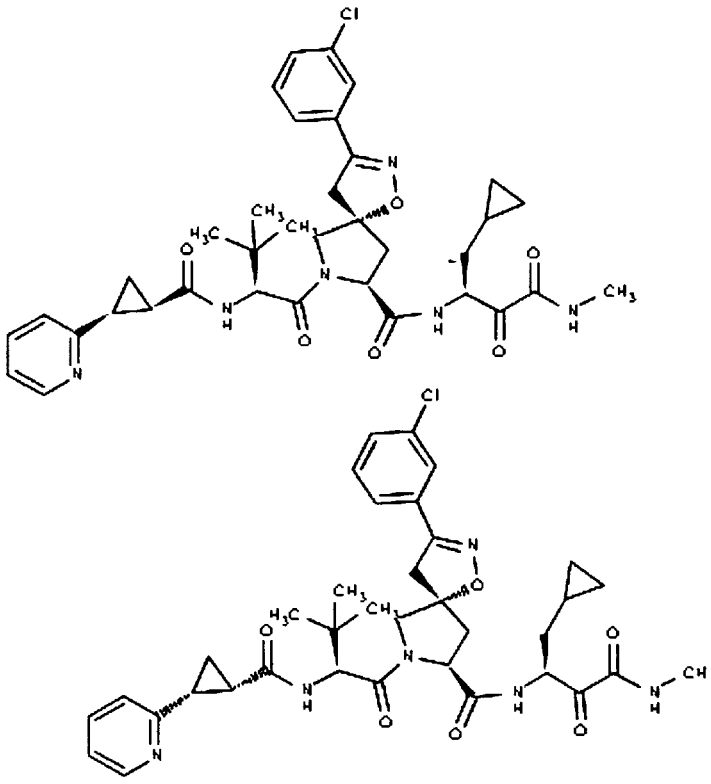
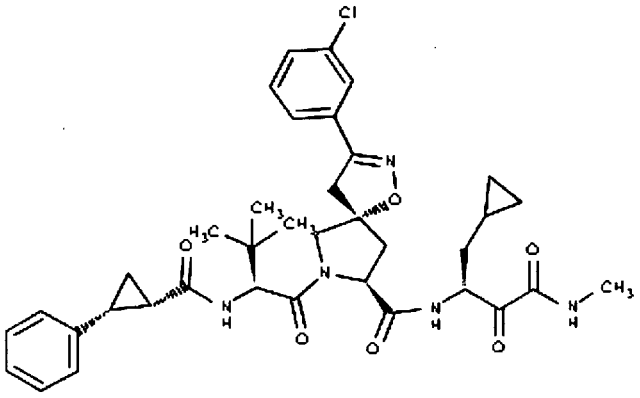
	872
	873
	874

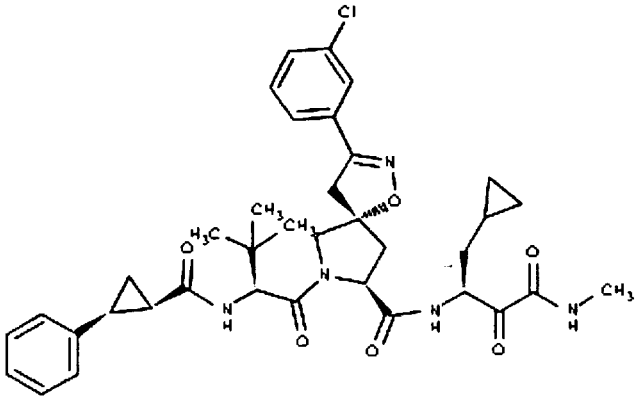
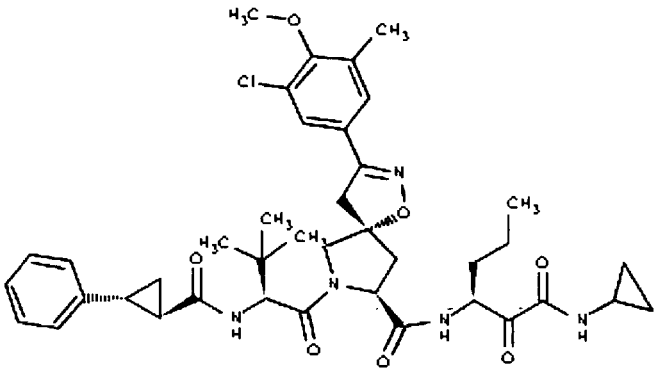
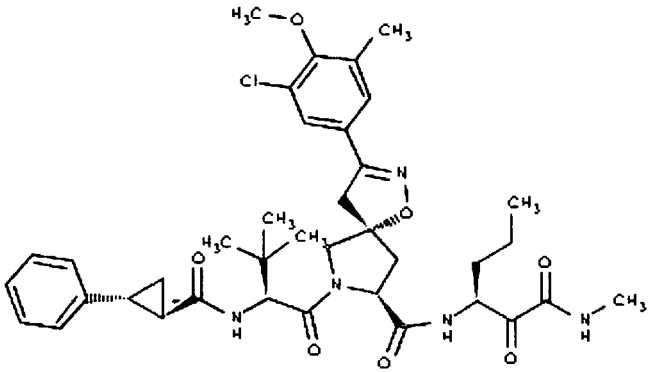
	875
	876
	877

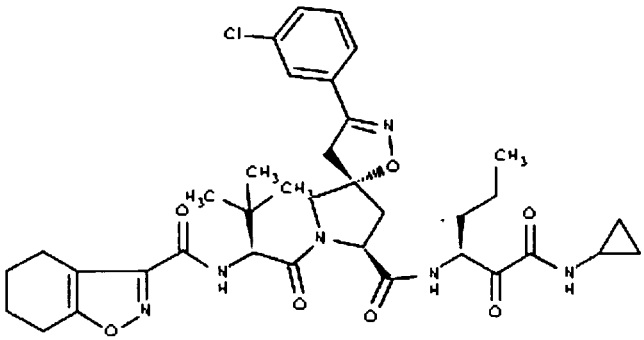
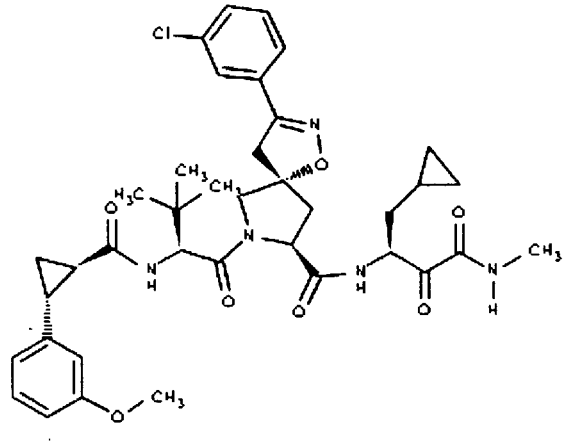
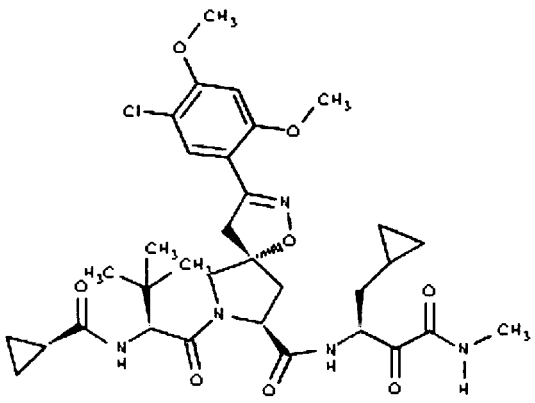
	878
	879
	880

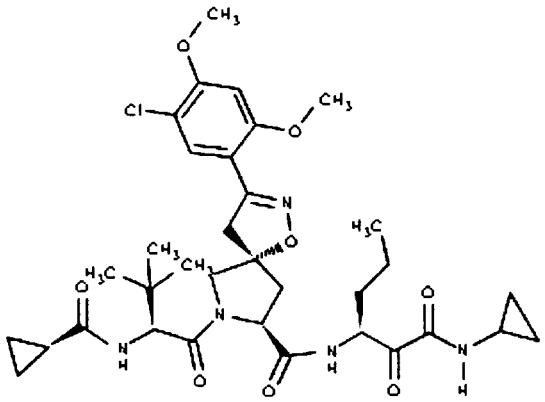
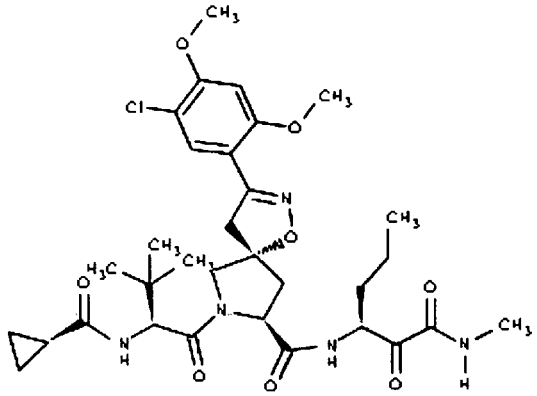
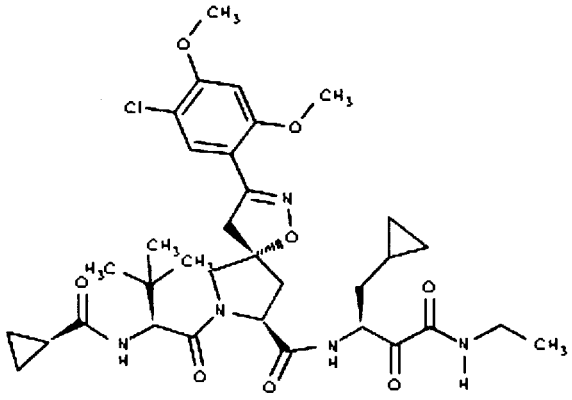
	881
	882
	883

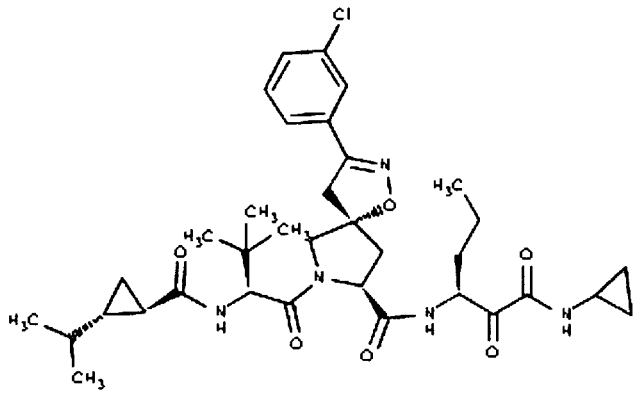
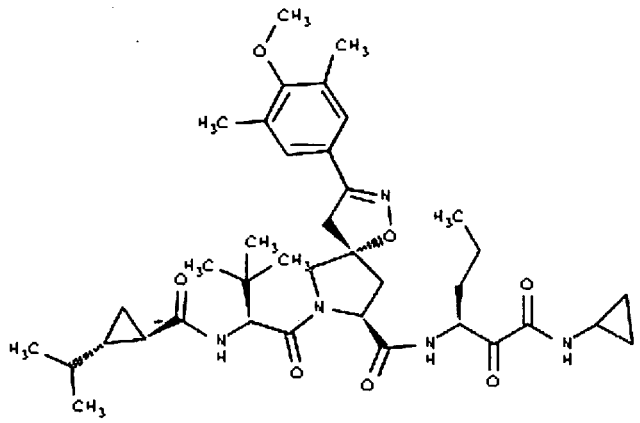
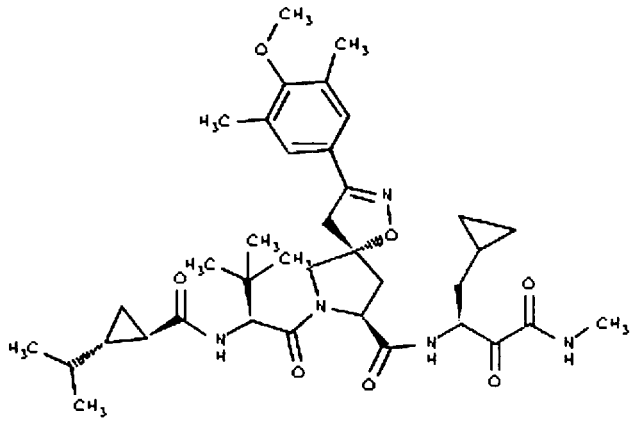
	884
	885

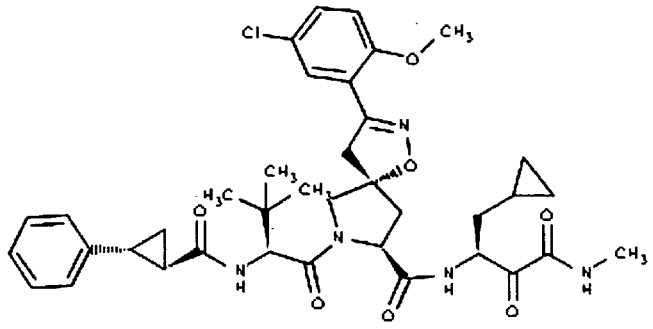
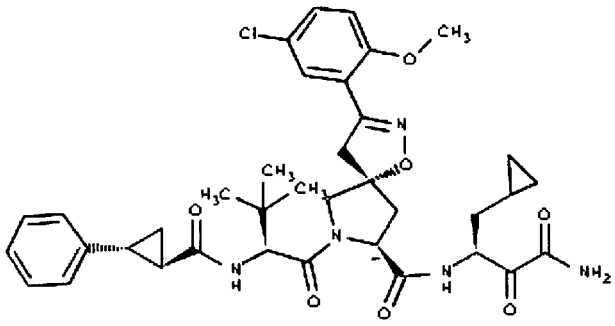
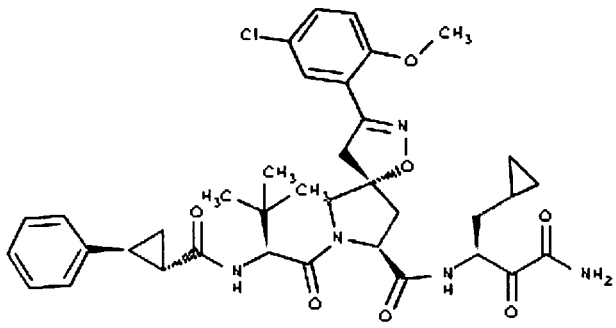
	886
	887

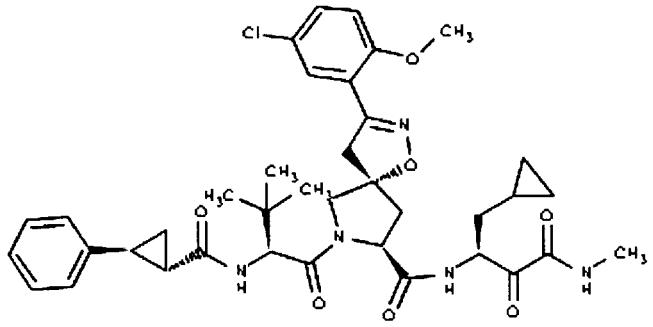
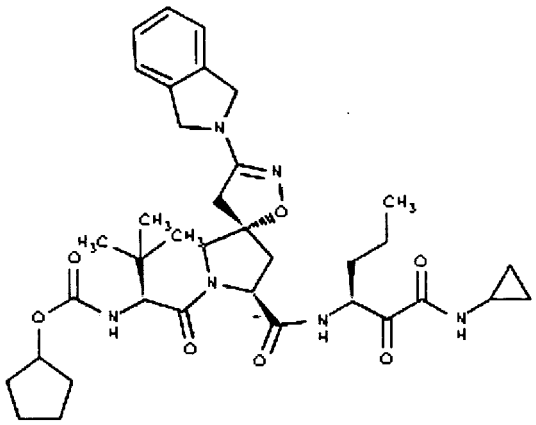
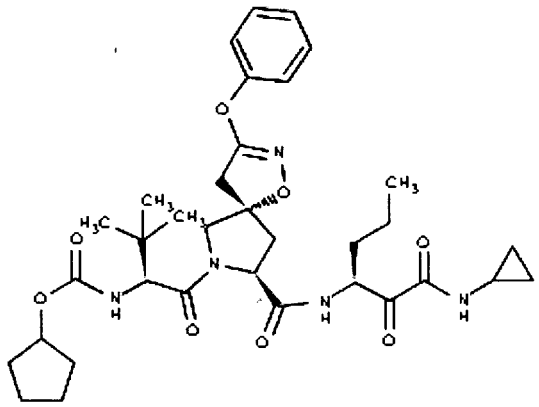
	888
	889
	890

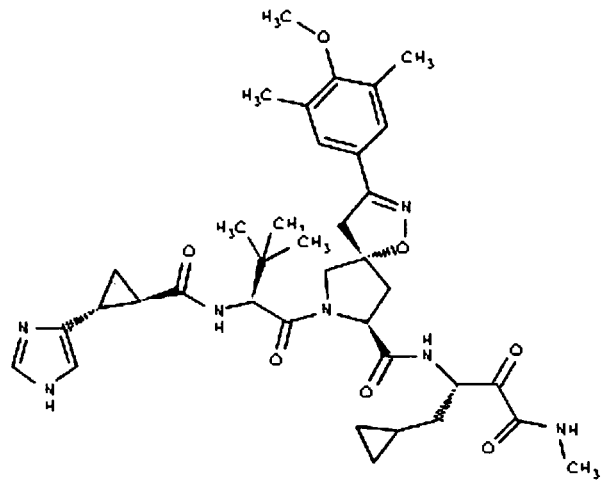
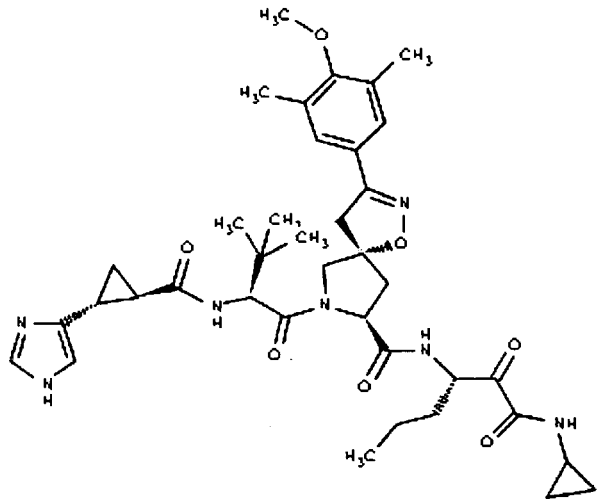
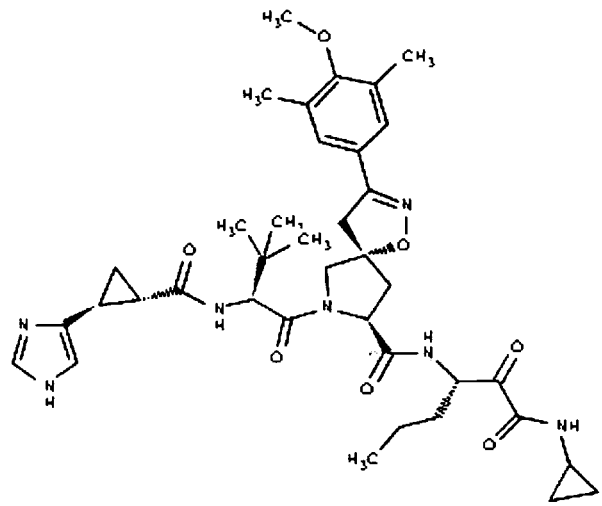
	891
	892
	893

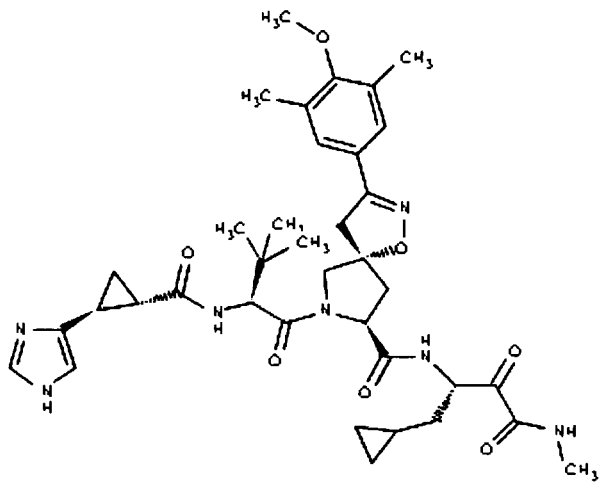
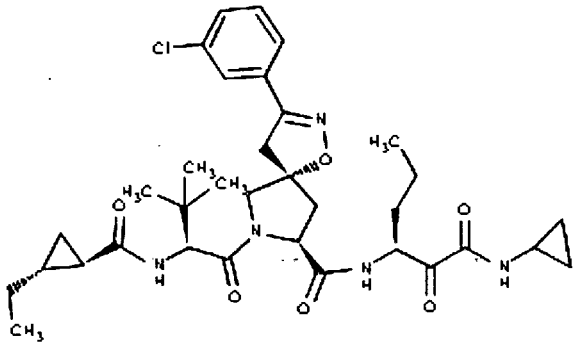
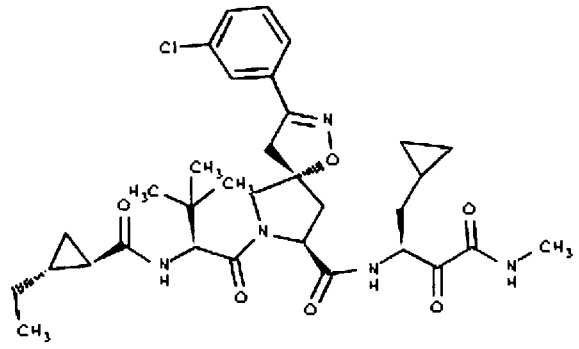
	894
	895
	896

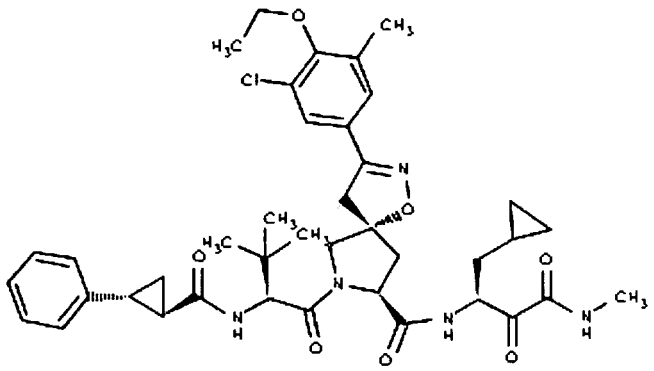
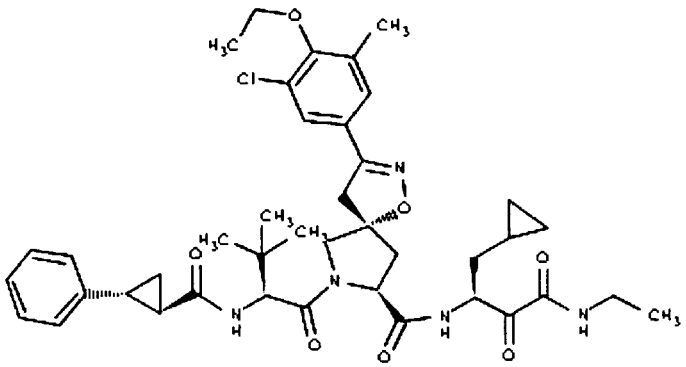
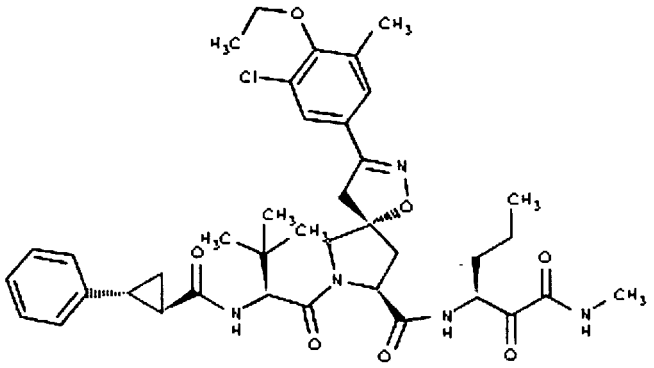
	897
	898
	899

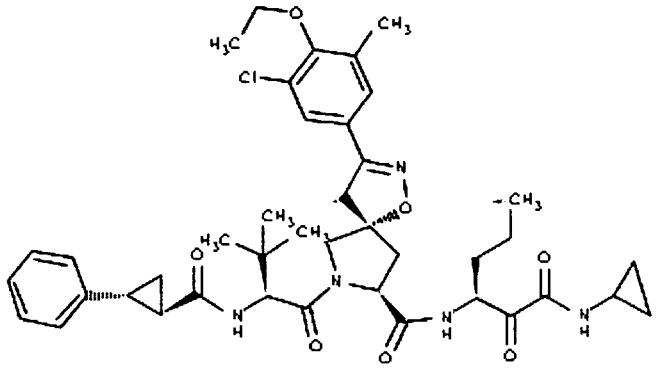
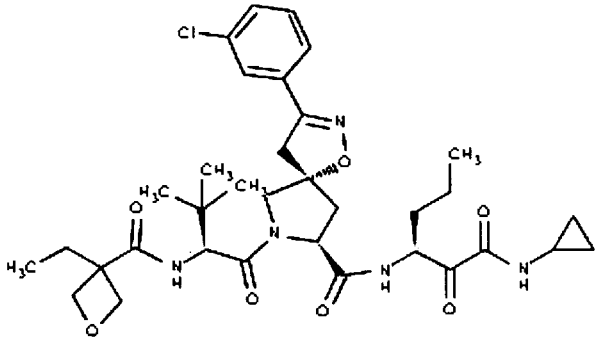
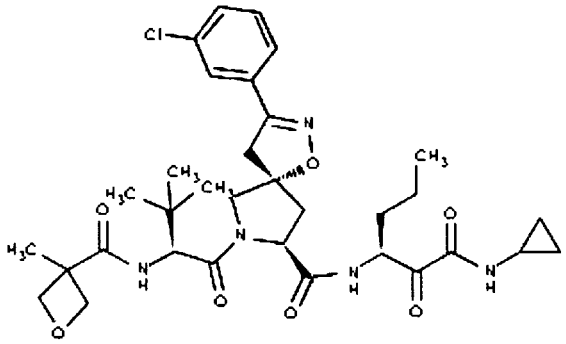
	900
	901
	902

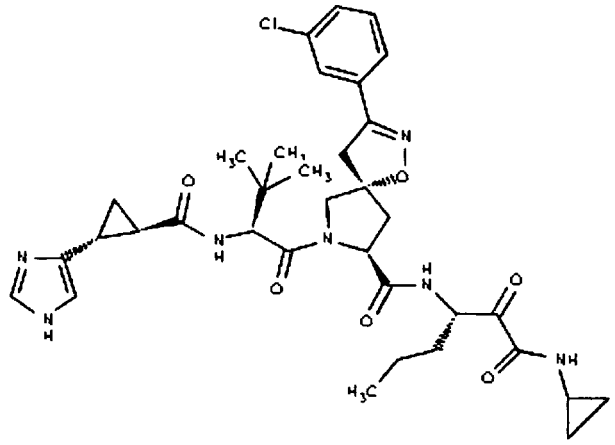
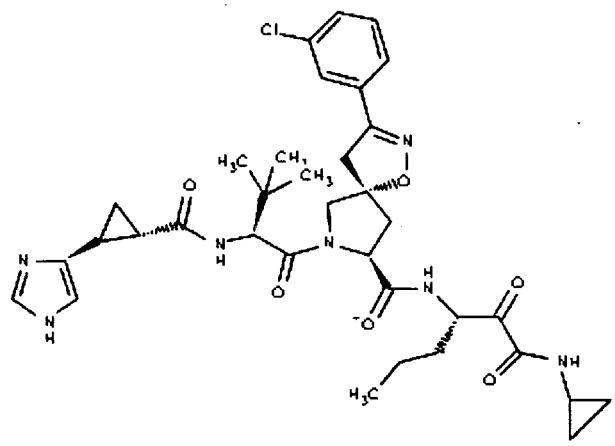
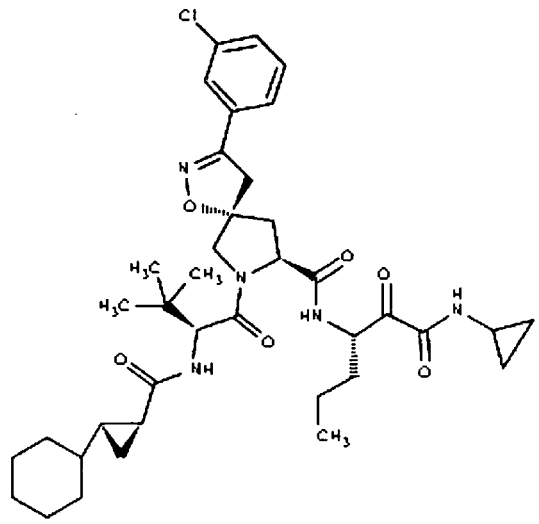
	903
	904
	905

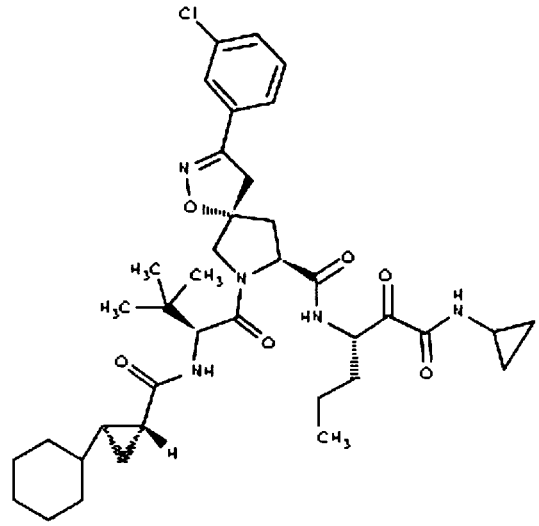
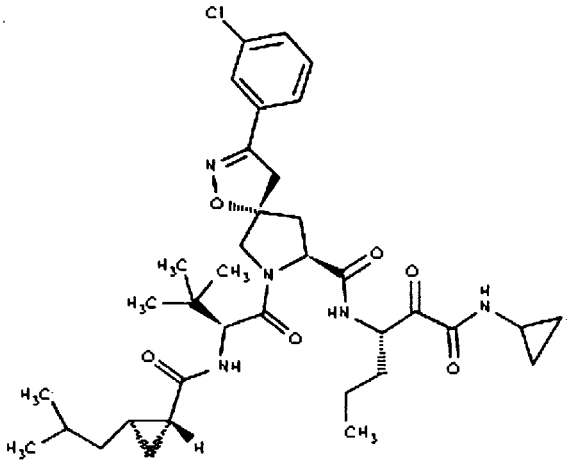
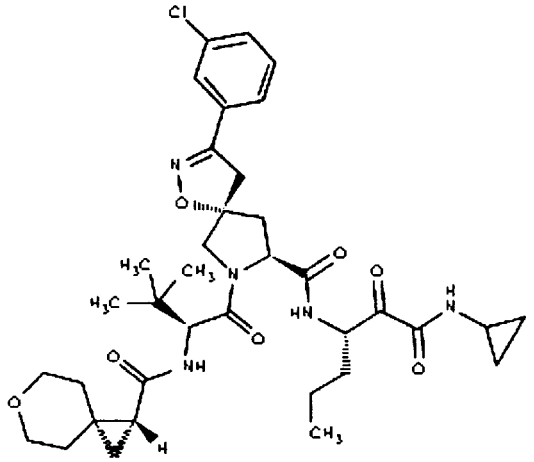
	906
	907
	908

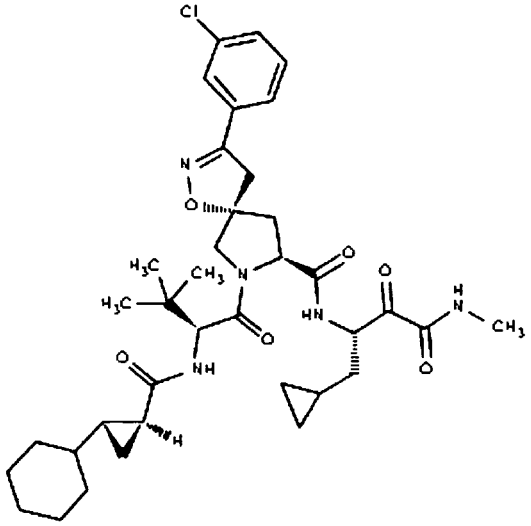
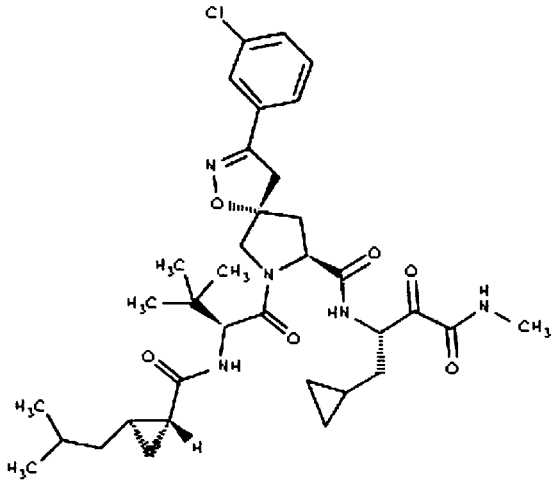
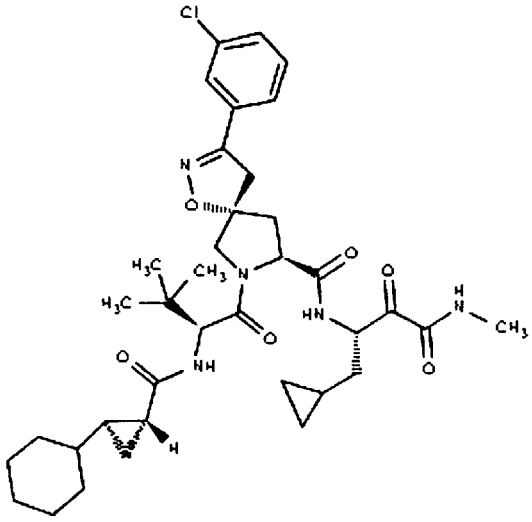
	909
	910
	911

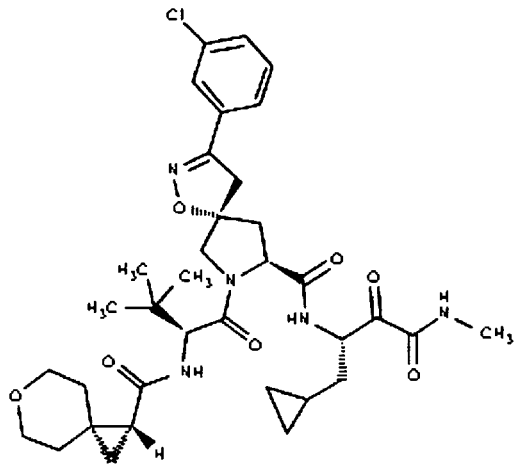
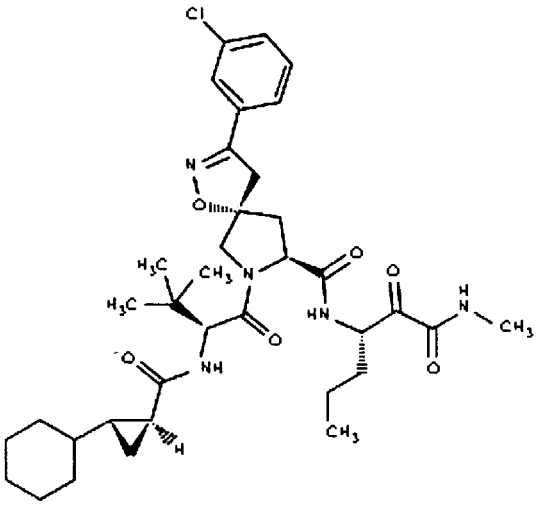
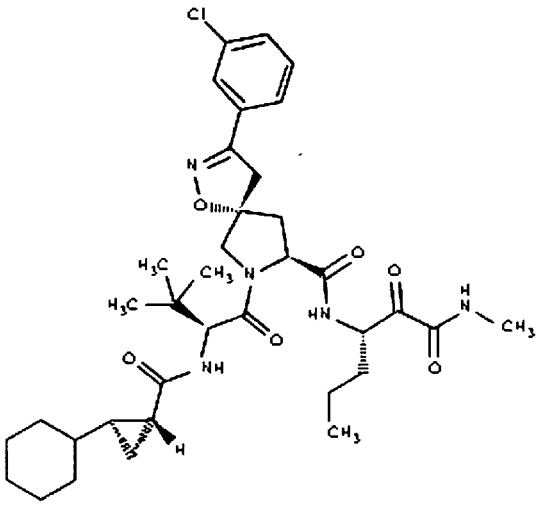
	912
	913
	914

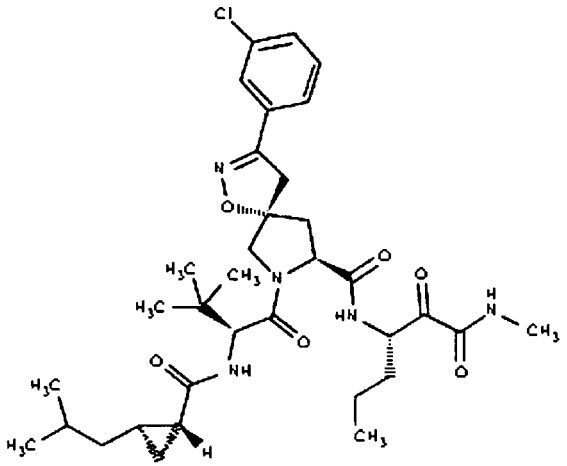
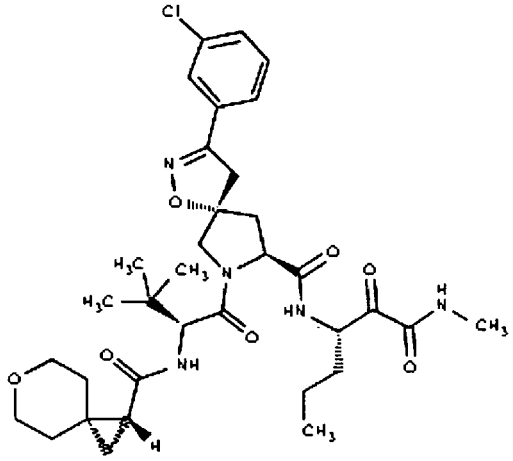
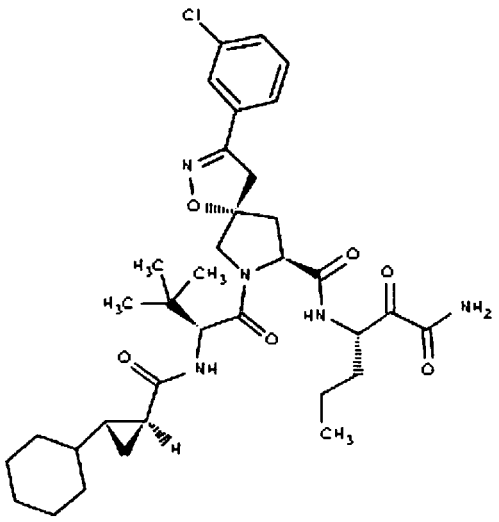
	915
	916
	917

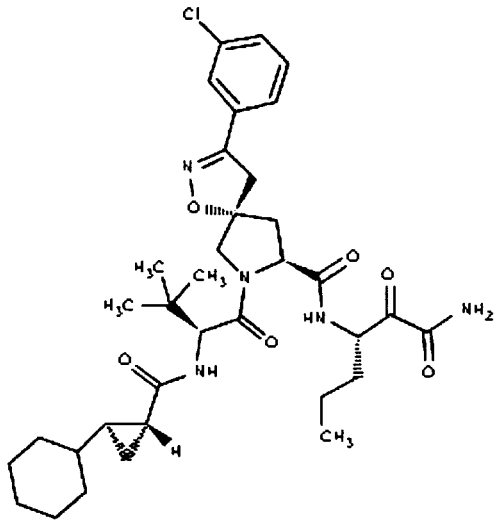
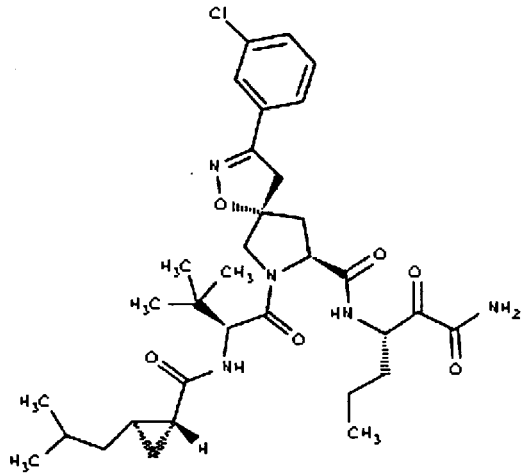
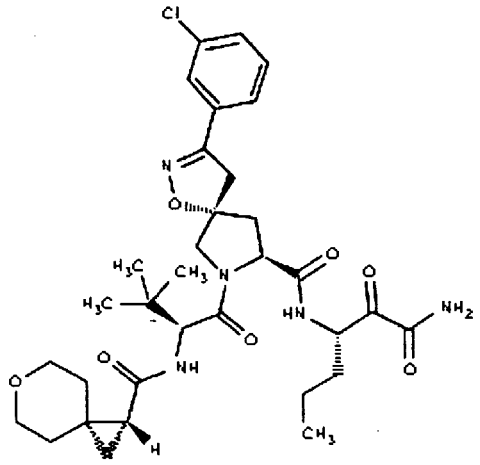
	918
	919
	920

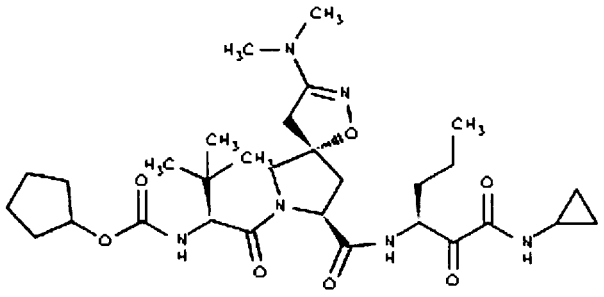
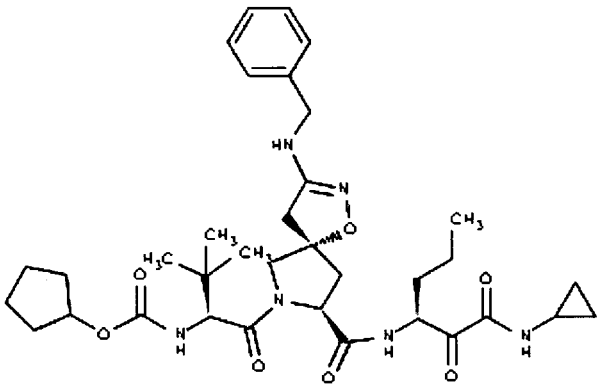
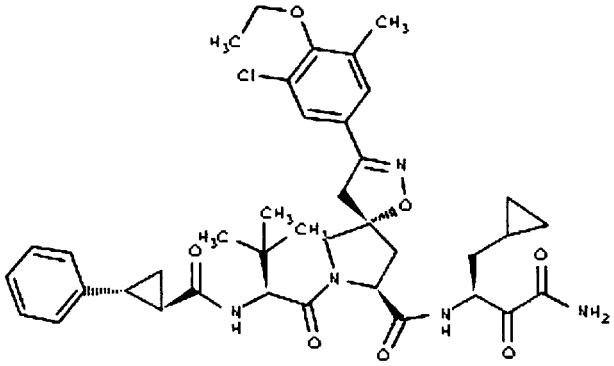
	921
	922
	923

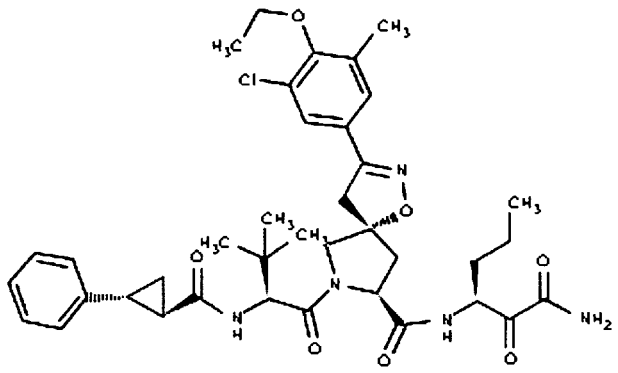
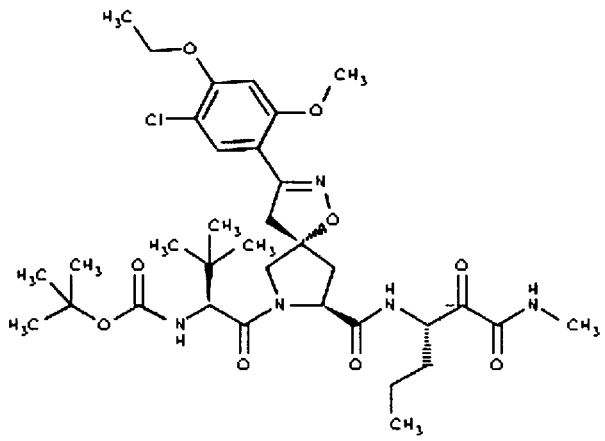
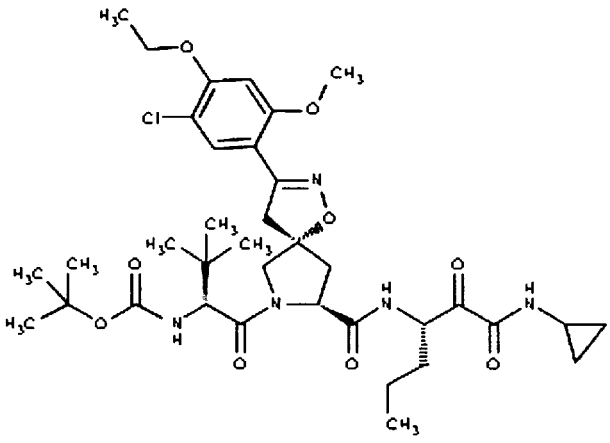
	924
	925
	926

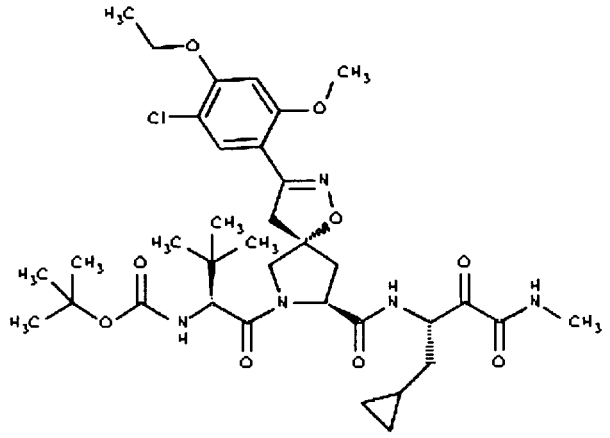
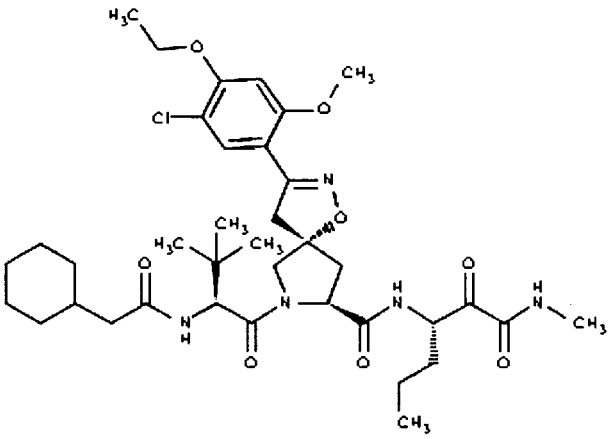
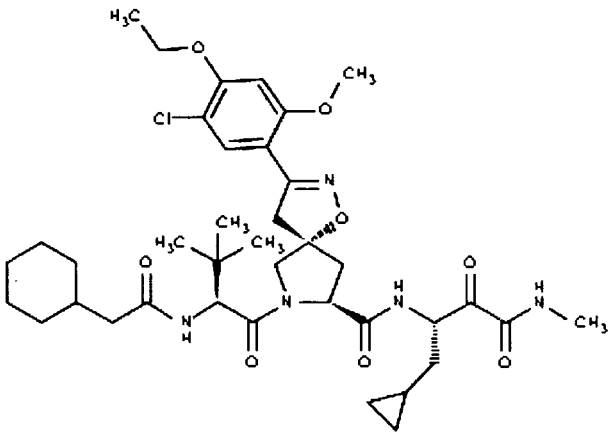
	927
	928
	929

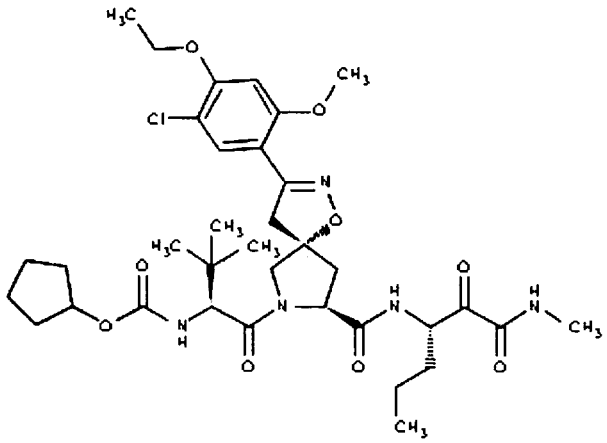
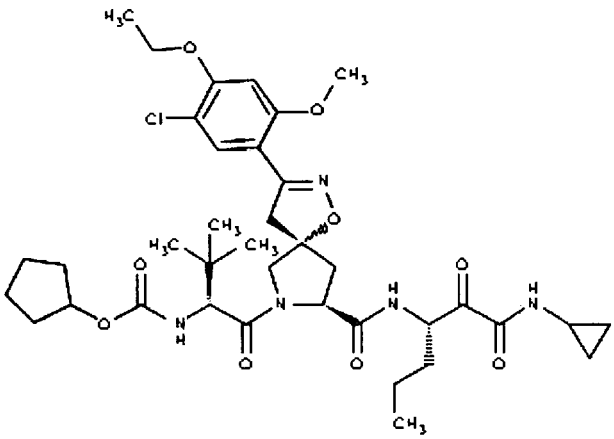
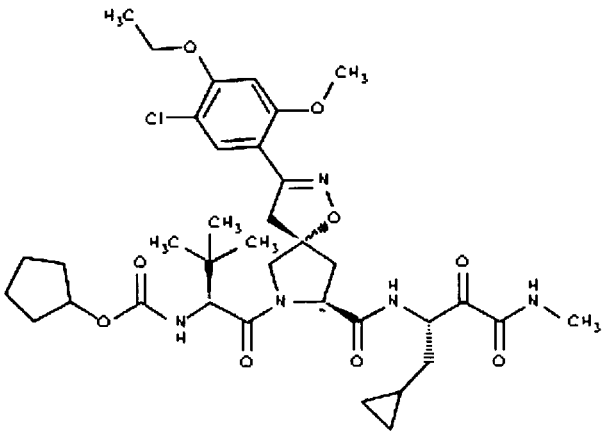
	930
	931
	932

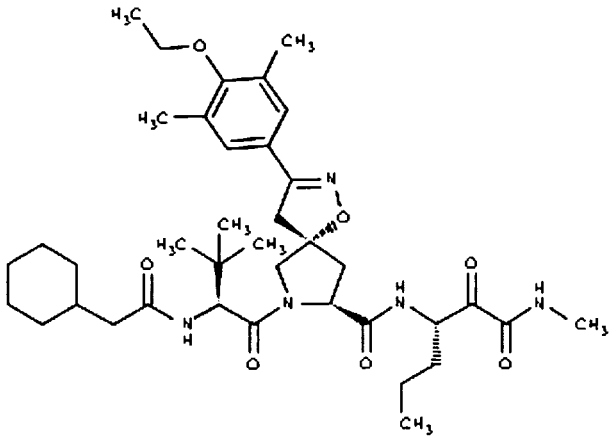
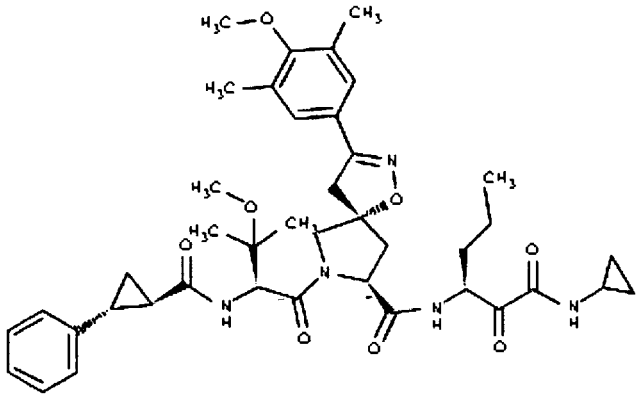
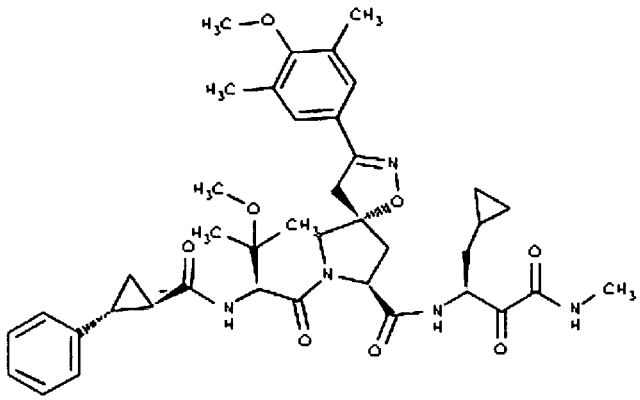
	933
	934
	935

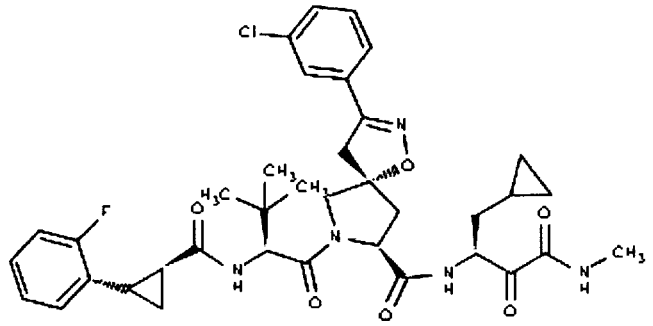
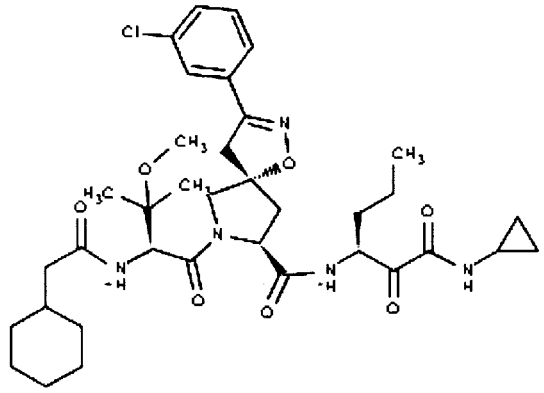
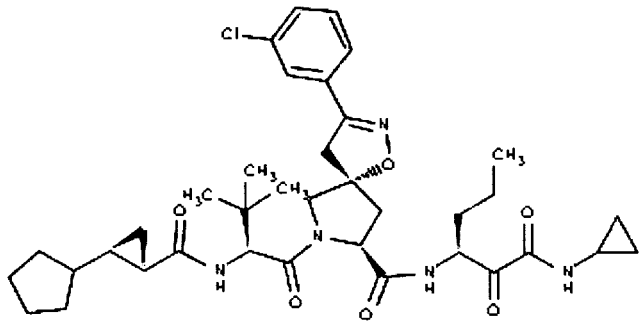
	936
	937
	938

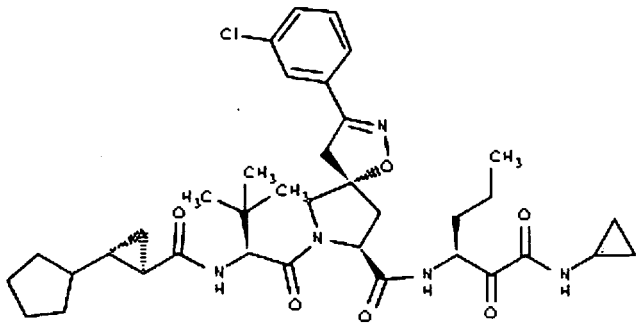
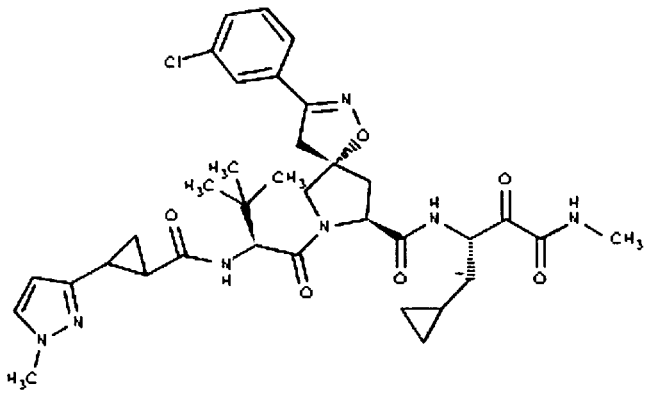
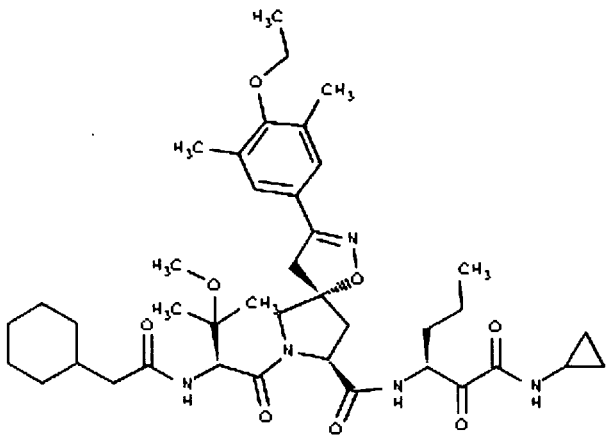
	939
	940
	941

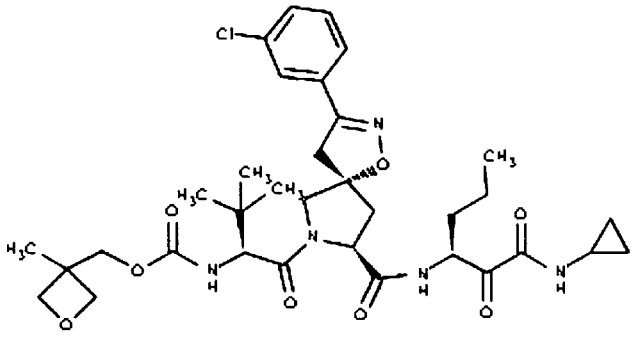
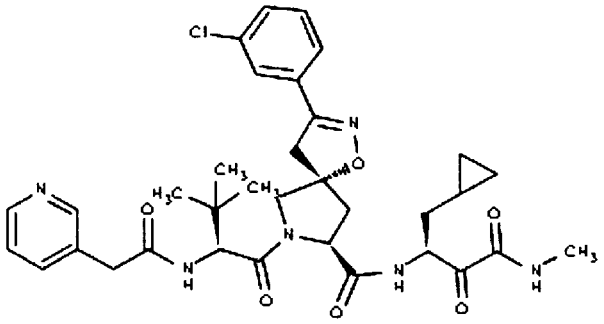
	942
	943
	944

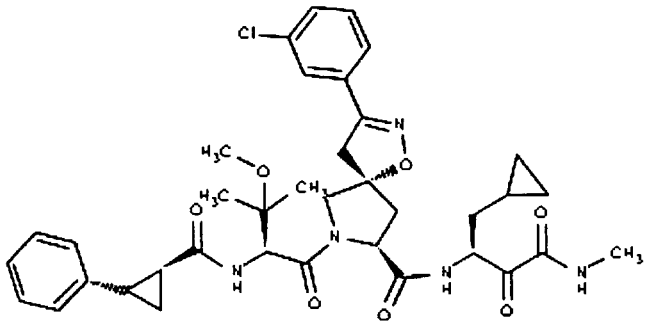
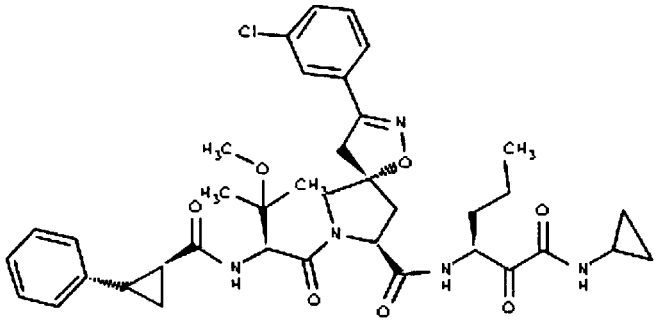
	945
	946
	947

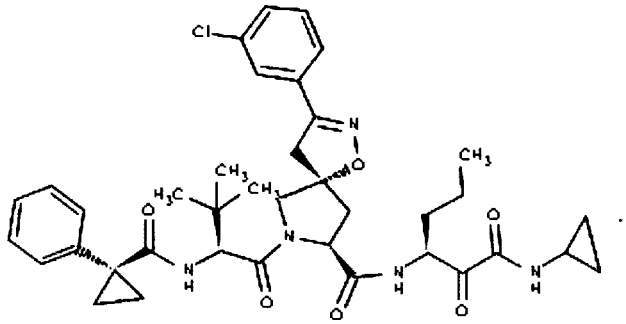
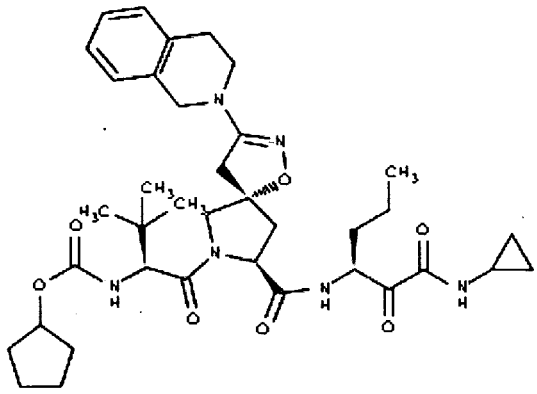
	948
	949
	950

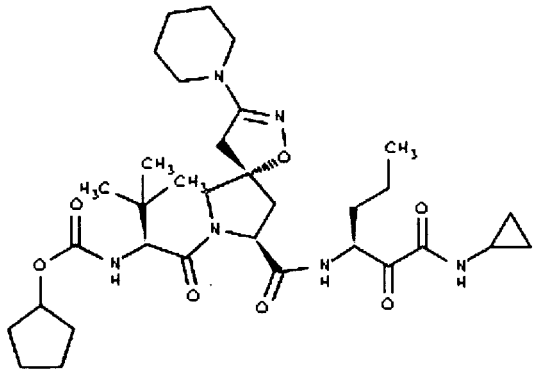
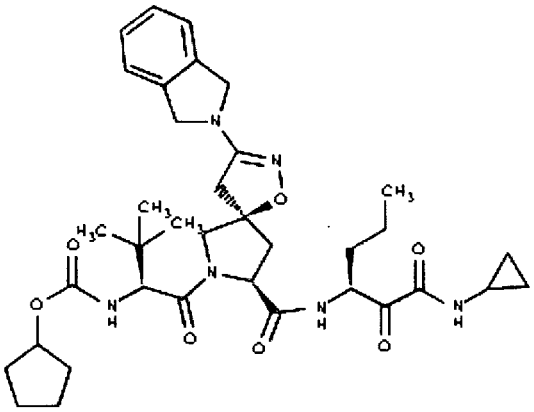
	951
	952
	953

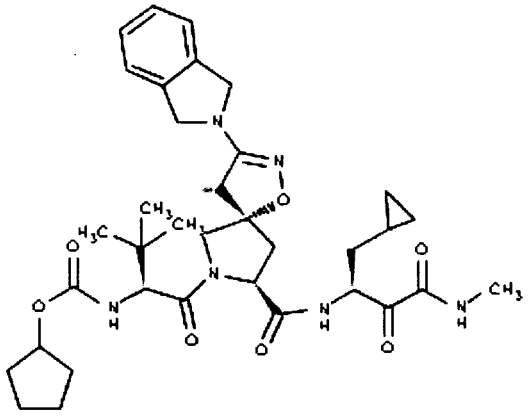
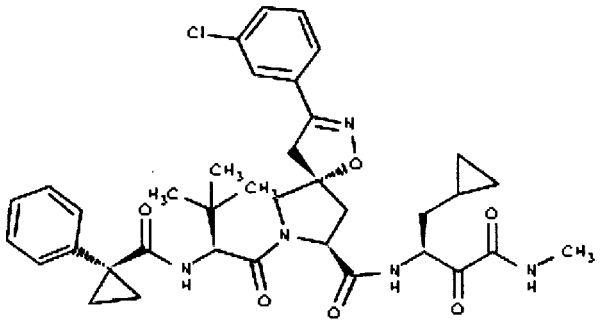
	954
	955
	956

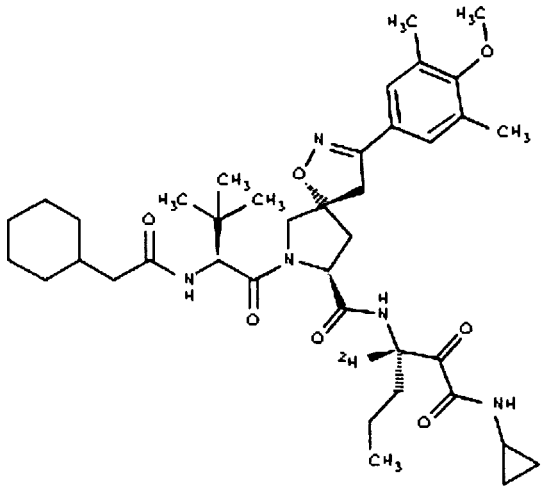
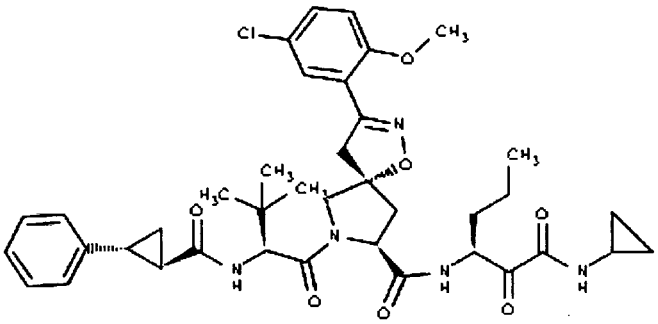
	957
	958

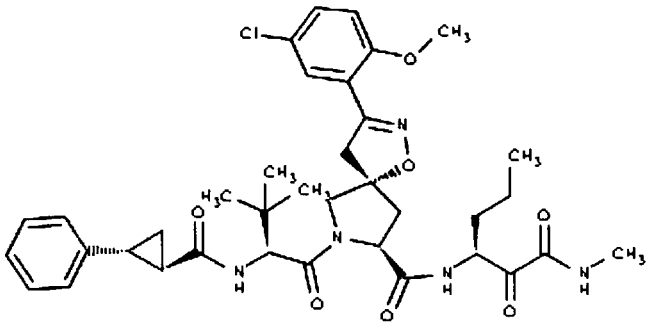
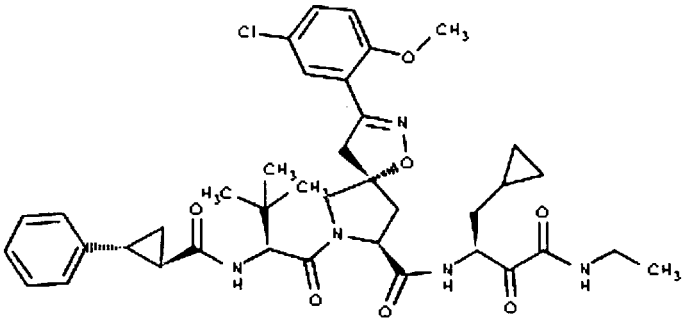
	959
	960

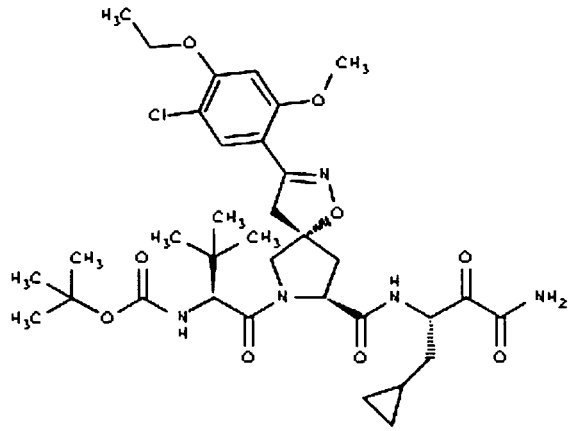
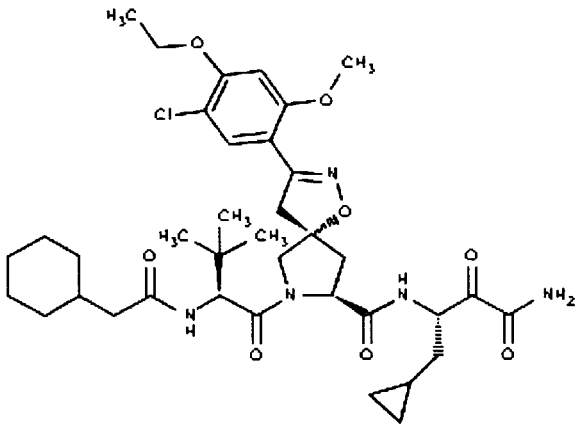
	961
	962

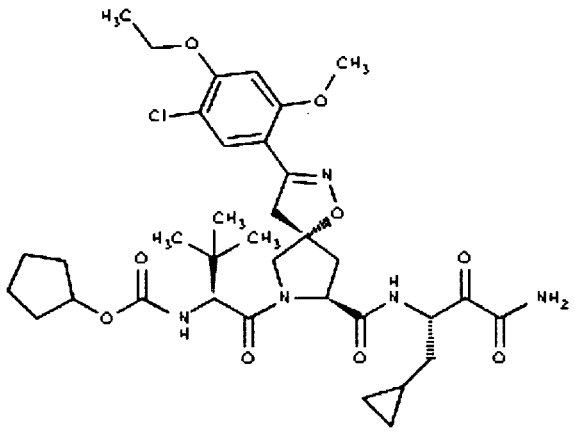
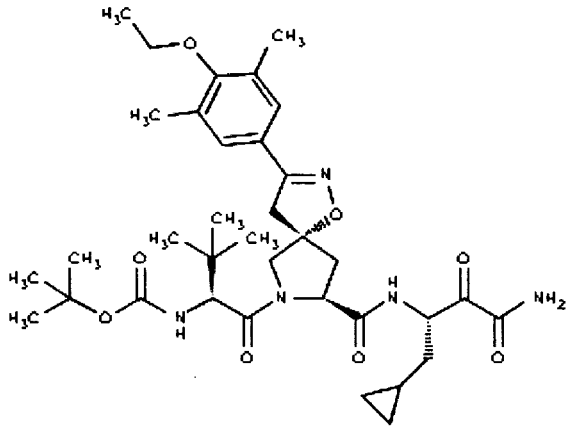
	963
	964

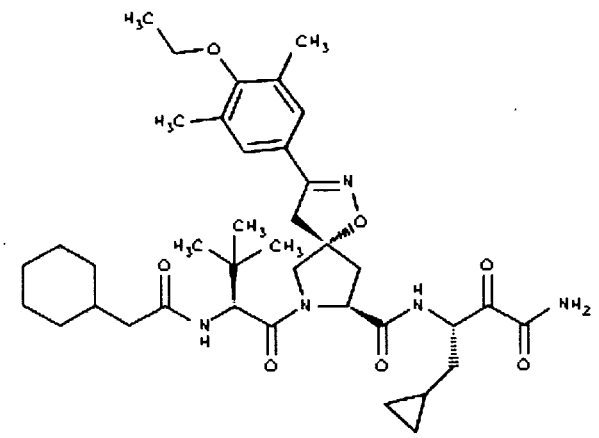
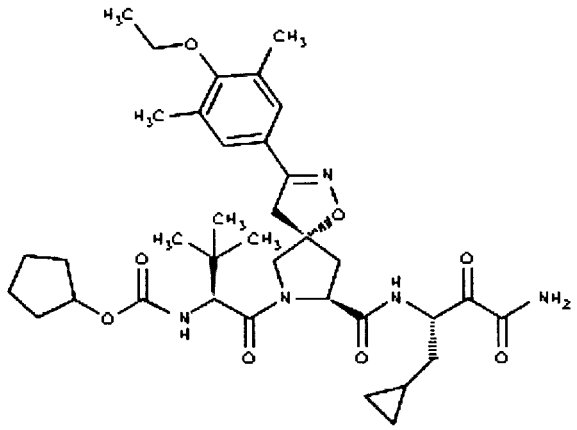
	965
	966

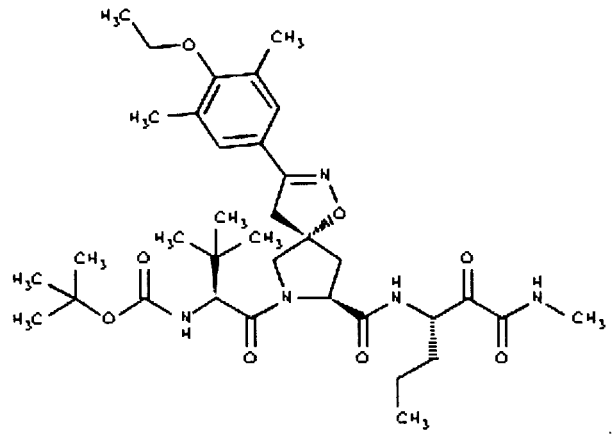
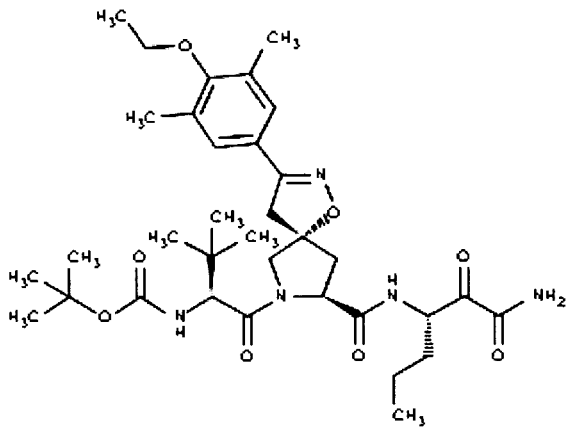
	967
	968

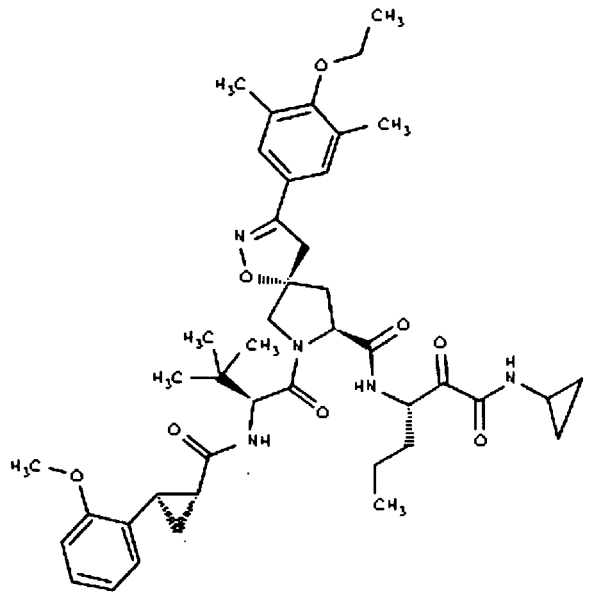
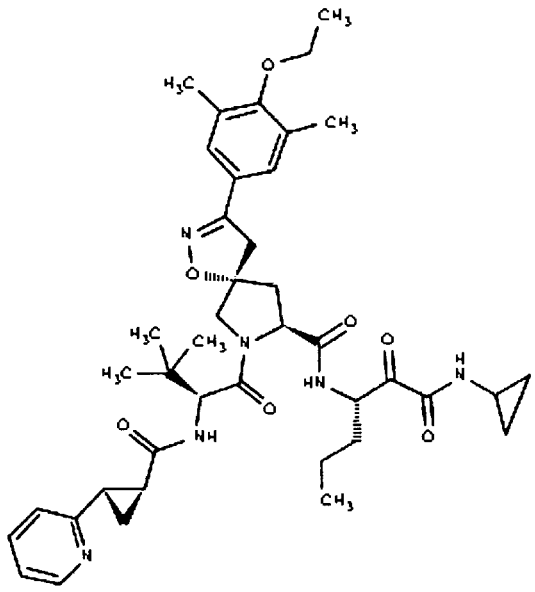
	969
	970

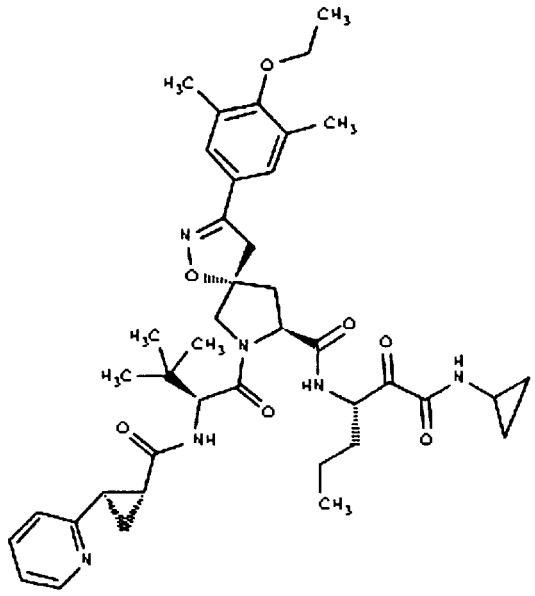
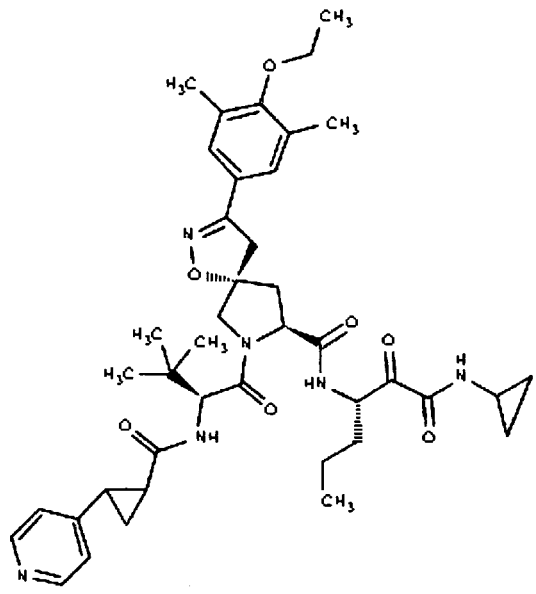
	971
	972

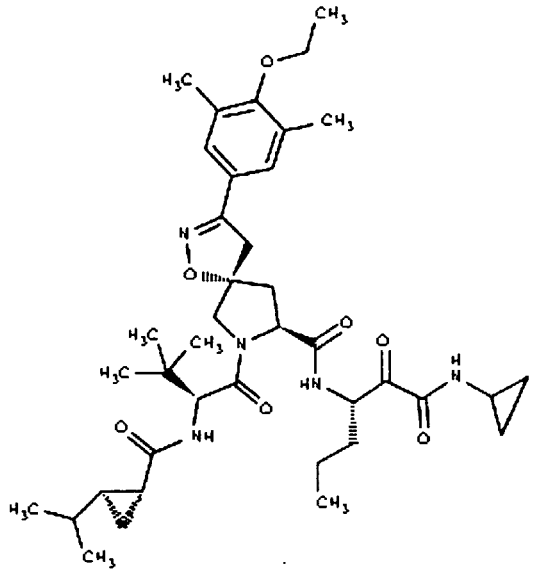
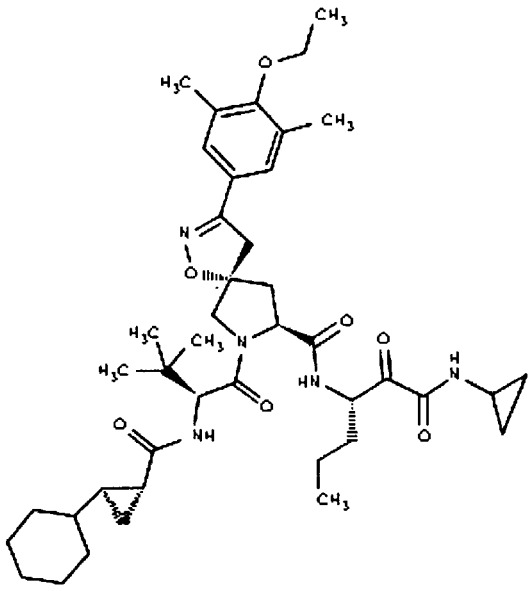
	973
	974

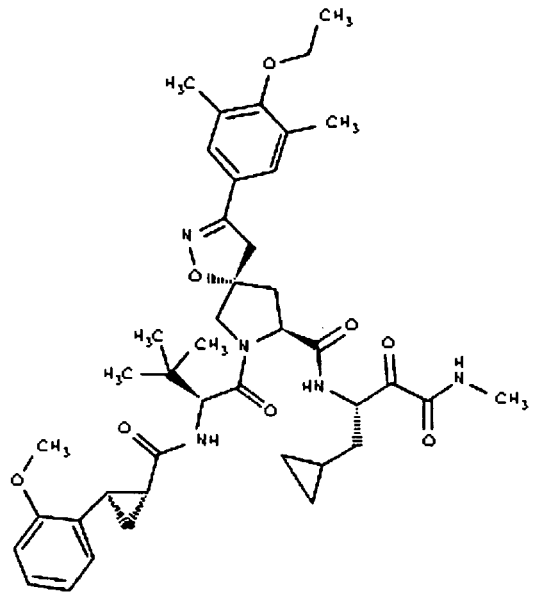
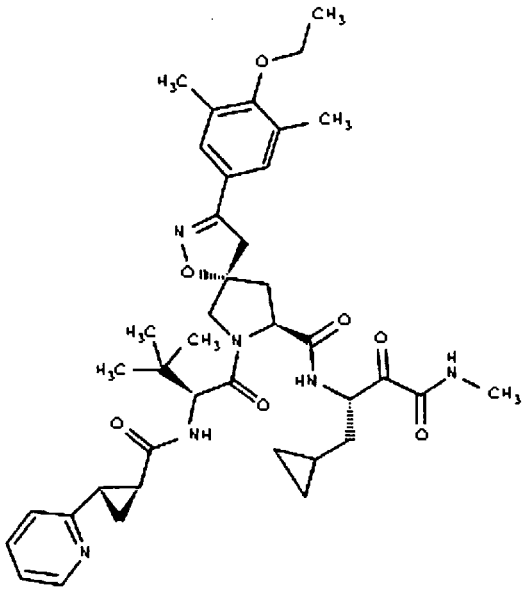
	975
	976

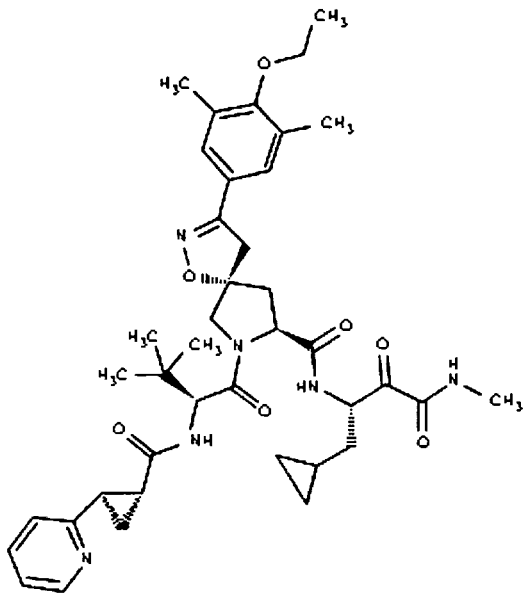
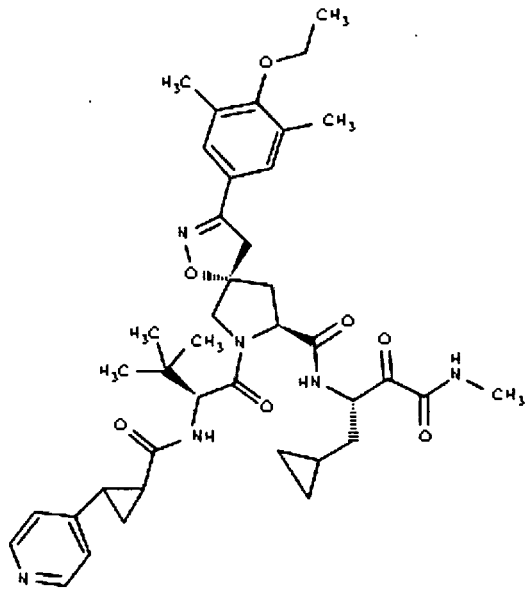
	977
	978

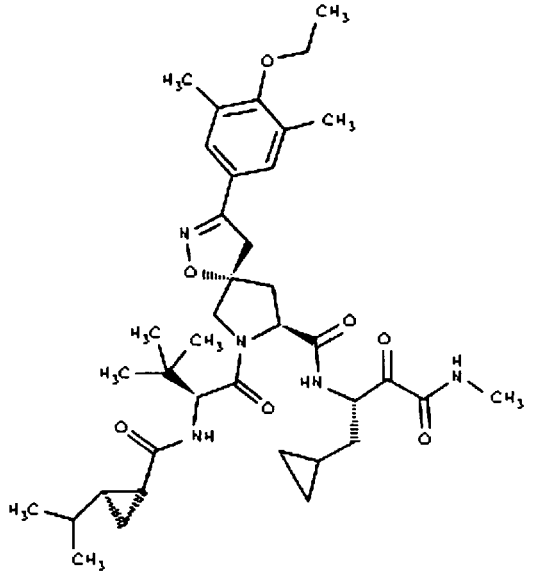
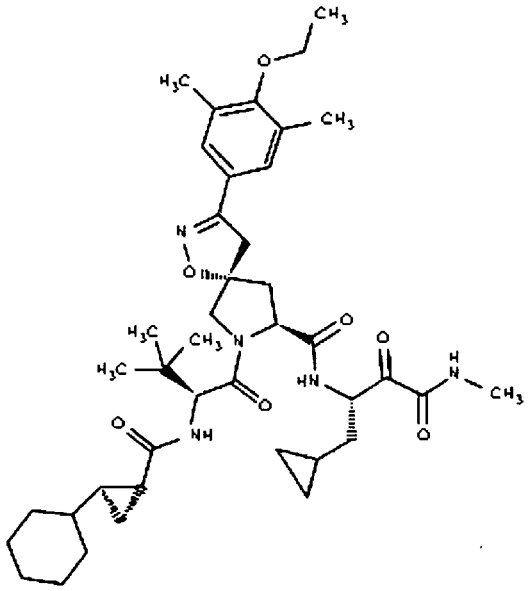
	979
	980

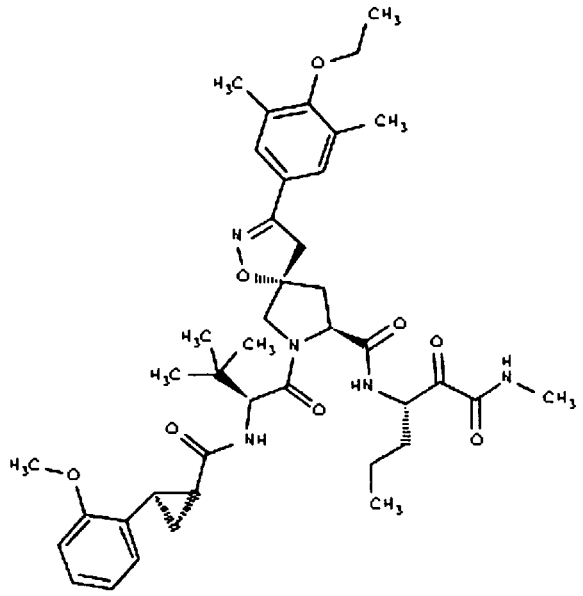
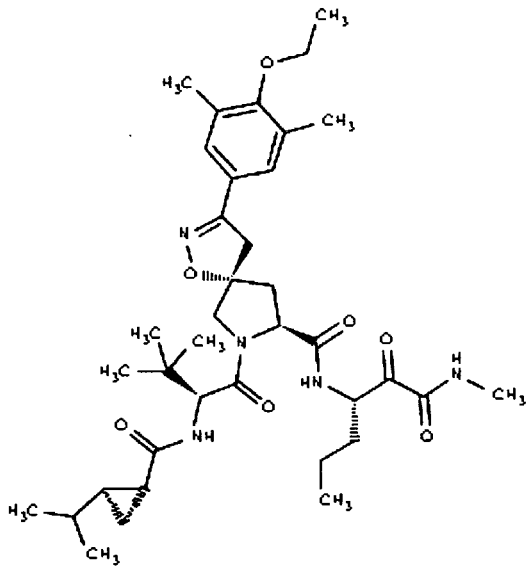
	981
	982

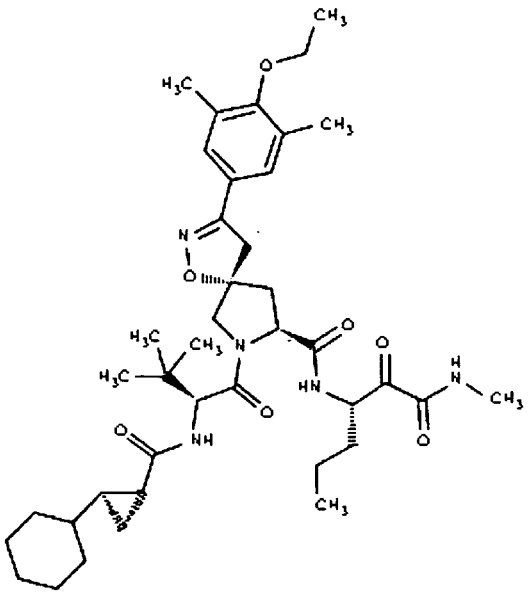
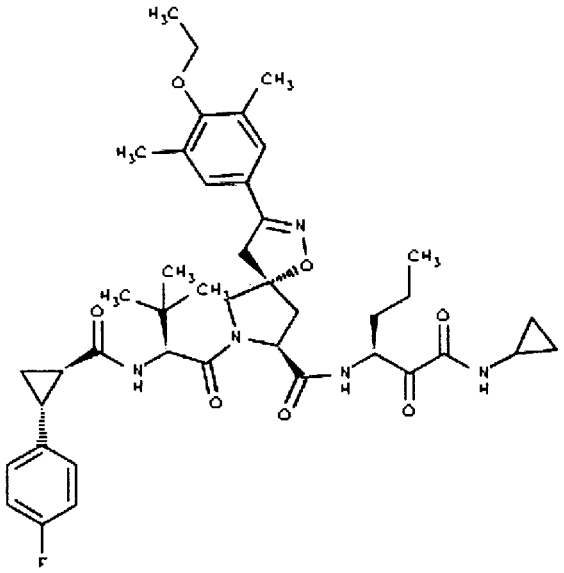
	983
	984

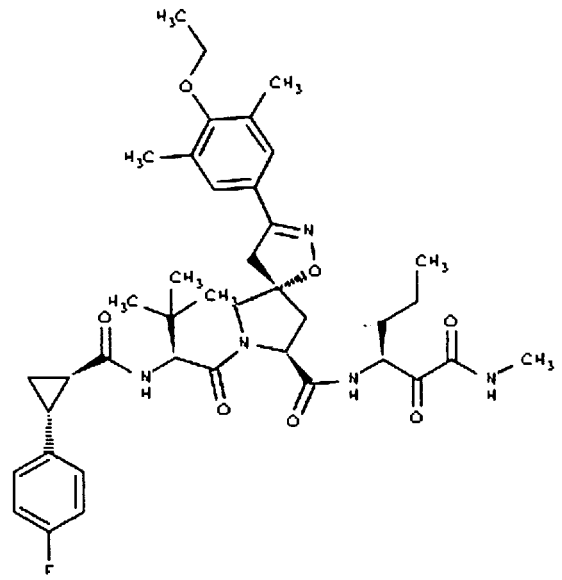
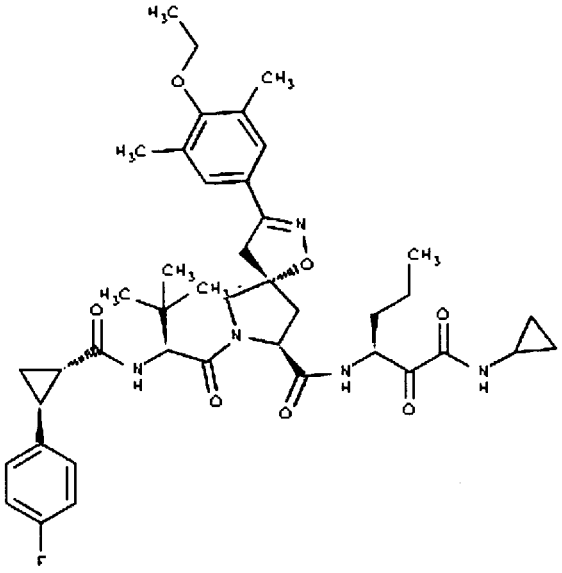
	985
	986

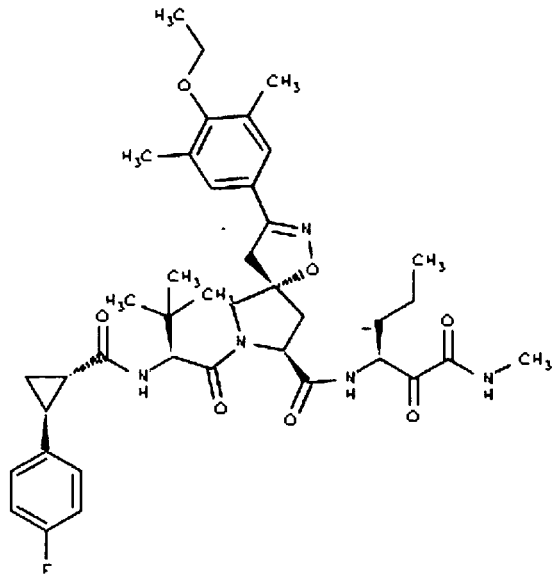
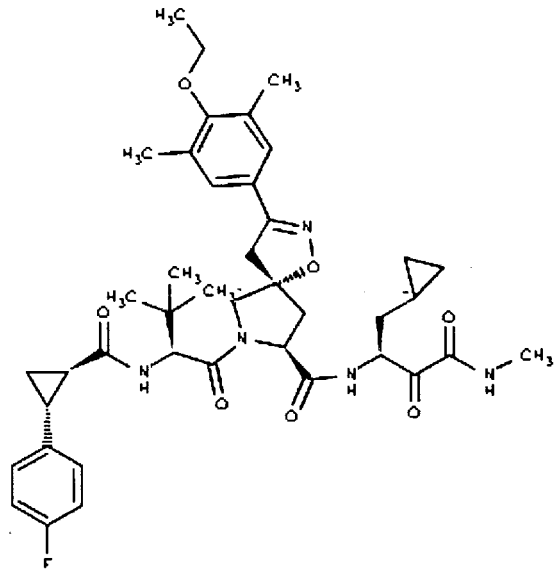
	987
	988

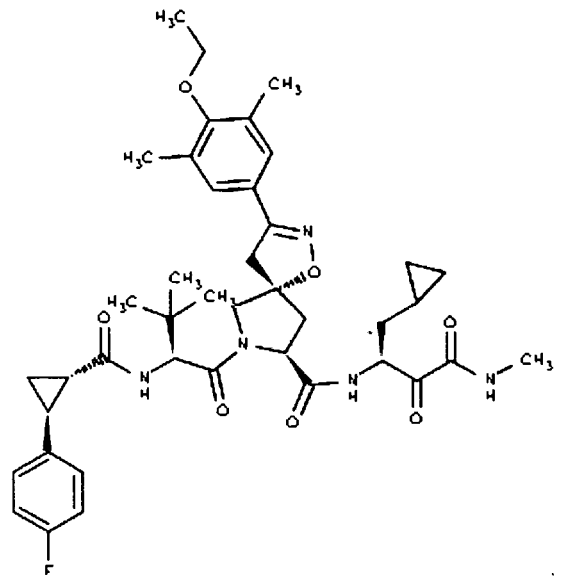
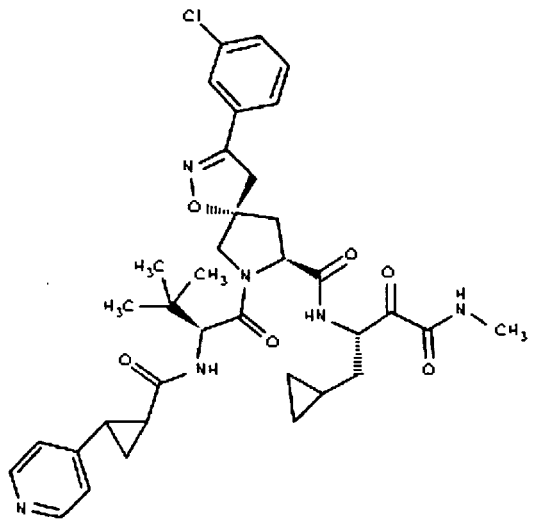
	989
	990

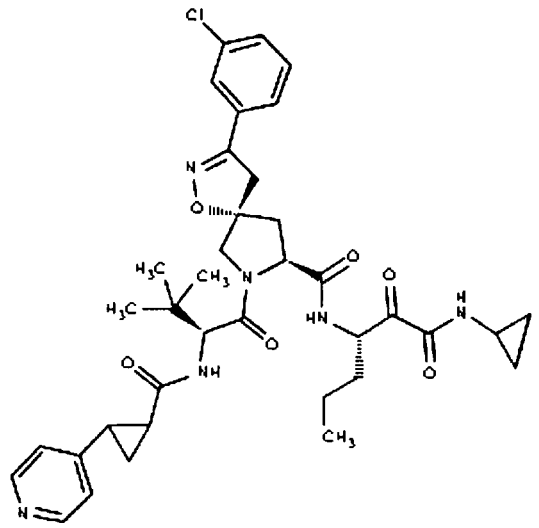
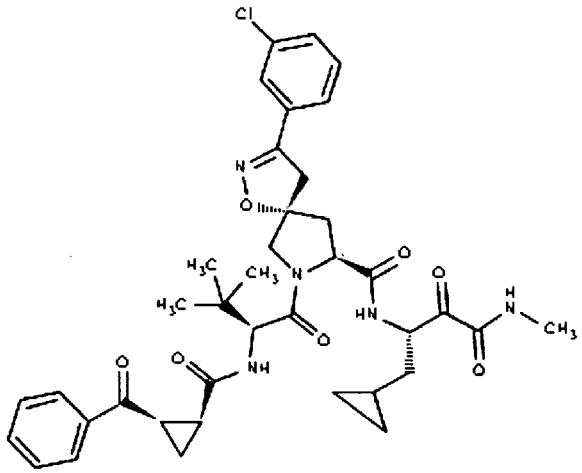
	991
	992

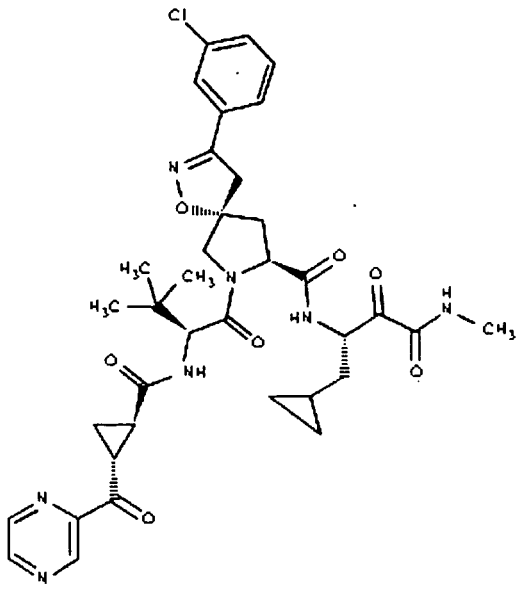
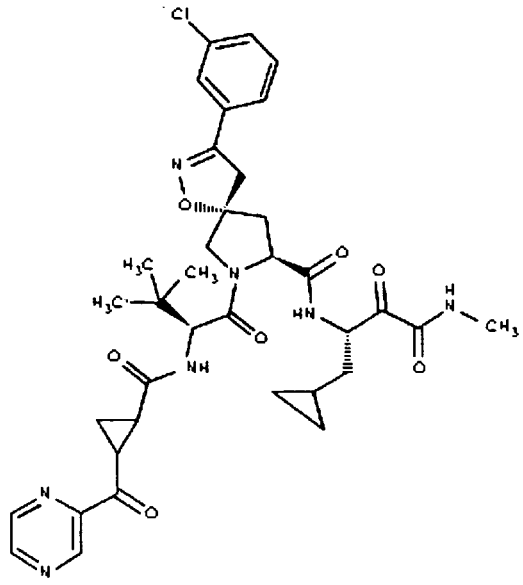
	993
	994

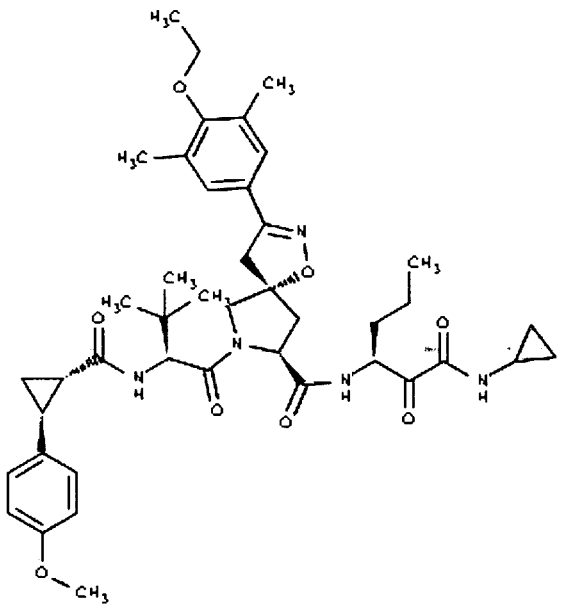
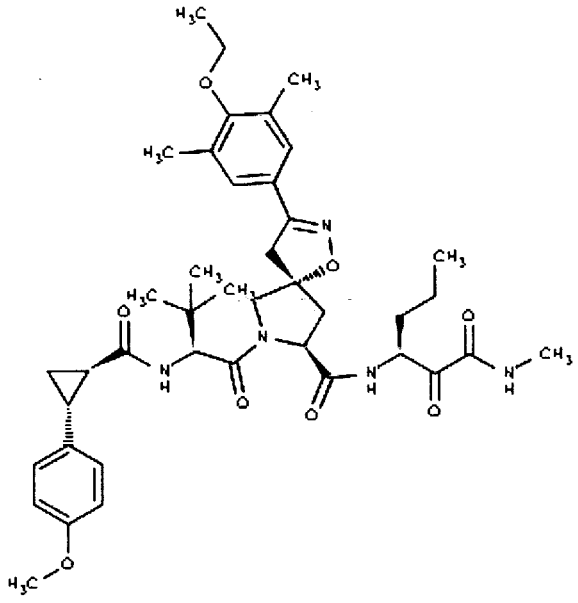
	995
	996

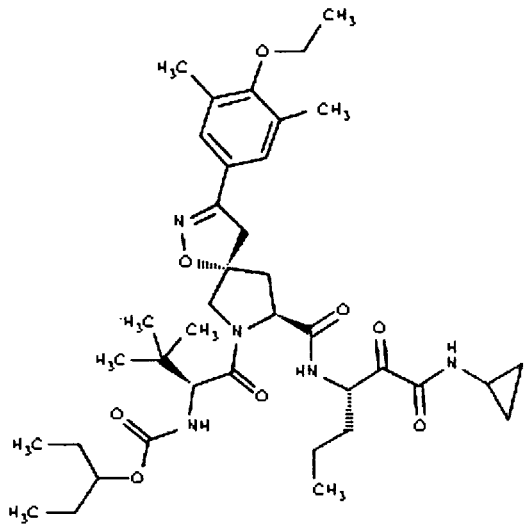
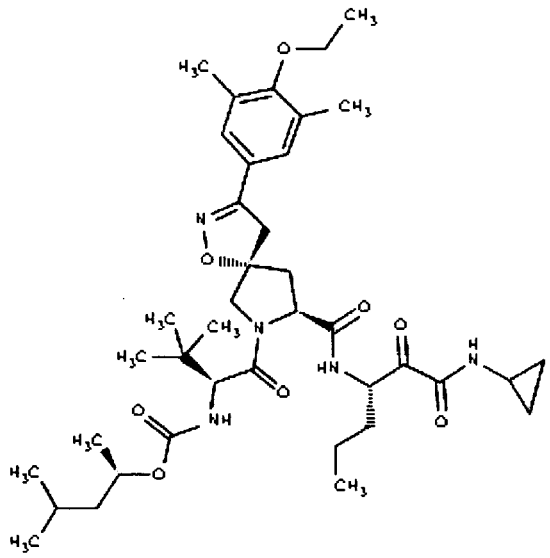
	997
	998

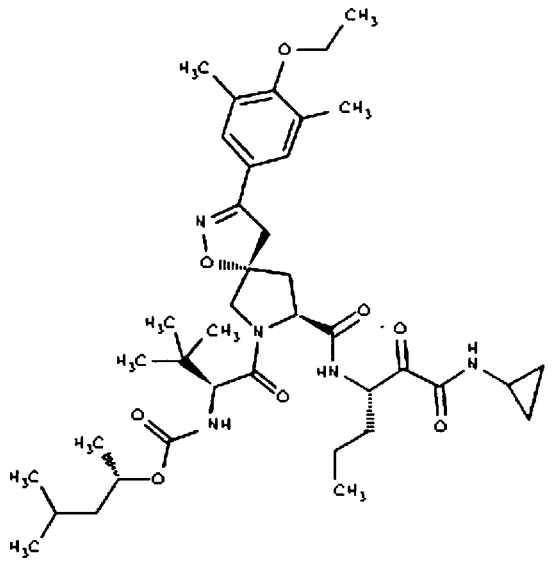
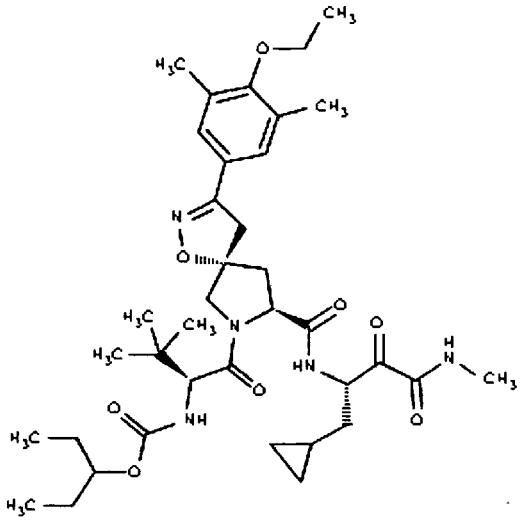
	999
	1000

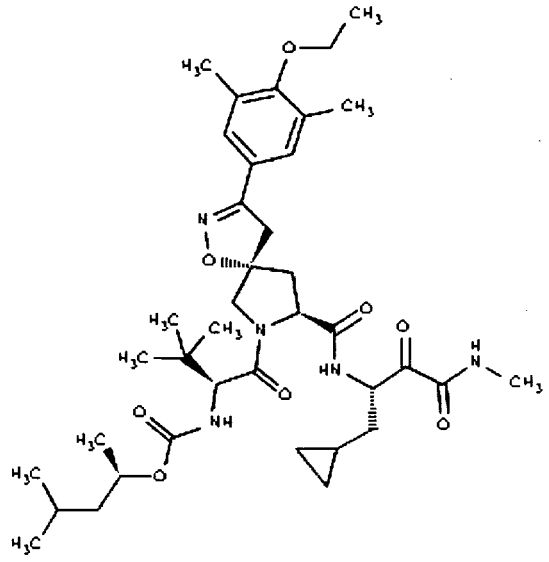
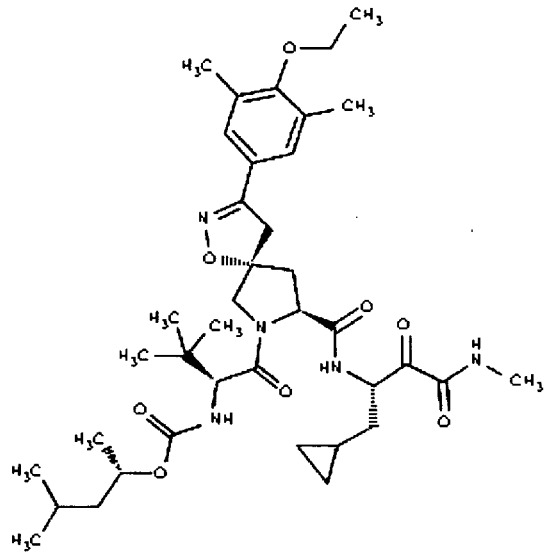
	1001
	1002

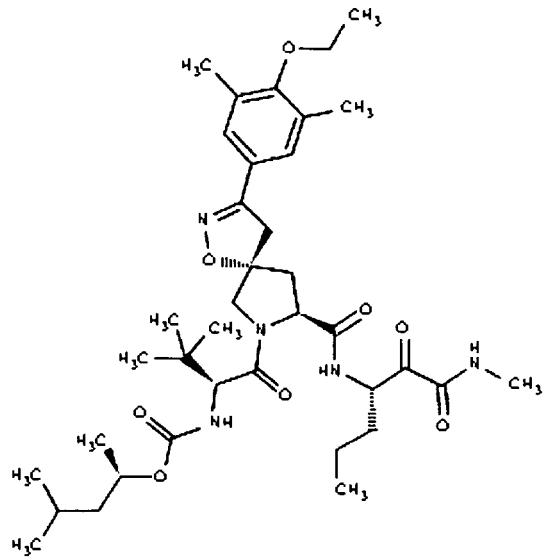
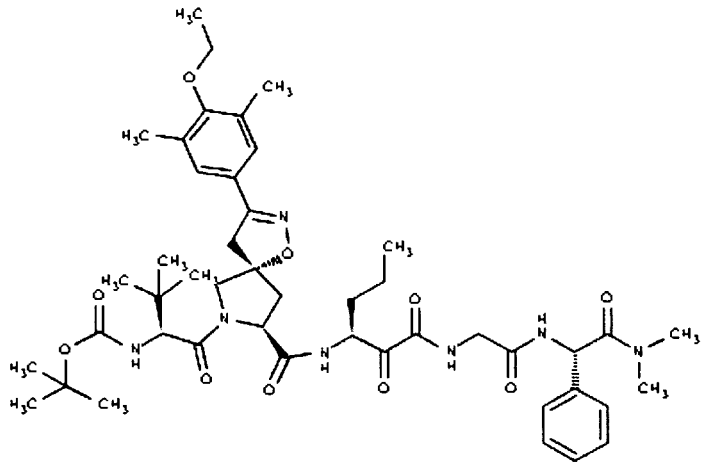
	1003
	1004

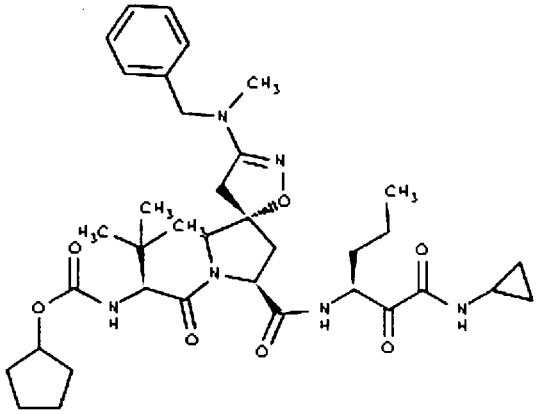
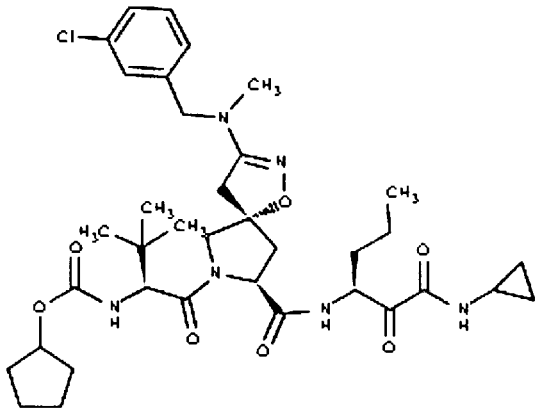
	1005
	1006

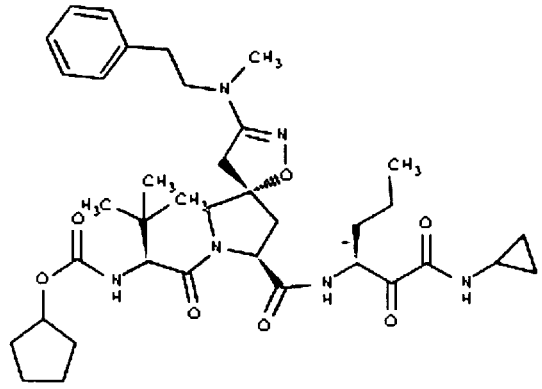
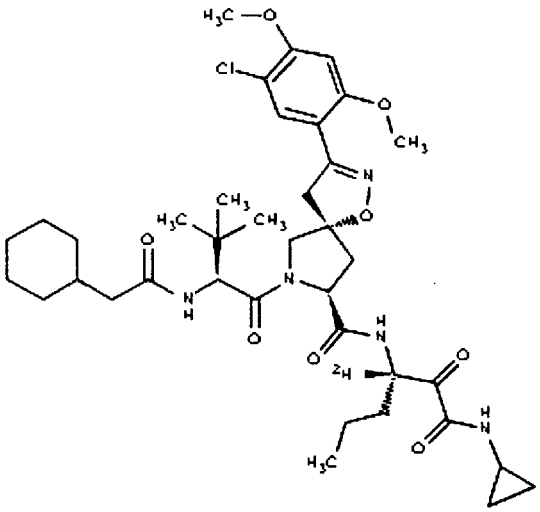
	1007
	1008

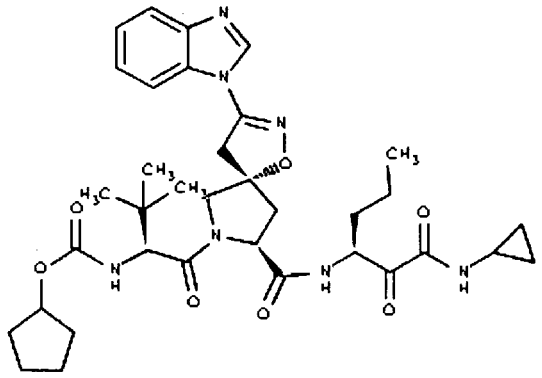
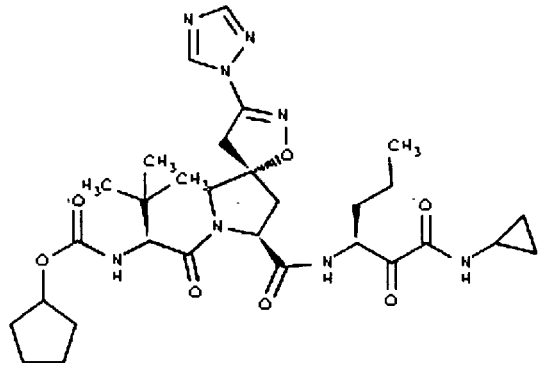
	1009
	1010

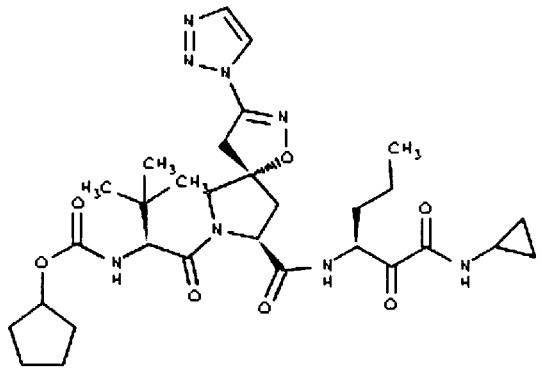
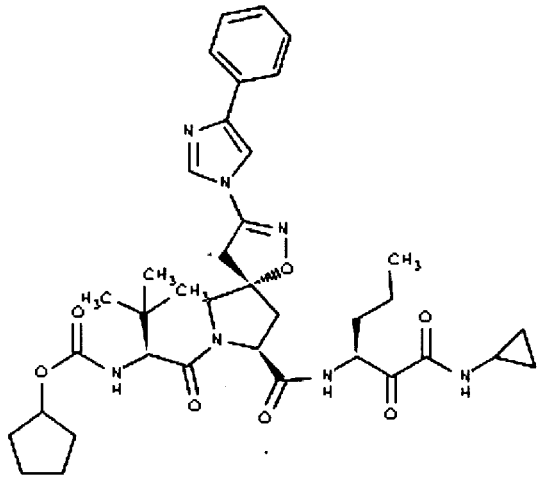
	1011
	1012

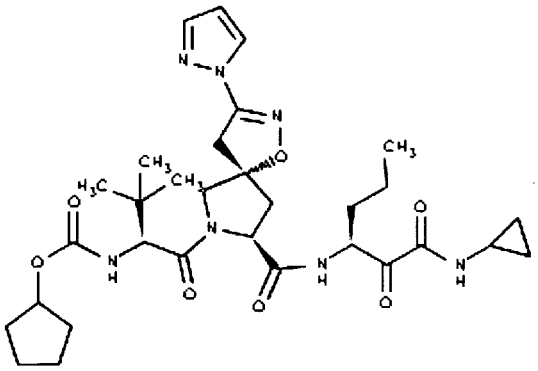
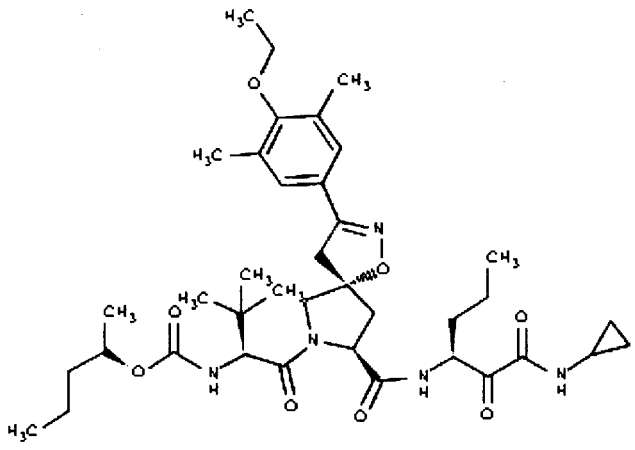
	1013
	1014

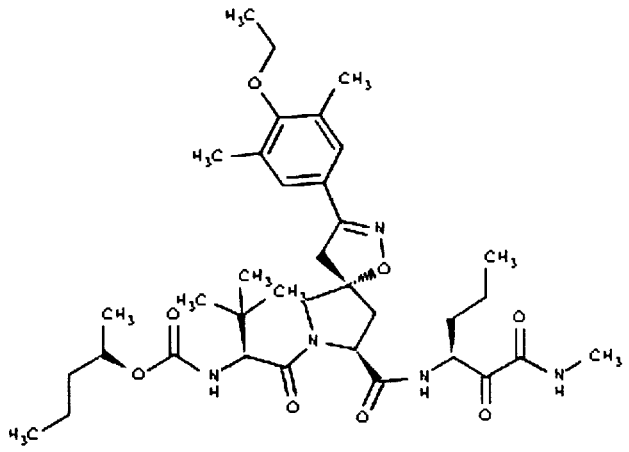
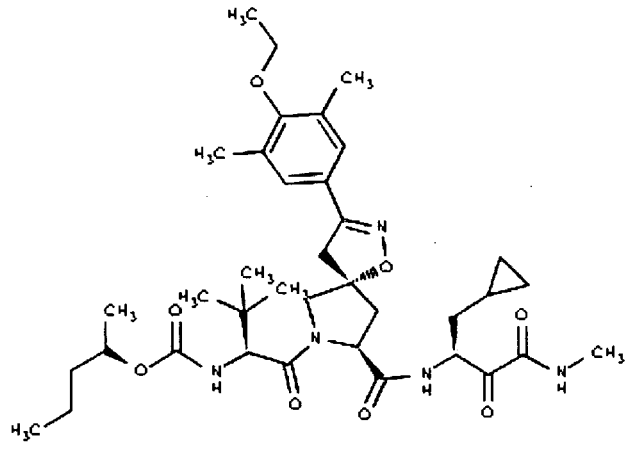
	1015
	1016

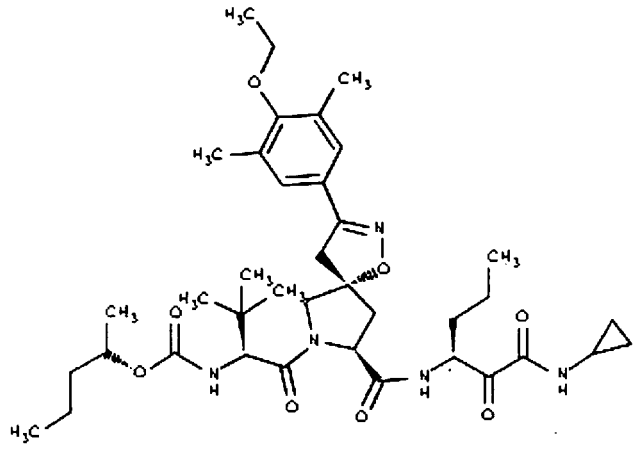
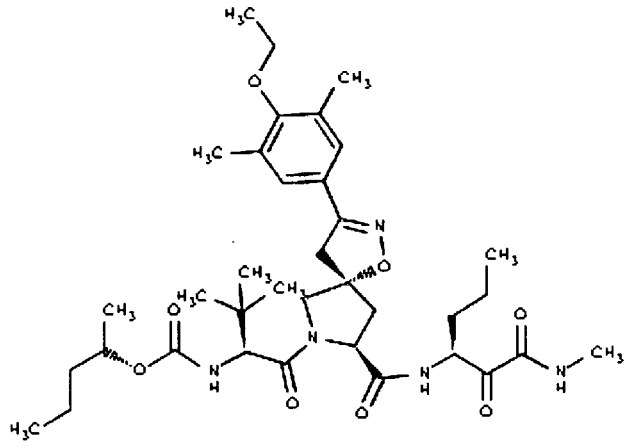
	1017
	1018

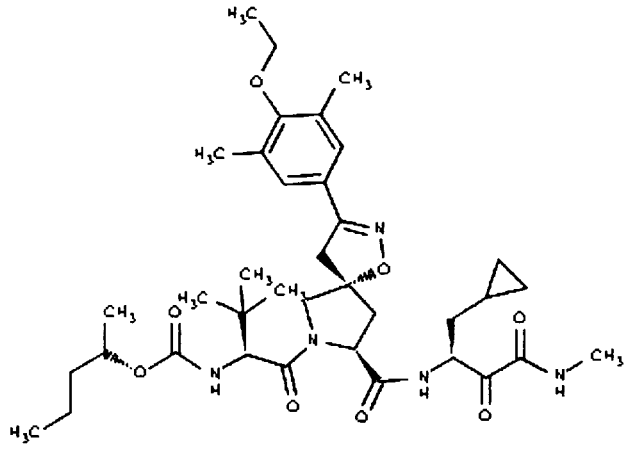
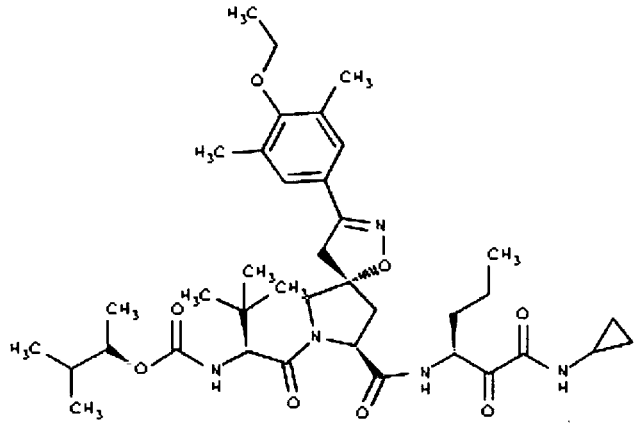
	1019
	1020

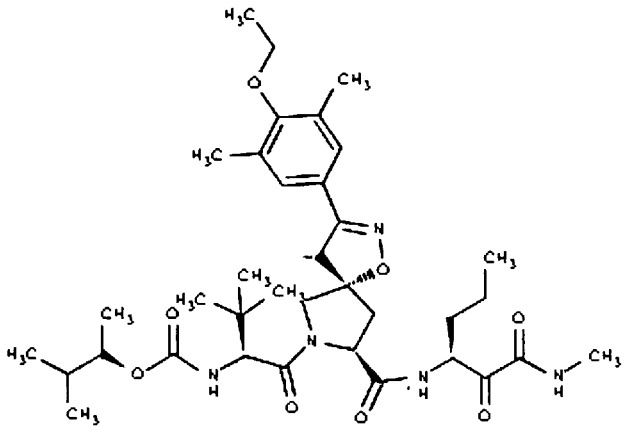
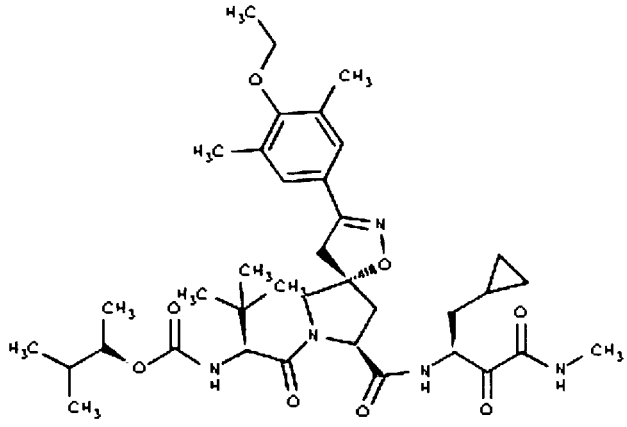
	1021
	1022

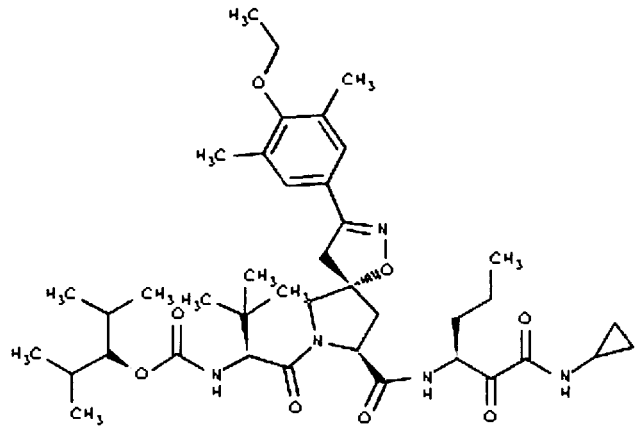
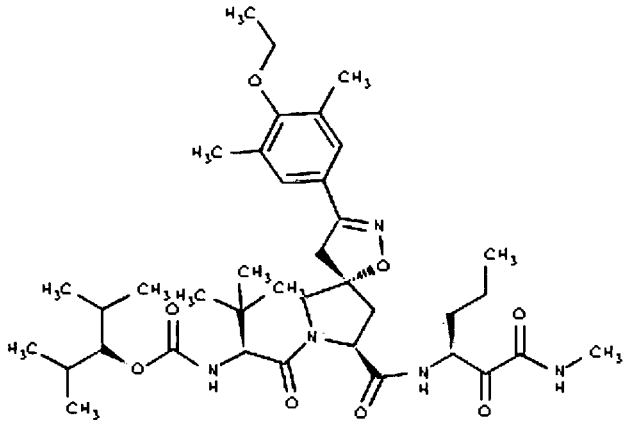
	1023
	1024

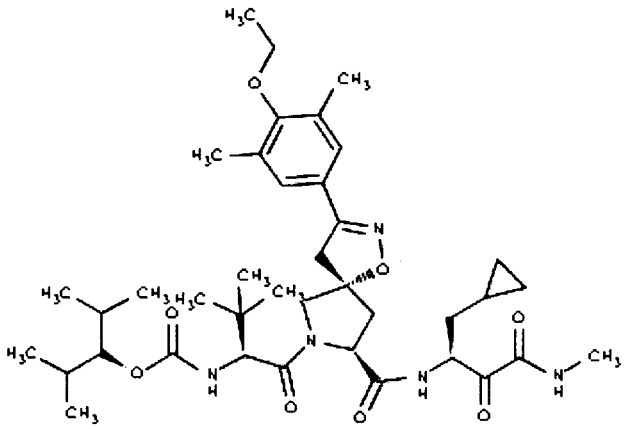
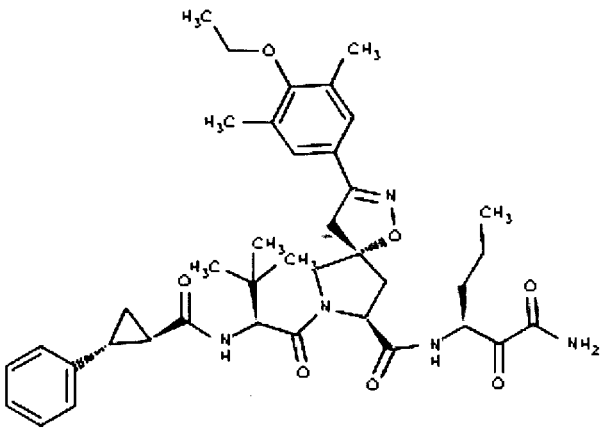
	1025
	1026

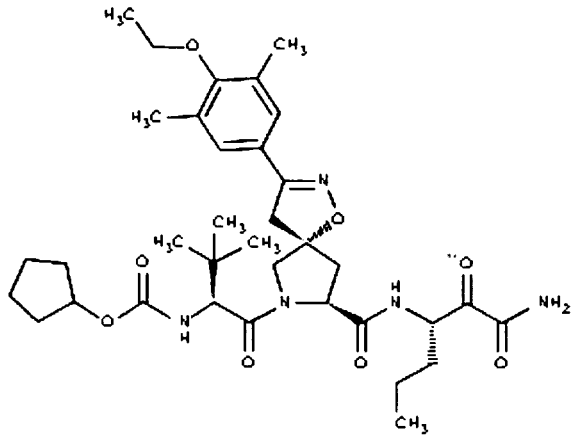
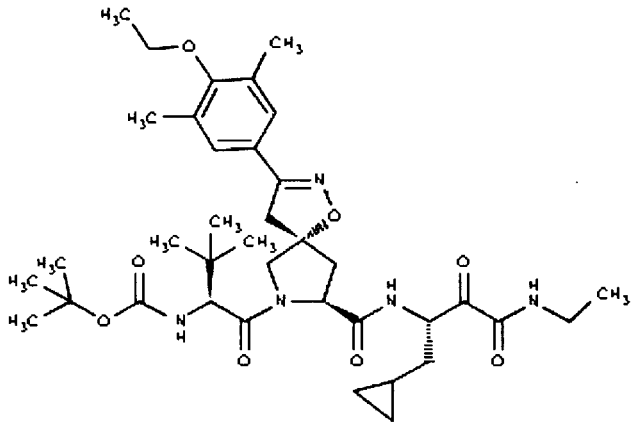
	1027
	1028

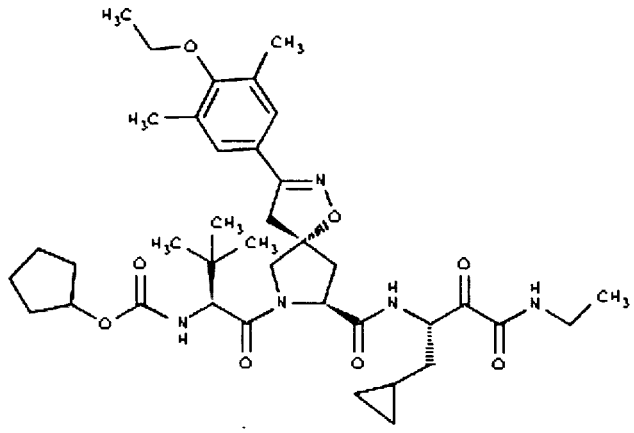
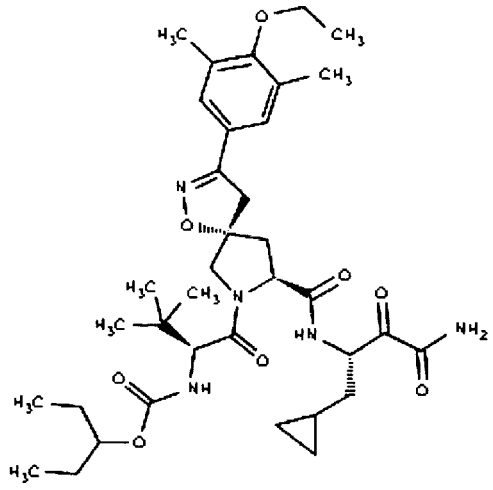
	1029
	1030

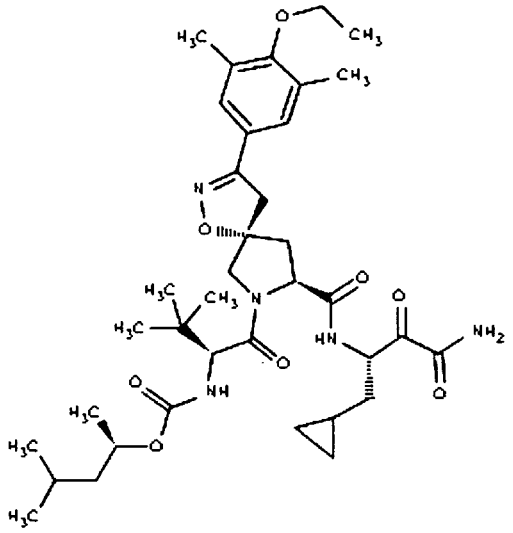
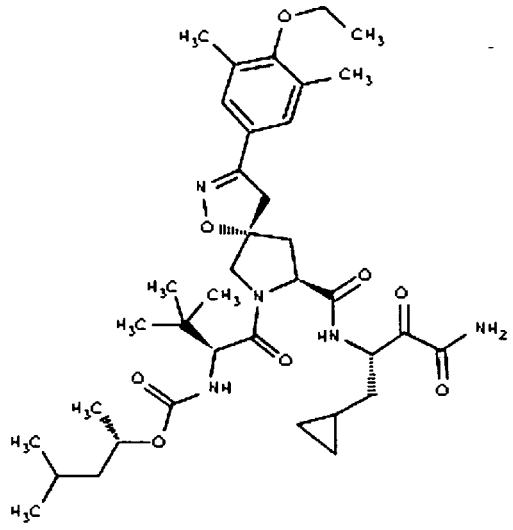
	1031
	1032

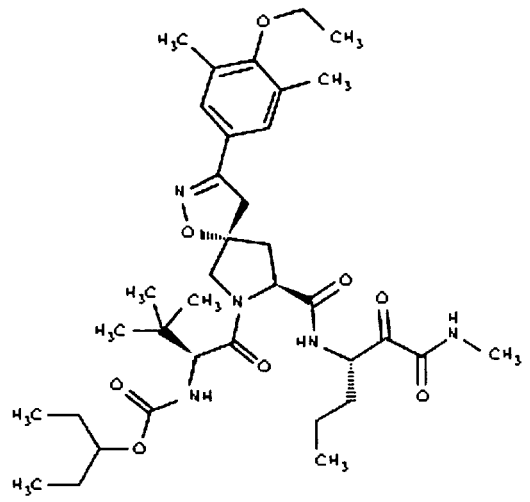
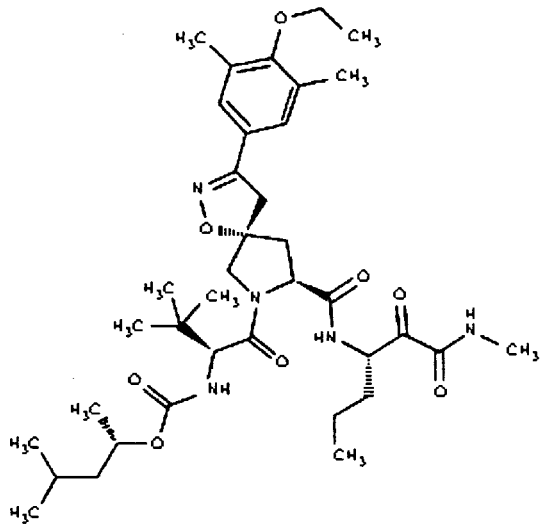
	1033
	1034

	1035
	1036

	1037
	1038

	1039
	1040

	1041
	1042

	1043
	1044

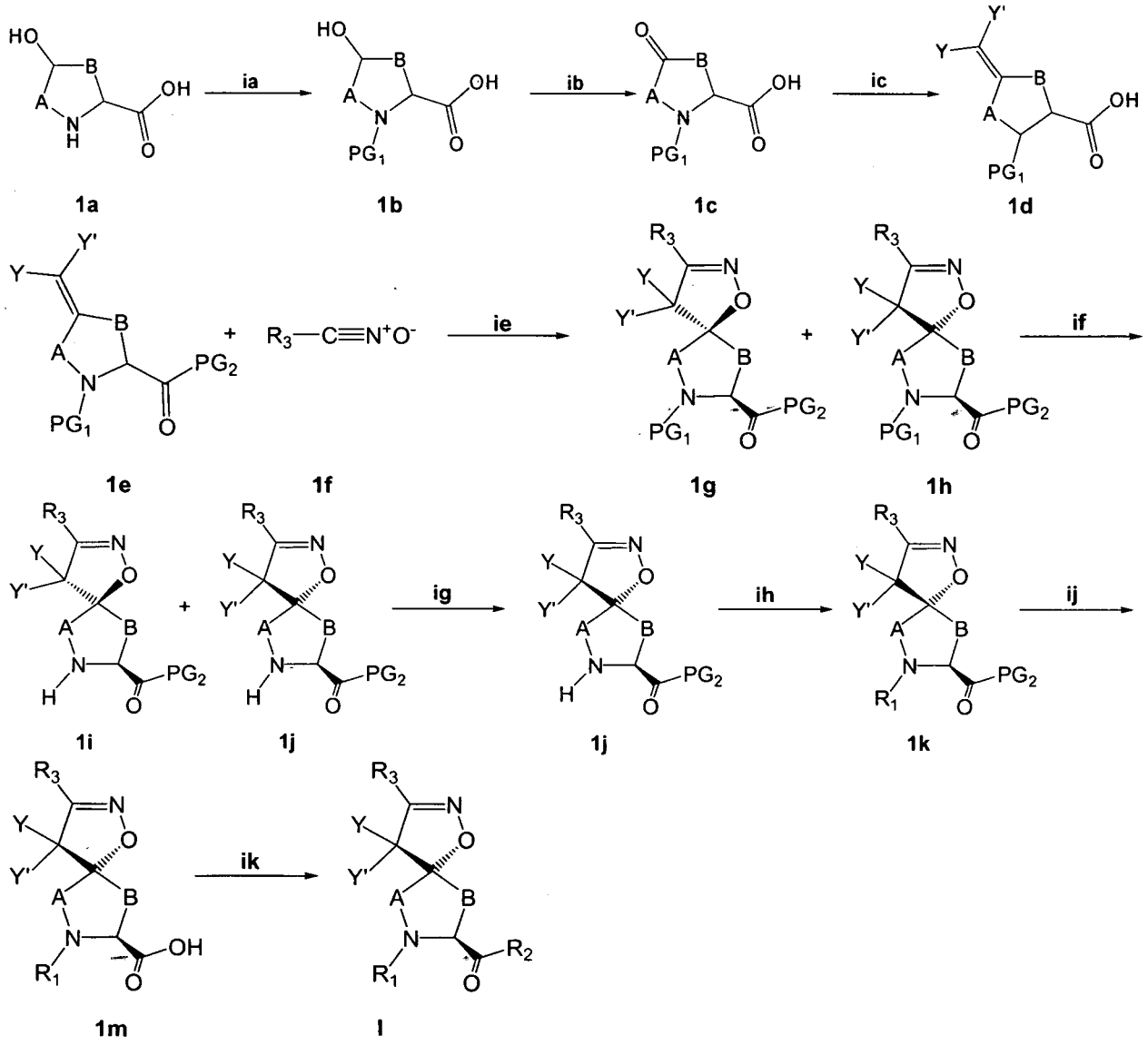
II. SYNTHESIS OF THE COMPOUNDS

[0221] Compounds of Formula I may be readily synthesized from commercially available starting materials using the exemplary synthetic routes provided below. Exemplary synthetic routes to produce compounds of Formula I are provided below in the Preparations, Methods, Examples, and Schemes. For example, the spiroisoxazoline moiety may be prepared by 1,3-dipolar addition between a nitrile oxide and a methylene proline as reported by Kurth, M.J., et al., in J. Org. Chem., 2002, 67, pp. 5673-5677, and as illustrated in Scheme 1 below. The

nitrile oxides can be generated from cholooximes or nitroalkanes using known methods.

[0222] Scheme I provides a general representation of processes for preparing compounds of Formula (I). Its overall strategy is to construct a compound of formula **1h** followed by selective removal of the protecting group PG_1 in the presence of PG_2 to provide the intermediate **1j**. The substituent R_1 may then be coupled to **1j**, which provides intermediates of formula **1k** containing R_1. In some embodiments, R_1 may itself contain a protecting group which may be selectively removed in the presence of PG_2, followed by further elaboration. Subsequent to the addition of the R_1 moiety, the PG_2 group is removed to provide the intermediate **1m**. Coupling of **1m** with an R_2 moiety then provides the peptidomimetic compounds of Formula (**I**).

Scheme 1



[0223] Referring again to Scheme 1, in one example, the hydroxy proline **1a** is protected as the Boc derivative (i.e., step **ia**) to provide the protected proline **1b,** wherein PG_1 is t-

butyloxycarbonate, using known methods. See, e.g., T.W. Greene and P. G. M. Wuts, Protective Groups in Organic Synthesis, 3rd edition, John Wiley and Sons, Inc. (1999). Oxidation of **1b** (i.e., step **ib**) provides the keto-pyrrolidine acid **1c.** The oxidation is achieved with a suitable reagent such as, for example, sodium hypochlorite in the presence of TEMPO. Next, in step **ic,** the keto-pyrrolidine acid **1c** is reacted with a Wittig reagent such as, for example, a triphenylphosphonium ylid of the formula $(Ph)_3P=C(Y)(Y')$ and using known conditions, to provide an exomethylene compound of formula **1d**. Use of the free acid **1c** to provide the corresponding free acid **1d** is advantageous as the acid **1d** may be expediently purified from neutral or basic by-products by simple extraction of **1d** into aqueous basic solution. The acid **1d** is subsequently protected (step **id**) with a suitable protecting group such as, for example, a t-butyl ester under known conditions (ibid) to provide the intermediate **1e**.

[0224] Reaction of **1e** with a nitrile oxide **1f** provides a mixture of the syn and anti isomers of the spiroisoxazolines **1g** and **1h**. As referred to herein, syn- means that the 2-carboxyl moiety of the proline ring and the oxygen of the isoxazoline ring are on the same side of a plane as described by the proline ring. The term anti- means that the 2-carboxyl moiety of the proline ring and the oxygen of the isoxazoline ring are on the opposite side of a plane as described by the proline ring. Thus, **1g** represents a syn- compound of the invention and **1h** represents an anti- compound of the invention.

[0225] In some embodiments, when PG_1 is Boc and PG_2 is *t*-butoxy, selective removal of the protecting group PG_1 from **1g** and **1h** in the presence of the protecting group PG_2 may be achieved with a sulfonic acid such as, for example, methane sulfonic acid in a suitable organic solvent at temperatures from about -40 °C to about 40 °C, from about -20 °C to about 20 °C and from about -5 °C to about 5 °C. Suitable organic solvents include, for example, methylene chloride and tetrahydrofuran.

[0226] The isomers **1i** and **1j** may be separated advantageously by crystallization of a mixture of the corresponding organic acid salts which avoids more complicated methods such as, e.g., chromatography. Suitable organic salts include those of organic carboxylic acids, e.g., acetic acid, optionally substituted benzoic acids, tartaric acid, malonic acid, fumaric acid, oxalic acid, mandelic acid, citric acid, *p*-toluoyl tartaric acid and maleic acid; organic sulfonic acids, e.g., methane sulfonic acid, optionally substituted benzene sulfonic acids, trifluromethane sulfonic acid and camphor sulfonic acid.

[0227] A single spiroisoxazoline isomer, for example **1j**, is coupled with an acid R_1COOH in the presence of a coupling reagent such as, for example, EDCI to provide the intermediate

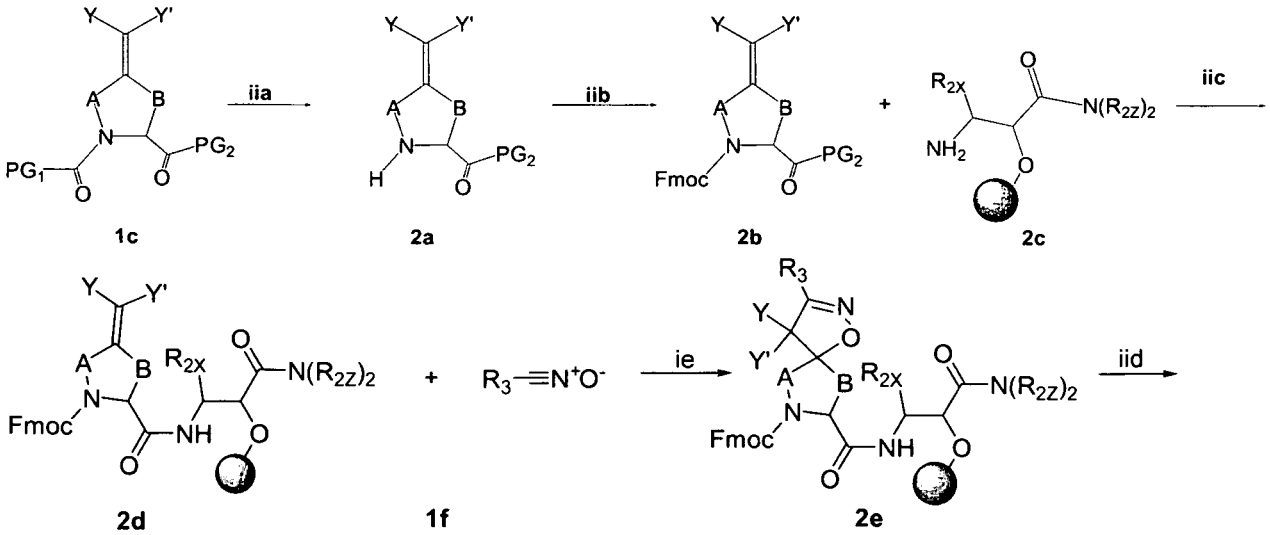
spiroisoxazoline **1k**. Selective removal of the protecting group PG_2 of **1k** to give **1m** with minimum racemization or cleavage of the R_1 side chain is achieved by a suitable mineral acid in a suitable organic solvent at temperatures from about -40 °C to about 40 °C, from about -20° C to about 20 °C and from about -5 °C to about 5 °C. Suitable mineral acids include, for example, concentrated hydrochloric acid or concentrated sulfuric acid. Suitable organic solvents include, for example, methylene chloride and tetrahydrofuran. The spiroisoxazoline **1m** is then coupled with an amine moiety R_2 to provide the compounds of Formula I.

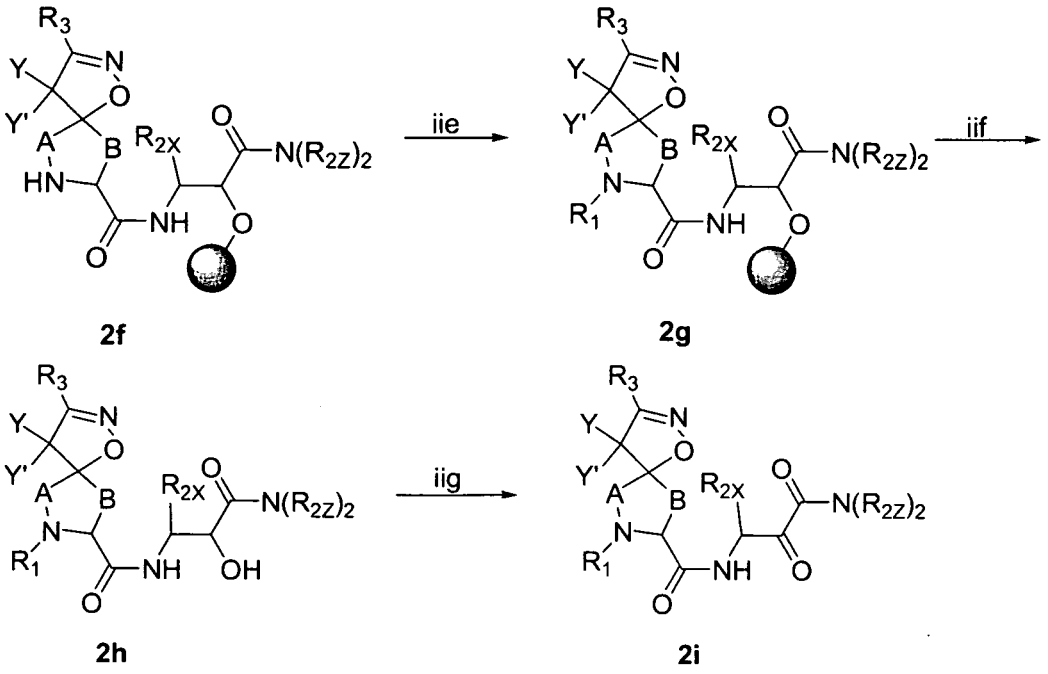
[0228] Referring again to Scheme 1, $PG_1(CO)$- can be an amine protecting group, wherein PG_1 is, for example, methoxycarbonyl, t-butyloxycarbonyl, 9-flourenylmethyloxycarbonyl, or benzyloxycarbonyl. $PG_2(CO)$- can be an acid or acid protecting group wherein PG_2 is, for example, -OH, methoxy, t-butyloxy or benzyloxy.

[0229] Each of PG_1 and PG_2 groups may be incorporated into the core spiroisoxazoline structure either individually or together using known methods and as further described herein. For example, if the desired R_1 substituted is a group other than a PG_1 group (e.g., a protecting group), the PG_1 group may be removed to provide a compound with a free amine group. That amine group and an appropriate moiety may be coupled under known coupling conditions to provide a compound wherein R_1 is a moiety of a protease inhibitor. For example, if the PG_2 moiety is protected, the protecting group may be removed and an R_2 moiety may be incorporated.

[0230] Another method for producing compounds of the present invention is illustrated below in Scheme 2.

Scheme 2





[0231] Referring to Scheme 2, the symbol represents a polymeric resin to which reactants are bound by a functionality that allows further modification and subsequent removal of the product from the resin. A suitable resin is a polymer bound dihydropyran (DHP) resin as described by Ellman et.al. in Tetrahedron Letters, 1994, 35, 9333.

[0232] In step **iia**, simultaneous deprotection of both the amine and acid may be achieved by contacting the proline **1e** with an acid, for example, trifluoroacetic acid in methylene chloride to give the amino acid **2a**. Reaction of **2a**, step **iib**, with an activated Fmoc derivative, for example, N-(9H-Fluoren-9ylmethoxycarbonyloxy)succinimide (Fmoc-OSu), in the presence of a mild inorganic base, such as sodium carbonate, gives the Fmoc derivative **2b**.

[0233] Preparation of the resin bound peptide **2d** may be accomplished by reacting the Fmoc derivative **2b** with the DHP resin bound amino-alcohol **2c**, step iiic, which reacts with the free acid **2b**, in the presence of a coupling reagent such as, for example, O-Benzotriazole-N,N,N',N'-tetramethyl-uronium-hexafluoro-phosphate (HBTU), a racemization suppressant, such as 1-hydroxybenzotriazole (HOBT) and a tertiary amine, such as di-isopropylethyl amine (DIEA).

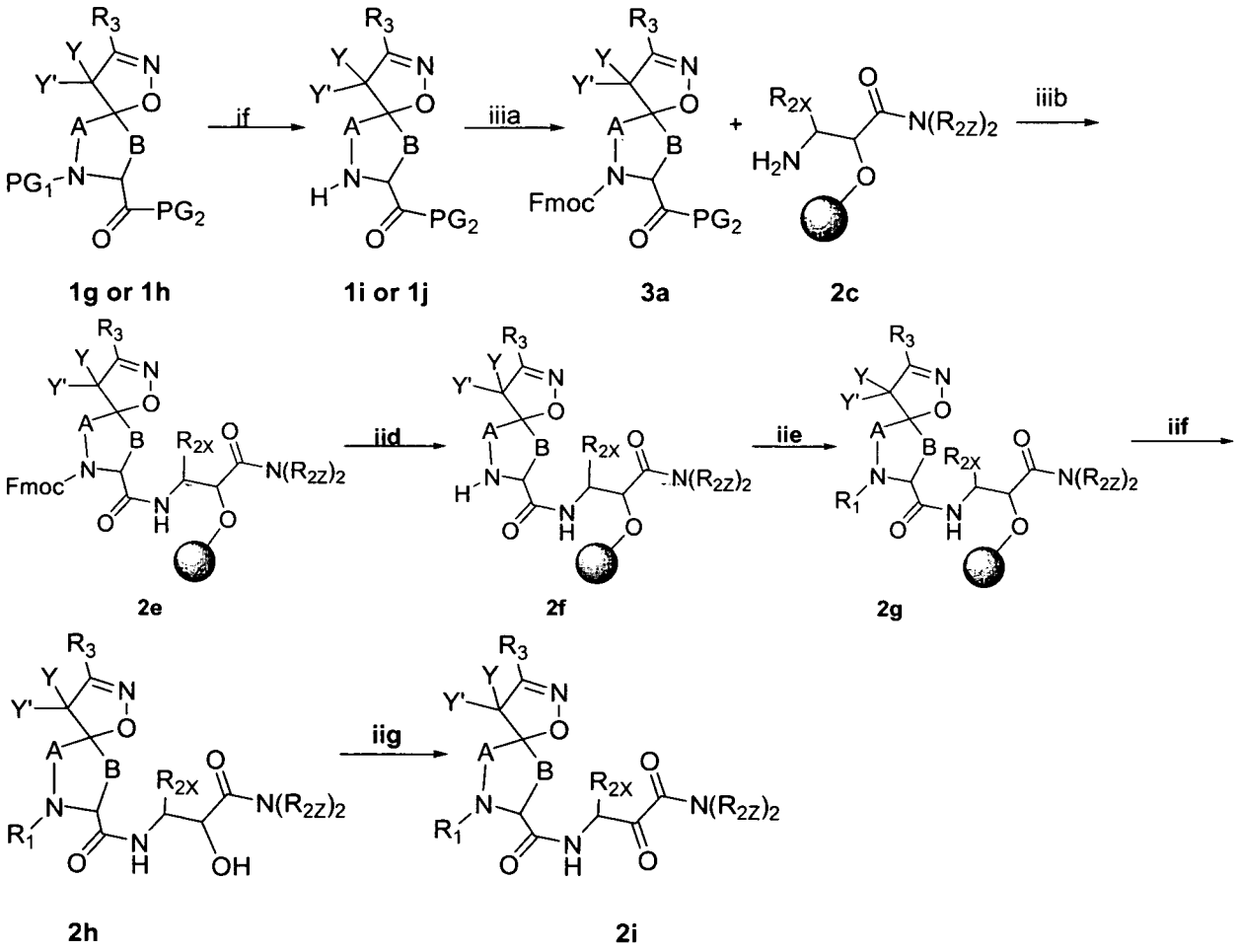
[0234] As in Scheme 1, an R_3-substituted nitrile oxide **1f** may undergo a dipolar cycloaddition reaction with the resin bound peptide **2d** to provide two isomers, syn- and anti-, of the compound **2e**. Next in step **iid**, the Fmoc protecting group is removed by contacting **2e** with a secondary amine such as, for example, piperidine in a polar solvent such as dimethylformamide to give **2f**. Formation of the peptide **2g**, via step **iie**, can be achieved

through reaction of **2f** with a carboxylic acid in the presence of a coupling reagent such as HBTU, a racemization suppressant such as HOBt, and a tertiary amine such as DIEA. Cleavage of the peptide-resin **2g**, step **iif**, to give the alpha-hydroxy-amide **2h**, can be achieved by contacting **2g** with a strong acid such as, for example, trifluoroacetic acid and water.

[0235] In the final step, **iig**, the alpha-hydroxy-amide **2h** is oxidized to **2i** using a Dess-Martin periodinane oxidation or a Pfitzner-Moffat oxidation.

[0236] Alternatively, compounds of Formula I may be prepared using resin bound reagents as illustrated below in Scheme 3.

Scheme 3



[0237] In Scheme 3, the selective removal of the PG_1 in the presence of PG_2 (step **if**) provides spiroisoxazoline isomer(s) **1i** and/or **1j**. Reaction of **1i** and/or **1j**, in step **iiia**, with an activated Fmoc derivative, e.g., N-(9H-Fluoren-9-ylmethoxycarbonyloxy)succinimide (Fmoc-OSu), in the presence of a mild inorganic base, such as sodium carbonate, provides the Fmoc derivative **3a**.

[0238] Preparation of the resin bound peptide **2e** may be accomplished by reaction of the

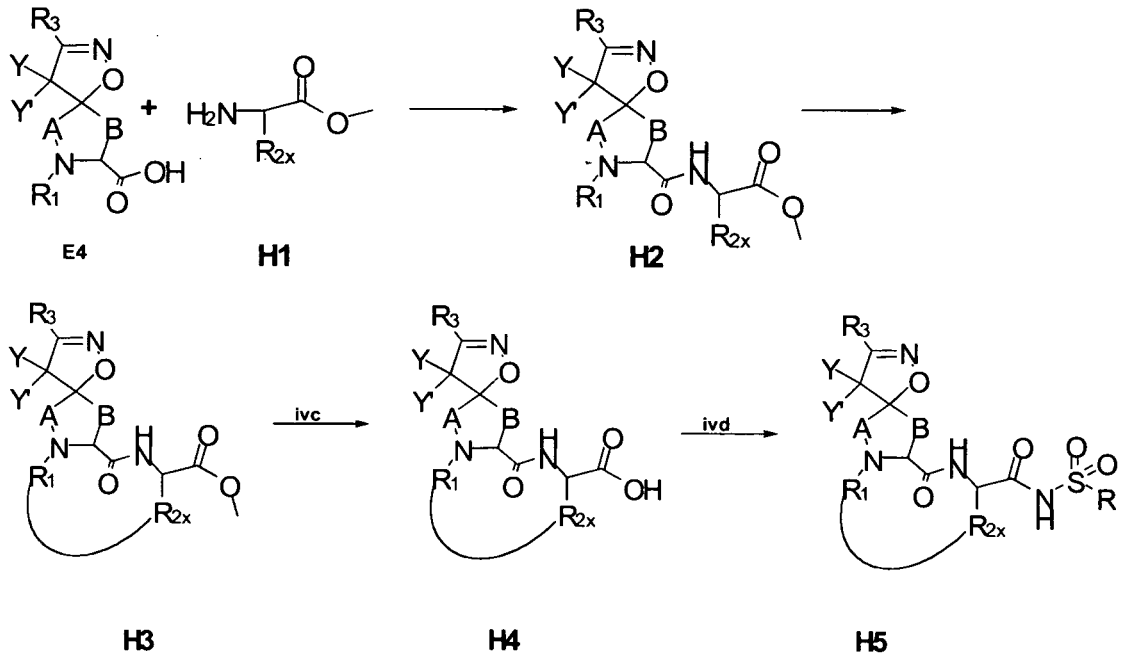
Fmoc derivative **3a** with the DHP resin bound amino-alcohol **2c**, via step **iiib**, which reacts with a free acid **3b**, in the presence of a coupling reagent (e.g., O-Benzotriazole-N,N,N',N'-tetramethyl-uronium-hexafluoro-phosphate (HBTU)), a racemization suppressant (e.g., 1-hydroxybenzotriazole (HOBT)), and a tertiary amine (e.g., di-isopropylethyl amine (DIEA)).

[0239] In step **iid**, the Fmoc protecting group is removed by contacting **2e** with a secondary amine such as, e.g., piperidine in a polar solvent such as dimethylformamide to give **2f**. Formation of the peptide **2g** can be achieved, e.g., by reacting **2f** with a carboxylic acid in the presence of a coupling reagent (e.g., HBTU), a racemization suppressant (e.g., HOBt) and a tertiary amine (e.g., DIEA). Cleavage of the peptide-resin **2g** to give the free peptide **2h** can be achieved, e.g., by contacting **2g** with a strong acid (e.g., trifluoroacetic acid) and water.

[0240] In the final step, **iig**, the alcohol of **2h** can be oxidized to **2i**, e.g., with Dess-Martin periodinane or sodium hypochlorite and TEMPO.

[0241] Scheme 4 below illustrates a synthetic pathway for compounds of Formula I in which R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted macrocyclic heterocycloaliphatic.

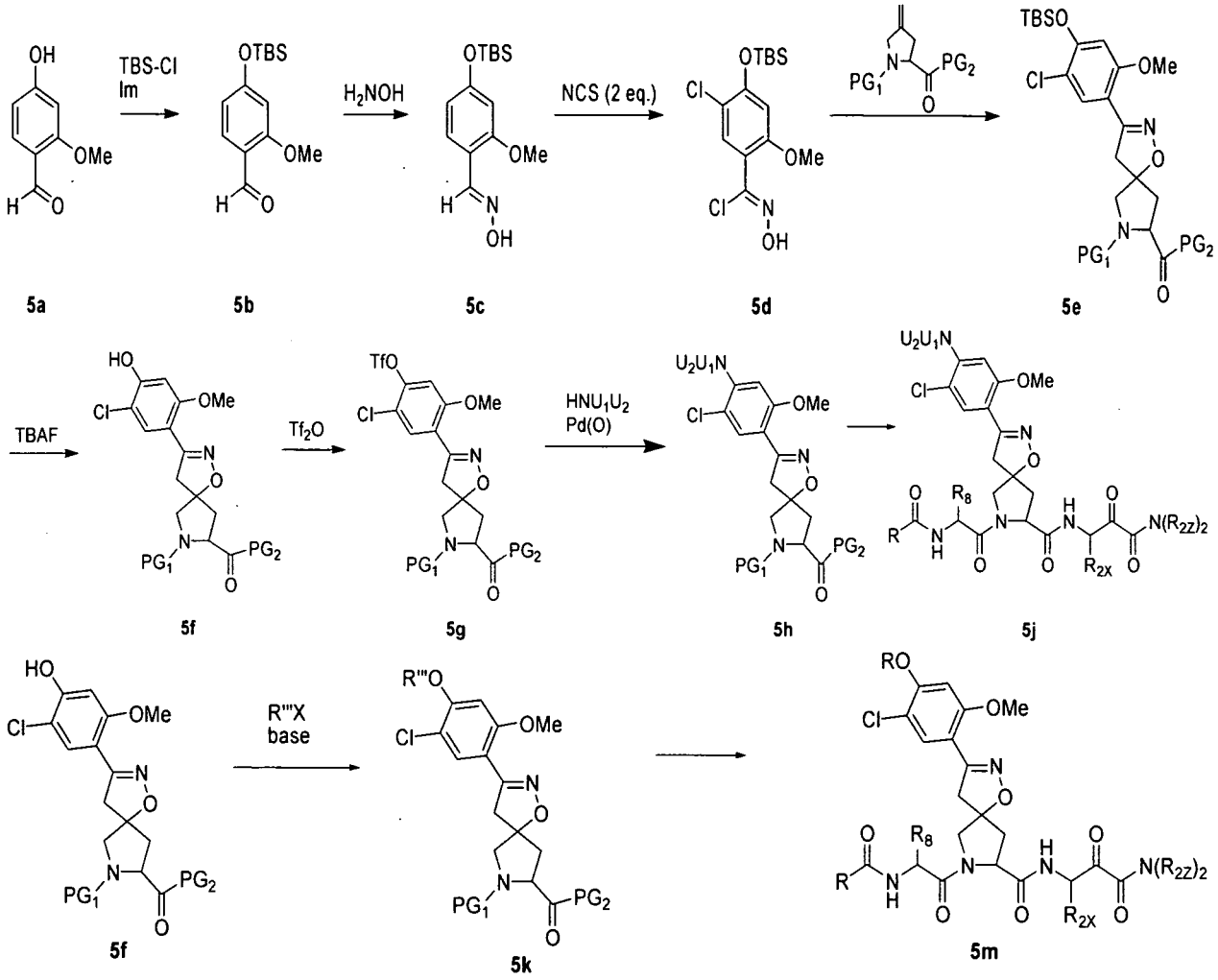
Scheme 4



[0242] Referring to Scheme 4, the spiroisoxazoline acid **E4** reacts with the amino ester **H1** in the presence of a coupling reagent to provide the intermediate **H2**. Macrocyclization of **H2** results in compound **H3**. Hydrolysis of the ester **H2** provides acid **H4**. Reaction of acid **H4** with a sulfonamide or sulfamide in the presence of a coupling reagent provides the product **H5**.

[0243] Shown below in Schemes 5, 6, 7, 8, and 9 are examples of total synthesis of compounds of formula (**I**) according to one of the methods described above.

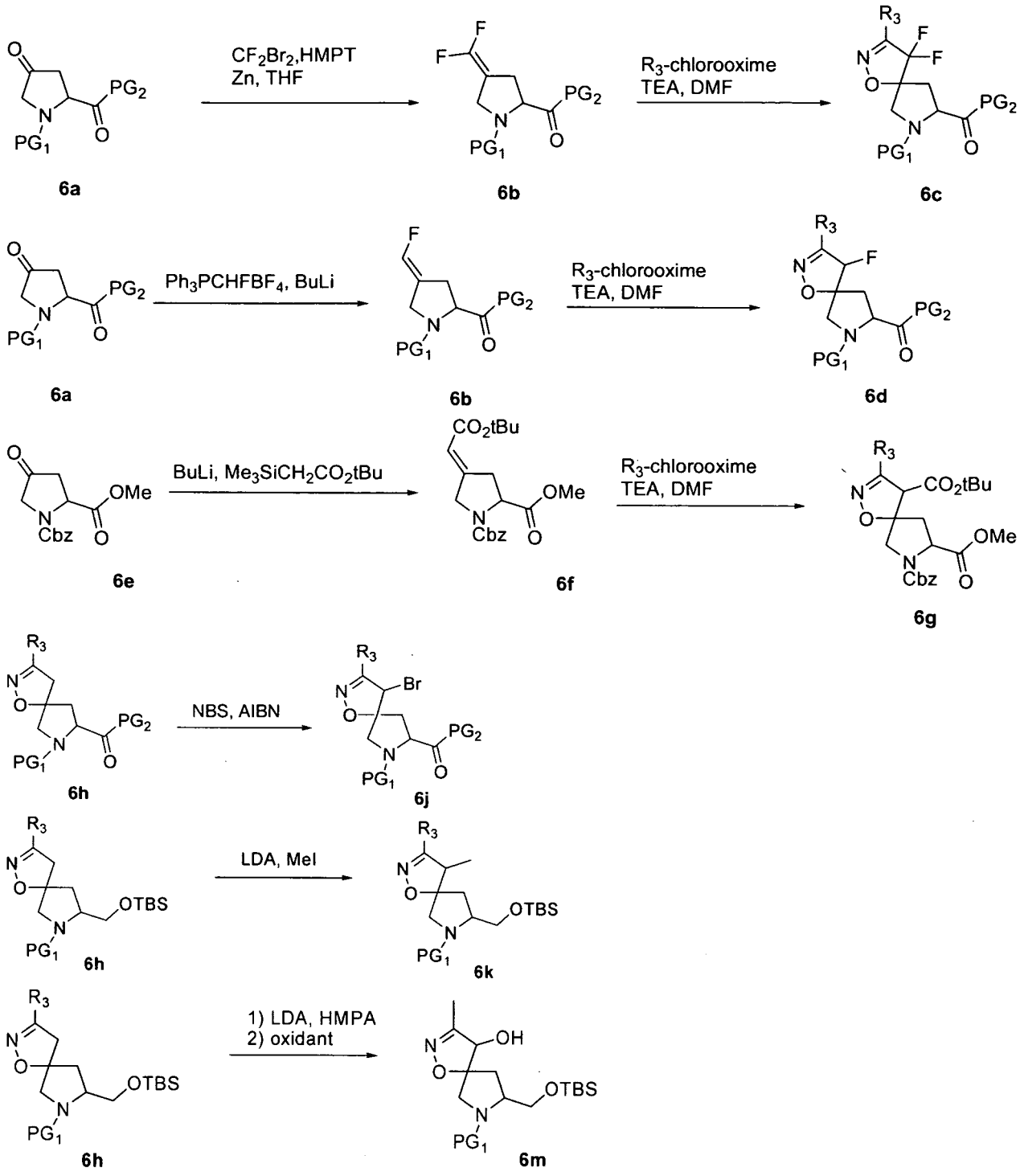
Scheme 5



[0244] Referring to Scheme 5, the protected *t*-butyldimethylsilyl-hydroxybenzaldehyde **5b** is converted to the hydroxamoyl chloride **5d** as previously described. Reaction of **5d** with the exomethylene pyrrolidine provides the spiroisoxazoline **5e**. Deprotection of **5e** to **5f** followed by reaction with triflic anhydride provides the triflate **5g**. Reaction of **5f** with an amine HNU_1U_2 provides the intermediate spiroisoxazoline **5h** which is converted to compounds of the invention as previously described.

[0245] Alternatively, the hydroxy-spiroisoxazoline intermediate **5f** may be alkylated to provide the intermediate **5k** which may be similarly converted to compounds of the invention.

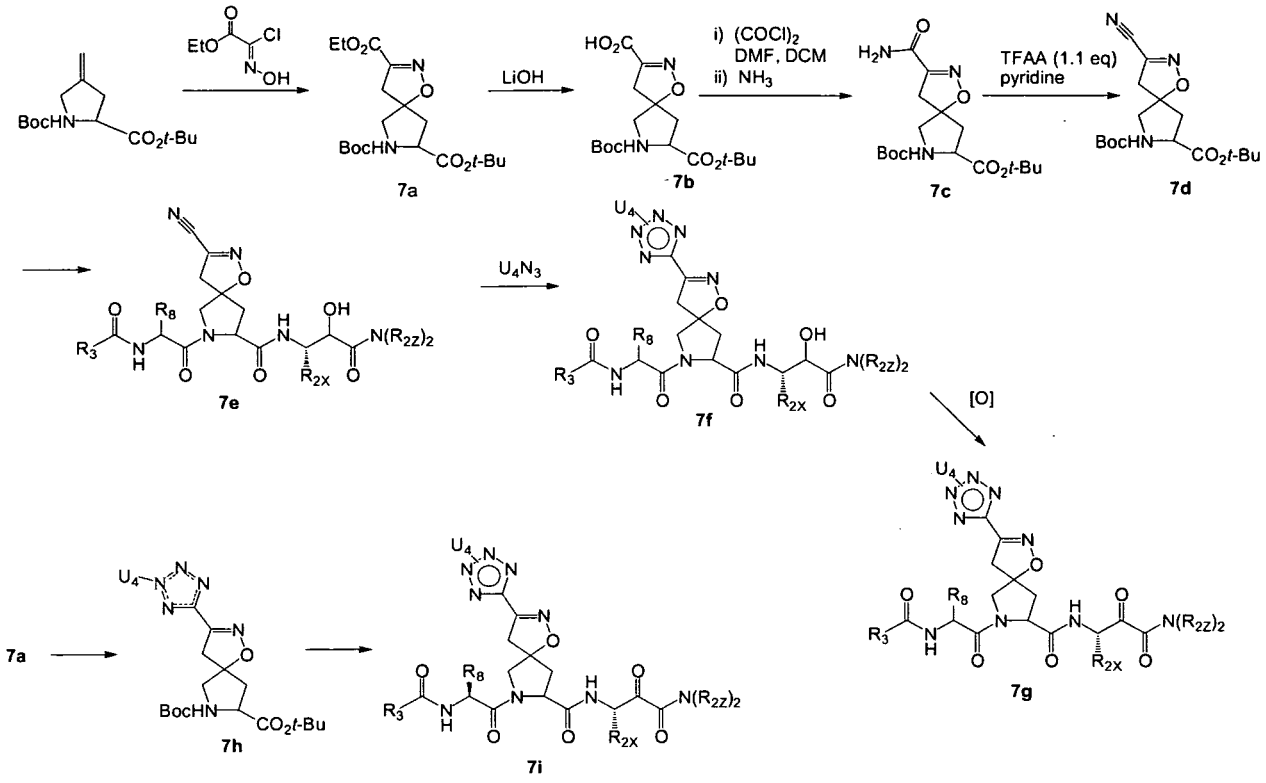
Scheme 6:



[0246] Referring to Scheme 6, reaction of the diprotected pyrrolidinone with difluorodibromomethane in the presence of HMPT and zinc provides the difluroexomethylene intermediate **6b**. Dipolar addition with the nitrile oxide **1f** as previously described provides the diflurospiroisoxazoline **6c**. In a similar fashion, the intermediates **6b** and **6f** are prepared from 6a and 6e respectively and converted to the corresponding substituted isooxazolines **6d** and **6g**.

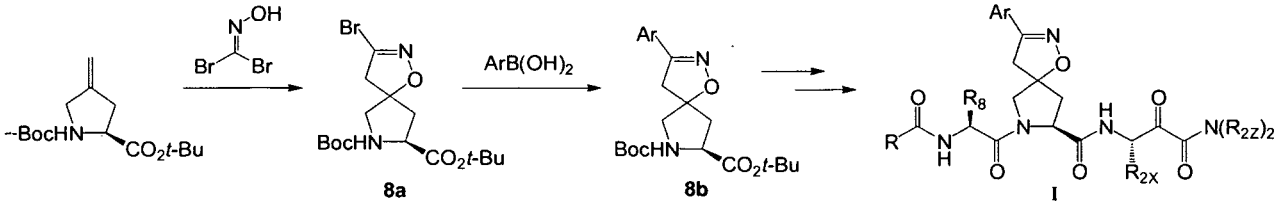
In other variations, the intermediate **6h** may be brominated to give **6j**, alkylated to provide **6k** or oxidized to provide **6m** using the reagents illustrated.

Scheme 7:



[0247] Referring to Scheme 7, dipolar addition of the exomethylene pyrrolidine shown with **1f** wherein R_3 is -COOEt, leads to the ester **7a**. Hydrolysis of the ethyl ester in **7a**, conversion to the acid chloride (not shown) and reaction with ammonia provides the amide **7c**. Reaction of **7c** with trifluroacetic anhdride provides the nitrile **7d** which is converted to the peptidic intermediate **7e** by methods previously described. The intermediate **7e** reacts with an azide U_4N_3 to provide the tetrazole **7f** which is oxidized to a compound of the invention **7g**. In a variation of this scheme, the ester **7a** may be converted to the triazole **7h** and subsequently to compounds of the invention **7i**.

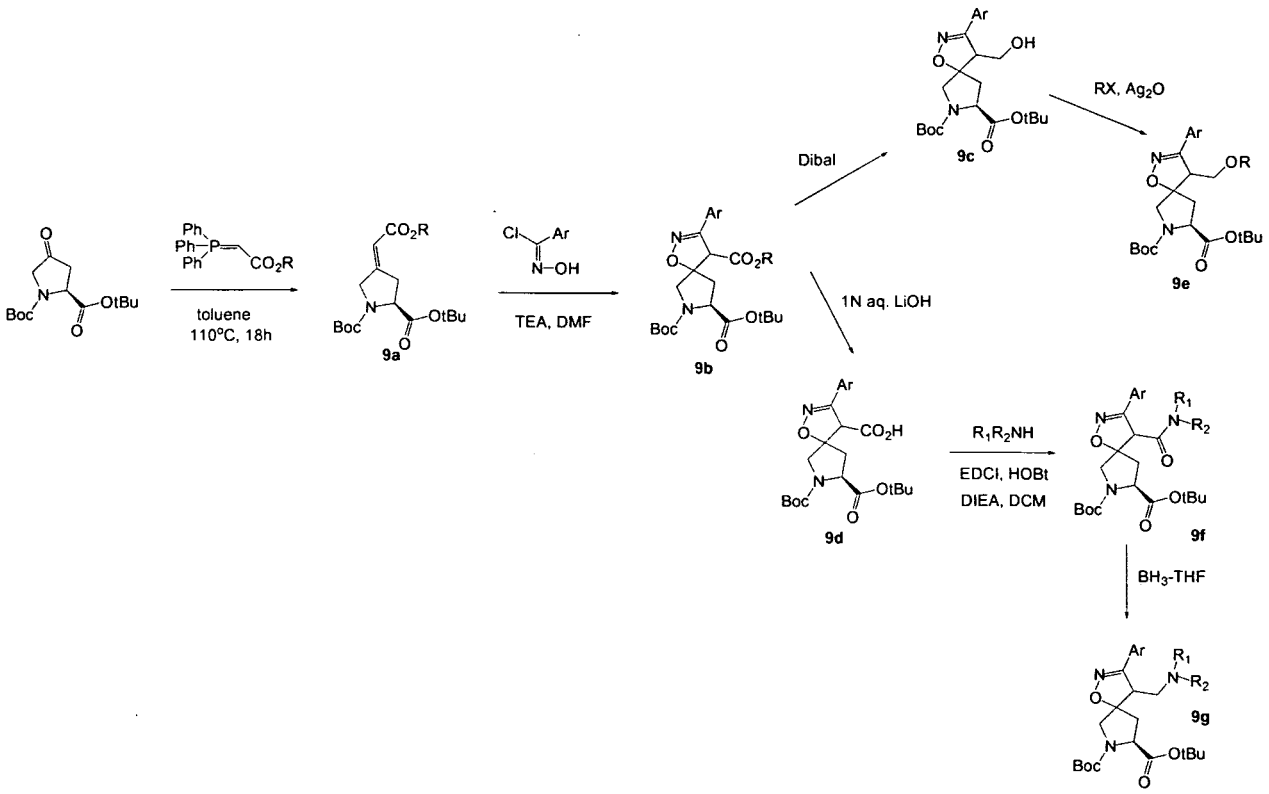
Scheme 8:



[0248] Referring to Scheme 8, dipolar addition as previously described but using hydroxycarbonimidic dibromide provides the bromoisoxazoline **8a**. Reaction of **8a** with an

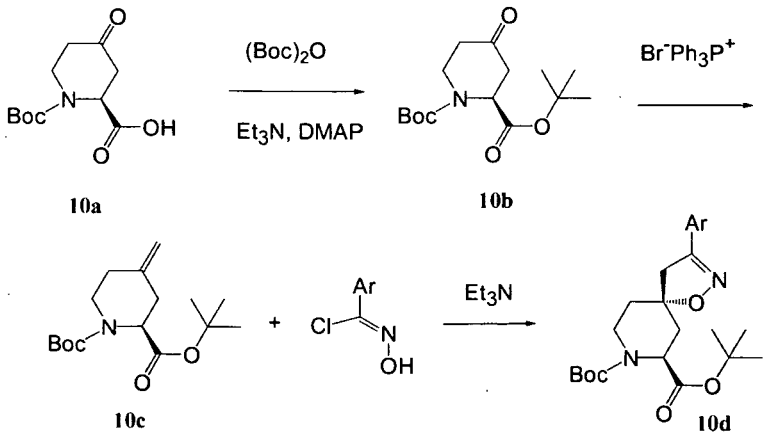
arylboronic acid in the presence of a palladium catalyst (Suzuki conditions) provides the intermediate **8b** which is converted to compounds of the invention by methods previously described. The AR in step 8a and 8b represents aryl or heteroaryl.

Scheme 9:



[0249] Referring to Scheme 9, the Wittig product **9a** undergoes a dipolar addition to provide the spiroisoxazoline **9b**. Reduction of **9b** with, for example, DIBAL provides the alcohol **9c** which may be alkylated to provide the intermediate **9e** which subsequently may be converted to compounds of the invention by methods previously described. Hydrolysis of ester **9b** with, e.g., LiOH, will provide carboxylic acid **9d** which can be converted to compounds of formula I as described herein.

Scheme 10:



[0250] Referring to scheme 10, the diprotected piperidinone **10b** undergoes a Wittig type reaction to form the exomethylene compound **10c** which undergoes dipolar addition as previously described to provide a 4.5 spiroisoxazoline **10d** which may be converted to compounds of the invention as previously described.

III. FORMULATIONS, ADMINISTRATIONS, AND USES

[0251] Another embodiment of this invention provides a pharmaceutical composition comprising a compound of formula (**I**) or pharmaceutically acceptable salts or mixtures of salts thereof. According to another embodiment, the compound of formula (**I**) is present in an amount effective to decrease the viral load in a sample or in a patient, wherein said virus encodes a serine protease necessary for the viral life cycle, and a pharmaceutically acceptable carrier.

[0252] If pharmaceutically acceptable salts of the compounds of this invention are utilized in these compositions, those salts are preferably derived from inorganic or organic acids and bases. Included among such acid salts are the following: acetate, adipate, alginate, aspartate, benzoate, benzene sulfonate, bisulfate, butyrate, citrate, camphorate, camphor sulfonate, cyclopentane-propionate, digluconate, dodecylsulfate, ethanesulfonate, fumarate, glucoheptanoate, glycerophosphate, hemisulfate, heptanoate, hexanoate, hydrochloride, hydrobromide, hydroiodide, 2 hydroxyethanesulfonate, lactate, maleate, methanesulfonate, 2 naphthalenesulfonate, nicotinate, oxalate, pamoate, pectinate, persulfate, 3 phenyl propionate, picrate, pivalate, propionate, succinate, tartrate, thiocyanate, tosylate and undecanoate. Base salts include ammonium salts, alkali metal salts, such as sodium and potassium salts, alkaline earth metal salts, such as calcium and magnesium salts, salts with organic bases, such as dicyclohexylamine salts, N methyl D glucamine, and salts with amino acids such as arginine, lysine, and so forth.

[0253] Also, the basic nitrogen containing groups may be quaternized with such agents as lower alkyl halides, such as methyl, ethyl, propyl, and butyl chloride, bromides and iodides; dialkyl sulfates, such as dimethyl, diethyl, dibutyl and diamyl sulfates, long chain halides such as decyl, lauryl, myristyl and stearyl chlorides, bromides and iodides, aralkyl halides, such as benzyl and phenethyl bromides and others. Water or oil soluble or dispersible products are thereby obtained.

[0254] The compounds utilized in the compositions and methods of this invention may also be modified by appending appropriate functionalities to enhance selective biological properties. Such modifications are known in the art and include those which increase biological penetration into a given biological system (e.g., blood, lymphatic system, central

nervous system), increase oral availability, increase solubility to allow administration by injection, alter metabolism and alter rate of excretion.

[0255] Pharmaceutically acceptable carriers that may be used in these compositions include, but are not limited to, ion exchangers, alumina, aluminum stearate, lecithin, serum proteins, such as human serum albumin, buffer substances such as phosphates, glycine, sorbic acid, potassium sorbate, partial glyceride mixtures of saturated vegetable fatty acids, water, salts or electrolytes, such as protamine sulfate, disodium hydrogen phosphate, potassium hydrogen phosphate, sodium chloride, zinc salts, colloidal silica, magnesium trisilicate, polyvinyl pyrrolidone, cellulose based substances, polyethylene glycol, sodium carboxymethylcellulose, polyacrylates, waxes, polyethylene polyoxypropylene block polymers, polyethylene glycol and wool fat.

[0256] According to another embodiment, the compositions of this invention are formulated for pharmaceutical administration to a mammal. In one embodiment said mammal is a human being.

[0257] Such pharmaceutical compositions of the present invention may be administered orally, parenterally, by inhalation spray, topically, rectally, nasally, buccally, vaginally or via an implanted reservoir. The term "parenteral" as used herein includes subcutaneous, intravenous, intramuscular, intra articular, intra synovial, intrasternal, intrathecal, intrahepatic, intralesional and intracranial injection or infusion techniques. Preferably, the compositions are administered orally or intravenously.

[0258] Sterile injectable forms of the compositions of this invention may be aqueous or oleaginous suspension. These suspensions may be formulated according to techniques known in the art using suitable dispersing or wetting agents and suspending agents. The sterile injectable preparation may also be a sterile injectable solution or suspension in a non toxic parenterally acceptable diluent or solvent, for example as a solution in 1,3 butanediol. Among the acceptable vehicles and solvents that may be employed are water, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium. For this purpose, any bland fixed oil may be employed including synthetic mono- or diglycerides. Fatty acids, such as oleic acid and its glyceride derivatives are useful in the preparation of injectables, as are natural pharmaceutically-acceptable oils, such as olive oil or castor oil, especially in their polyoxyethylated versions. These oil solutions or suspensions may also contain a long-chain alcohol diluent or dispersant, such as carboxymethyl cellulose or similar dispersing agents which are commonly used in the formulation of pharmaceutically acceptable dosage forms

including emulsions and suspensions. Other commonly used surfactants, such as Tweens, Spans and other emulsifying agents or bioavailability enhancers which are commonly used in the manufacture of pharmaceutically acceptable solid, liquid, or other dosage forms may also be used for the purposes of formulation.

[0259] In one embodiment, dosage levels of between about 0.01 and about 100 mg/kg body weight per day of the protease inhibitor compounds described herein are useful in a monotherapy for the prevention and treatment of antiviral, particularly anti-HCV mediated disease. In another embodiment, dosage levels of between about 0.5 and about 75 mg/kg body weight per day of the protease inhibitor compounds described herein are useful in a monotherapy for the prevention and treatment of antiviral, particularly anti-HCV mediated disease. Typically, the pharmaceutical compositions of this invention will be administered from about 1 to about 5 times per day or alternatively, as a continuous infusion. Such administration can be used as a chronic or acute therapy. The amount of active ingredient that may be combined with the carrier materials to produce a single dosage form will vary depending upon the host treated and the particular mode of administration. A typical preparation will contain from about 5% to about 95% active compound (w/w). In one embodiment, such preparations contain from about 20% to about 80% active compound.

[0260] When the compositions of this invention comprise a combination of a compound of formula I and one or more additional therapeutic or prophylactic agents, both the compound and the additional agent should be present at dosage levels of between about 10 to 100% of the dosage normally administered in a monotherapy regimen. In another embodiment, the additional agent should be present at dosage levels of between about 10 to 80% of the dosage normally administered in a monotherapy regimen.

[0261] The pharmaceutical compositions of this invention may be orally administered in any orally acceptable dosage form including, but not limited to, capsules, tablets, aqueous suspensions or solutions. In the case of tablets for oral use, carriers that are commonly used include lactose and corn starch. Lubricating agents, such as magnesium stearate, are also typically added. For oral administration in a capsule form, useful diluents include lactose and dried cornstarch. When aqueous suspensions are required for oral use, the active ingredient is combined with emulsifying and suspending agents. If desired, certain sweetening, flavoring or coloring agents may also be added.

[0262] Alternatively, the pharmaceutical compositions of this invention may be administered in the form of suppositories for rectal administration. These may be prepared by mixing the agent with a suitable non irritating excipient which is solid at room

temperature but liquid at rectal temperature and therefore will melt in the rectum to release the drug. Such materials include cocoa butter, beeswax and polyethylene glycols.

[0263] The pharmaceutical compositions of this invention may also be administered topically, especially when the target of treatment includes areas or organs readily accessible by topical application, including diseases of the eye, the skin, or the lower intestinal tract. Suitable topical formulations are readily prepared for each of these areas or organs.

[0264] Topical application for the lower intestinal tract may be effected in a rectal suppository formulation (see above) or in a suitable enema formulation. Topically transdermal patches may also be used.

[0265] For topical applications, the pharmaceutical compositions may be formulated in a suitable ointment containing the active component suspended or dissolved in one or more carriers. Carriers for topical administration of the compounds of this invention include, but are not limited to, mineral oil, liquid petrolatum, white petrolatum, propylene glycol, polyoxyethylene, polyoxypropylene compound, emulsifying wax and water. Alternatively, the pharmaceutical compositions may be formulated in a suitable lotion or cream containing the active components suspended or dissolved in one or more pharmaceutically acceptable carriers. Suitable carriers include, but are not limited to, mineral oil, sorbitan monostearate, polysorbate 60, cetyl esters wax, cetearyl alcohol, 2 octyldodecanol, benzyl alcohol and water.

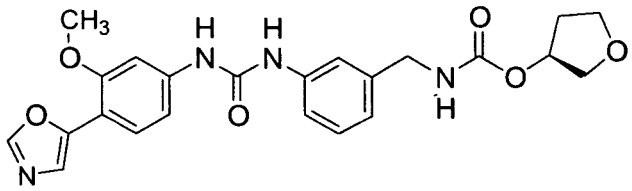
[0266] For ophthalmic use, the pharmaceutical compositions may be formulated as micronized suspensions in isotonic, pH adjusted sterile saline, or, preferably, as solutions in isotonic, pH adjusted sterile saline, either with our without a preservative such as benzylalkonium chloride. Alternatively, for ophthalmic uses, the pharmaceutical compositions may be formulated in an ointment such as petrolatum.

[0267] The pharmaceutical compositions of this invention may also be administered by nasal aerosol or inhalation. Such compositions are prepared according to techniques well known in the art of pharmaceutical formulation and may be prepared as solutions in saline, employing benzyl alcohol or other suitable preservatives, absorption promoters to enhance bioavailability, fluorocarbons, and/or other conventional solubilizing or dispersing agents.

[0268] In one embodiment, the pharmaceutical compositions are formulated for oral administration.

[0269] In another embodiment, the compositions of this invention additionally comprise another anti-viral agent, preferably an anti-HCV agent. Such anti-viral agents include, but are not limited to, immunomodulatory agents, such as α, β–, and γ–interferons, pegylated

derivatized interferon–α compounds, and thymosin; other anti-viral agents, such as ribavirin, amantadine, and telbivudine; other inhibitors of hepatitis C proteases (NS2-NS3 inhibitors and NS3-NS4A inhibitors); inhibitors of other targets in the HCV life cycle, including helicase and polymerase inhibitors; inhibitors of internal ribosome entry; broad-spectrum viral inhibitors, such as IMPDH inhibitors (e.g., compounds of U.S. Pat. Nos. 5,807,876, 6,498,178, 6,344,465, and 6,054,472, WO 97/40028, WO 98/40381, WO 00/56331, and mycophenolic acid and derivatives thereof, and including, but not limited to VX-497, VX-148, and/or VX-944); or combinations of any of the above. See also W. Markland et al., Antimicrobial & Antiviral Chemotherapy, 44, p. 859 (2000) and U.S. Pat. No. 6,541,496.



[0270] The following definitions are used herein (with trademarks referring to products available as of this application's filing date):

"Peg-Intron" means PEG-INTRON®, peginteferon alfa-2b, available from Schering Corporation, Kenilworth, NJ;

"Intron" means INTRON-A®, interferon alfa-2b available from Schering Corporation, Kenilworth, NJ;

"ribavirin" means ribavirin (1-beta-D-ribofuranosyl-1H-1,2,4-triazole-3-carboxamide, available from ICN Pharmaceuticals, Inc., Costa Mesa, CA; described in the Merck Index, entry 8365, Twelfth Edition; also available as REBETROL® from Schering Corporation, Kenilworth, NJ, or as COPEGASUS® from Hoffmann-La Roche, Nutley, NJ;

"Pagasys" means PEGASYS®, peginterferon alfa-2a available Hoffmann-La Roche, Nutley, NJ;

"Roferon" mean ROFERON®, recombinant interferon alfa-2a available from Hoffmann-La Roche, Nutley, NJ;

"Berefor" means BEREFOR®, interferon alfa 2 available from Boehringer Ingelheim Pharmaceutical, Inc., Ridgefield, CT;

SUMIFERON®, a purified blend of natural alpha interferons such as Sumiferon available from Sumitomo, Japan;

WELLFERON®, interferon alpha n1 available from Glaxo_Wellcome LTd., Great Britain; and

ALFERON®, a mixture of natural alpha interferons made by Interferon Sciences, and

available from Purdue Frederick Co., CT.

[0271] The term "interferon" as used herein means a member of a family of highly homologous species-specific proteins that inhibit viral replication and cellular proliferation, and modulate immune response, such as interferon alpha, interferon beta, or interferon gamma. The Merck Index, entry 5015, Twelfth Edition.

[0272] According to one embodiment of the present invention, the interferon is α-interferon. According to another embodiment, a therapeutic combination of the present invention utilizes natural alpha interferon 2a. Or, the therapeutic combination of the present invention utilizes natural alpha interferon 2b. In another embodiment, the therapeutic combination of the present invention utilizes recombinant alpha interferon 2a or 2b. In yet another embodiment, the interferon is pegylated alpha interferon 2a or 2b. Interferons suitable for the present invention include:

(a) INTRON-A® (interferon-alpha 2B, Schering Plough),

(b) PEG-INTRON®,

(c) PEGASYS®,

(d) ROFERON®,

(e) BEREFOR®,

(f) SUMIFERON®,

(g) WELLFERON®,

(h) consensus alpha interferon available from Amgen, Inc., Newbury Park, CA,

(i) ALFERON®;

(j) VIRAFERON®;

(k) INFERGEN®;

(l) ALBUFERON™.

[0273] As is recognized by skilled practitioners, a protease inhibitor would be preferably administered orally. Interferon is not typically administered orally. Nevertheless, nothing herein limits the methods or combinations of this invention to any specific dosage forms or regime. Thus, each component of a combination according to this invention may be administered separately, together, or in any combination thereof.

[0274] In one embodiment, the protease inhibitor and interferon are administered in separate dosage forms. In one embodiment, any additional agent is administered as part of a single dosage form with the protease inhibitor or as a separate dosage form. As this invention involves a combination of compounds, the specific amounts of each compound may be dependent on the specific amounts of each other compound in the combination. As

recognized by skilled practitioners, dosages of interferon are typically measured in IU (e.g., about 4 million IU to about 12 million IU).

[0275] Accordingly, agents (whether acting as an immunomodulatory agent or otherwise) that may be used in combination with a compound of this invention include, but are not limited to, Albuferon™ (albumin-Interferon alpha) available from Human Genome Sciences; PEG-INTRON® (peginterferon alfa-2b, available from Schering Corporation, Kenilworth, NJ); INTRON-A®, (interferon alfa-2b available from Schering Corporation, Kenilworth, NJ); ribavirin (1-beta-D-ribofuranosyl-1H-1,2,4-triazole-3-carboxamide, available from ICN Pharmaceuticals, Inc., Costa Mesa, CA; described in the Merck Index, entry 8365, Twelfth Edition); REBETROL® (Schering Corporation, Kenilworth, NJ), COPEGUS® (Hoffmann-La Roche, Nutley, NJ); PEGASYS® (peginterferon alfa-2a available Hoffmann-La Roche, Nutley, NJ); ROFERON® (recombinant interferon alfa-2a available from Hoffmann-La Roche, Nutley, NJ); BEREFOR® (interferon alfa 2 available from Boehringer Ingelheim Pharmaceutical, Inc., Ridgefield, CT); SUMIFERON® (a purified blend of natural alpha interferons such as Sumiferon available from Sumitomo, Japan); WELLFERON® (interferon alpha n1 available from Glaxo Wellcome Ltd., Great Britain); ALFERON® (a mixture of natural alpha interferons made by Interferon Sciences, and available from Purdue Frederick Co., CT); α-interferon; natural alpha interferon 2a; natural alpha interferon 2b; pegylated alpha interferon 2a or 2b; consensus alpha interferon (Amgen, Inc., Newbury Park, CA); VIRAFERON®; INFERGEN®; REBETRON® (Schering Plough, Interferon-alpha 2B + Ribavirin); pegylated interferon alpha (Reddy, K.R. et al. "Efficacy and Safety of Pegylated (40-kd) Interferon alpha-2a Compared with Interferon alpha-2a in Noncirrhotic Patients with Chronic Hepatitis C (Hepatology, 33, pp. 433-438 (2001); consensus interferon (Kao, J.H., et al., "Efficacy of Consensus Interferon in the Treatment of Chronic Hepatitis" J. Gastroenterol. Hepatol. 15, pp. 1418-1423 (2000); lymphoblastoid or "natural" interferon; interferon tau (Clayette, P. et al., "IFN-tau, A New Interferon Type I with Antiretroviral activity" Pathol. Biol. (Paris) 47, pp. 553-559 (1999); interleukin-2 (Davis, G.L. et al., "Future Options for the Management of Hepatitis C." Seminars in Liver Disease, 19, pp. 103-112 (1999); Interleukin-6 (Davis et al. "Future Options for the Management of Hepatitis C." Seminars in Liver Disease, 19, pp. 103-112 (1999); interleukin-12 (Davis, G.L. et al., "Future Options for the Management of Hepatitis C." Seminars in Liver Disease, 19, pp. 103-112 (1999); and compounds that enhance the development of type 1 helper T cell response (Davis et al., "Future Options for the Management of Hepatitis C." Seminars in Liver Disease, 19, pp. 103-112 (1999)). Also included are compounds that stimulate the synthesis of interferon

in cells (Tazulakhova, E.B. et al., "Russian Experience in Screening, analysis, and Clinical Application of Novel Interferon Inducers" J. Interferon Cytokine Res., 21 pp. 65-73) including, but are not limited to, double stranded RNA, alone or in combination with tobramycin, and Imiquimod (3M Pharmaceuticals; Sauder, D.N. "Immunomodulatory and Pharmacologic Properties of Imiquimod" J. Am. Acad. Dermatol., 43 pp. S6-11 (2000).

[0276] Compounds that stimulate the synthesis of interferon in cells (Tazulakhova, E.B. et al., "Russian Experience in Screening, analysis, and Clinical Application of Novel Interferon Inducers" J. Interferon Cytokine Res., 21 pp. 65-73) include, but are not limited to, double stranded RNA, alone or in combination with tobramycin, and Imiquimod (3M Pharmaceuticals; Sauder, D.N. "Immunomodulatory and Pharmacologic Properties of Imiquimod" J. Am. Acad. Dermatol., 43 pp. S6-11 (2000).

[0277] Other non-immunomodulatory or immunomodulatory compounds may be used in combination with a compound of this invention including, but not limited to, those specified in WO 02/18369, which is incorporated herein by reference (see, e.g., page 273, lines 9-22 and page 274, line 4 to page 276, line 11).

[0278] Still other agents include those described in various published U.S. Patent Applications. These publications provide additional teachings of compounds and methods that could be used in combination with VX-950 in the methods of this invention, particularly for the treatment of hepatitis. It is contemplated that any such methods and compositions may be used in combination with the methods and compositions of the present invention. For brevity, the disclosure the disclosures from those publications is referred to be reference to the publication number but it should be noted that the disclosure of the compounds in particular is specifically incorporated herein by reference. Exemplary such publications include U.S. Patent Publication No. 20040058982; U.S. Patent Publication No. 20050192212; U.S. Patent Publication No. 20050080005; U.S. Patent Publication No. 20050062522; U.S. Patent Publication No. 20050020503; U.S. Patent Publication No. 20040229818; U.S. Patent Publication No. 20040229817; U.S. Patent Publication No. 20040224900; U.S. Patent Publication No. 20040186125; U.S. Patent Publication No. 20040171626; U.S. Patent Publication No. 20040110747; U.S. Patent Publication No. 20040072788; U.S. Patent Publication No. 20040067901; U.S. Patent Publication No. 20030191067; U.S. Patent Publication No. 20030187018; U.S. Patent Publication No. 20030186895; U.S. Patent Publication No. 20030181363; U.S. Patent Publication No. 20020147160; U.S. Patent Publication No. 20040082574; U.S. Patent Publication No. 20050192212; U.S. Patent Publication No. 20050187192; U.S. Patent Publication No. 20050187165; U.S. Patent

Publication No. 20050049220; and U.S. Patent Publication No. US2005/0222236.

[0279] This invention may also involve administering a cytochrome P450 monooxygenase inhibitor. CYP inhibitors may be useful in increasing liver concentrations and/or increasing blood levels of compounds that are inhibited by CYP.

[0280] If an embodiment of this invention involves a CYP inhibitor, any CYP inhibitor that improves the pharmacokinetics of the relevant NS3/4A protease may be used in a method of this invention. These CYP inhibitors include, but are not limited to, ritonavir (WO 94/14436), ketoconazole, troleandomycin, 4-methyl pyrazole, cyclosporin, clomethiazole, cimetidine, itraconazole, fluconazole, miconazole, fluvoxamine, fluoxetine, nefazodone, sertraline, indinavir, nelfinavir, amprenavir, fosamprenavir, saquinavir, lopinavir, delavirdine, erythromycin, VX-944, and VX-497. Preferred CYP inhibitors include ritonavir, ketoconazole, troleandomycin, 4-methyl pyrazole, cyclosporin, and clomethiazole. For preferred dosage forms of ritonavir, see U.S. Pat. No. 6,037, 157, and the documents cited therein: U.S. Pat. No. 5,484,801, U.S. Application Serial No. 08/402,690, WO 95/07696 and WO 95/09614.

[0281] Methods for measuring the ability of a compound to inhibit cytochrome P450 monooxygenase activity are known. See, e.g., U.S. Pat. No. 6,037,157, and Yun, et al. Drug Metabolism & Disposition, vol. 21, pp. 403-407 (1993).

[0282] Upon improvement of a patient's condition, a maintenance dose of a compound, composition or combination of this invention may be administered, if necessary. Subsequently, the dosage or frequency of administration, or both, may be reduced, as a function of the symptoms, to a level at which the improved condition is retained when the symptoms have been alleviated to the desired level, treatment should cease. Patients may, however, require intermittent treatment on a long-term basis upon any recurrence of disease symptoms.

[0283] It should also be understood that a specific dosage and treatment regimen for any particular patient will depend upon a variety of factors, including the activity of the specific compound employed, the age, body weight, general health, sex, diet, time of administration, rate of excretion, drug combination, and the judgment of the treating physician and the severity of the particular disease being treated. The amount of active ingredients will also depend upon the particular described compound and the presence or absence and the nature of the additional anti-viral agent in the composition.

[0284] According to another embodiment, the invention provides a method for treating a patient infected with a virus characterized by a virally encoded serine protease that is

necessary for the life cycle of the virus by administering to said patient a pharmaceutically acceptable composition of this invention. In one embodiment, the methods of this invention are used to treat a patient suffering from a HCV infection. Such treatment may completely eradicate the viral infection or reduce the severity thereof. In another embodiment, the patient is a human being.

[0285] In an alternate embodiment, the methods of this invention additionally comprise the step of administering to said patient an anti-viral agent preferably an anti-HCV agent. Such anti-viral agents include, but are not limited to, immunomodulatory agents, such as α–, β–, and γ–interferons, pegylated derivatized interferon–α compounds, and thymosin; other anti-viral agents, such as ribavirin, amantadine, and telbivudine; other inhibitors of hepatitis C proteases (NS2-NS3 inhibitors and NS3-NS4A inhibitors); inhibitors of other targets in the HCV life cycle, including but not limited to helicase and polymerase inhibitors; inhibitors of internal ribosome entry; broad-spectrum viral inhibitors, such as IMPDH inhibitors (e.g., VX-497 and other IMPDH inhibitors disclosed in U.S. Pat. Nos. 5,807,876 and 6,498,178, mycophenolic acid and derivatives thereof); inhibitors of cytochrome P-450, such as ritonavir, or combinations of any of the above.

[0286] Such additional agent may be administered to said patient as part of a single dosage form comprising both a compound of this invention and an additional anti-viral agent. Alternatively the additional agent may be administered separately from the compound of this invention, as part of a multiple dosage form, wherein said additional agent is administered prior to, together with or following a composition comprising a compound of this invention.

[0287] Pharmaceutical compositions may also be prescribed to the patient in "patient packs" containing the whole course of treatment in a single package, usually a blister pack. Patient packs have an advantage over traditional prescriptions, where a pharmacist divides a patients supply of a pharmaceutical from a bulk supply, in that the patient always has access to the package insert contained in the patient pack, normally missing in traditional prescriptions. The inclusion of a package insert has been shown to improve patient compliance with the physician's instructions.

[0288] It will be understood that the administration of the combination of the invention by means of a single patient pack, or patient packs of each formulation, containing within a package insert instructing the patient to the correct use of the invention is a desirable additional feature of this invention.

[0289] According to a further aspect of the invention is a pack comprising at least one compound of formula I (in dosages according to this invention) and an information insert

containing directions on the use of the combination of the invention. Any composition, dosage form, therapeutic regimen or other embodiment of this invention may be presented in a pharmaceutical pack. In an alternative embodiment of this invention, the pharmaceutical pack further comprises one or more of additional agent as described herein. The additional agent or agents may be provided in the same pack or in separate packs.

[0290] Another aspect of this involves a packaged kit for a patient to use in the treatment of HCV infection or in the prevention of HCV infection (or for use in another method of this invention), comprising: a single or a plurality of pharmaceutical formulation of each pharmaceutical component; a container housing the pharmaceutical formulation(s) during storage and prior to administration; and instructions for carrying out drug administration in a manner effective to treat or prevent HCV infection.

[0291] Accordingly, this invention provides kits for the simultaneous or sequential administration of a dose of at least one compound of formula I (and optionally an additional agent). Typically, such a kit will comprise, e.g. a composition of each compound and optional additional agent(s) in a pharmaceutically acceptable carrier (and in one or in a plurality of pharmaceutical formulations) and written instructions for the simultaneous or sequential administration.

[0292] In another embodiment, a packaged kit is provided that contains one or more dosage forms for self administration; a container means, preferably sealed, for housing the dosage forms during storage and prior to use; and instructions for a patient to carry out drug administration. The instructions will typically be written instructions on a package insert, a label, and/or on other components of the kit, and the dosage form or forms are as described herein. Each dosage form may be individually housed, as in a sheet of a metal foil-plastic laminate with each dosage form isolated from the others in individual cells or bubbles, or the dosage forms may be housed in a single container, as in a plastic bottle. The present kits will also typically include means for packaging the individual kit components, i.e., the dosage forms, the container means, and the written instructions for use. Such packaging means may take the form of a cardboard or paper box, a plastic or foil pouch, etc.

[0293] A kit according to this invention could embody any aspect of this invention such as any composition, dosage form, therapeutic regimen, or pharmaceutical pack. The packs and kits according to this invention optionally comprise a plurality of compositions or dosage forms. Accordingly, included within this invention would be packs and kits containing one composition or more than one composition.

[0294] In yet another embodiment the present invention provides a method of pre-treating a

biological substance intended for administration to a patient comprising the step of contacting said biological substance with a pharmaceutically acceptable composition comprising a compound of this invention. Such biological substances include, but are not limited to, blood and components thereof such as plasma, platelets, subpopulations of blood cells and the like; organs such as kidney, liver, heart, lung, etc; sperm and ova; bone marrow and components thereof, and other fluids to be infused into a patient such as saline, dextrose, etc.

[0295] According to another embodiment the invention provides methods of treating materials that may potentially come into contact with a virus characterized by a virally encoded serine protease necessary for its life cycle. This method comprises the step of contacting said material with a compound according to the invention. Such materials include, but are not limited to, surgical instruments and garments (e.g. clothes, gloves, aprons, gowns, masks, eyeglasses, footwear, etc.); laboratory instruments and garments (e.g. clothes, gloves, aprons, gowns, masks, eyeglasses, footwear, etc.); blood collection apparatuses and materials; and invasive devices, such as, for example, shunts and stents.

[0296] In another embodiment, the compounds of this invention may be used as laboratory tools to aid in the isolation of a virally encoded serine protease. This method comprises the steps of providing a compound of this invention attached to a solid support; contacting said solid support with a sample containing a viral serine protease under conditions that cause said protease to bind to said solid support; and eluting said serine protease from said solid support. In one embodiment, the viral serine protease isolated by this method is HCV NS3-NS4A protease.

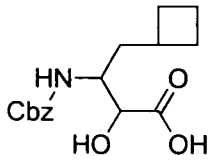
[0297] All references cited within this document are incorporated herein by reference.

IV. METHODS AND EXAMPLES

[0298] In order that the invention described herein may be more fully understood, the following methods and examples are provided. It should be understood that these methods and examples are for illustrative purposes only and are not to be construed as limiting this invention in any manner.

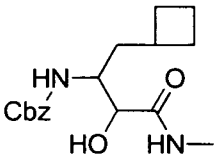
A. Preparation of Intermediates for Compounds of Formula I

[0299] Set forth below are various methods for preparing intermediates that can be used to synthesize the compound of Formula I.



Preparation of 3-(benzyloxycarbonylamino)-4-cyclobutyl-2-hydroxybutanoic acid.

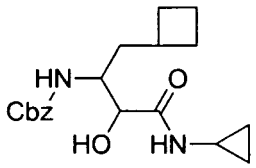
[0300] A solution of the cyanohydrin prepared according to methods described in WO 04/113294 (1 g, 3.65 mmol) in conc. HCl (12 mL) was heated to reflux for 18 hours. The reaction was concentrated in vacuo to afford the desired amino acid as an HCl salt (1.7 g) which was used in the next step without further purification. A solution of the above HCl salt in THF was treated with DIPEA (2.68 g) and Z-OSu (5.16 g). The reaction mixture was stirred at room temperature for 8 hours. The reaction mixture was diluted with toluene and HCl (12 N, until pH = 1). After separation, the organic layer was extracted with sat. $NaHCO_3$ (50 mL, twice). The aqueous layer was made acidic with HCl (6 N) until pH = 1 and extracted with EtOAc (200 mL). The combined organic layer was dried and concentrated in vacuo to afford the title compound (0.6 g). (M+1) 308.



Preparation of benzyl 1-cyclobutyl-3-hydroxy-4-(methylamino)-4-oxobutan-2-ylcarbamate.

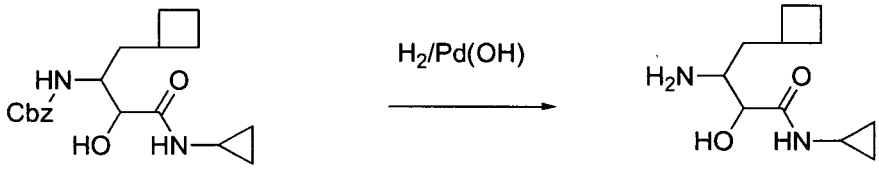
[0301] To a solution of 3-(benzyloxycarbonylamino)-4-cyclobutyl-2-hydroxybutanoic acid (250 mg, 0.81 mmol) in DCM (20 mL) was added HOSu (140 mg, 1.22 mmol), EDC (234 mg, 1.22 mmol). After stirring for 1 hour, methylamine in THF (2 N, 0.81 mL) was added to the above mixture. The reaction mixture was stirred for 18 hours and then concentrated in vacuo. The residue was purified by Gilson Prep to afford the title compound

(135 mg). ^{1}H-NMR ($CDCl_3$): δ 7.54-7.28 (m, 5H), 6.67 (NH, 1H), 5.03 (dd, 2H), 3.68 (m, 1H), 2.73 (m, 3H), 2.26 (m, 1H), 1.97-1.31 (m, 9H). (M+1) 321.



Preparation of benzyl 1-cyclobutyl-4-(cyclopropylamino)-3-hydroxy-4-oxobutan-2-ylcarbamate.

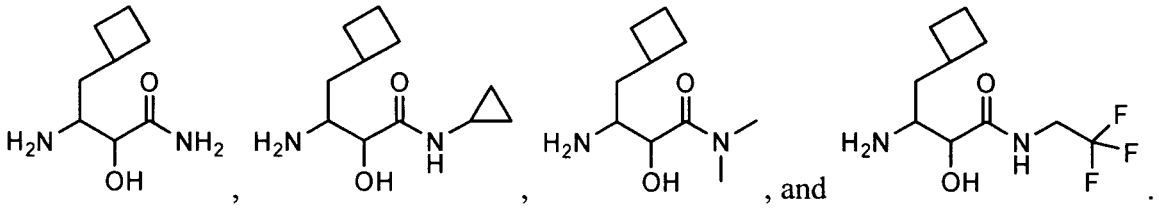
[0302] To a solution of 3-(benzyloxycarbonylamino)-4-cyclobutyl-2-hydroxybutanoic acid (600 mg, 1.95 mmol) in DCM (20 mL) was added HOSu (337 mg, 2.93 mmol), EDC (562 mg, 2.93 mmol). After stirring for 1 hour, cyclopropylamine (223 mg, 3.9 mmol) was added to the above mixture. The product was extracted with EtOAc. The combined organic layer was then washed with HCl (1N), water, $NaHCO_3$, and brine and then concentrated in vacuo to afford benzyl 1-cyclobutyl-4-(cyclopropylamino)-3-hydroxy-4-oxobutan-2-ylcarbamate (530 mg). (M+1) 347.



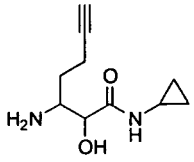
Preparation of 3-amino-4-cyclobutyl-N-cyclopropyl-2-hydroxybutanamide.

[0303] To a solution of the CBz amide (530 mg, 1.53 mmol) in MeOH (30 mL) was added $Pd(OH)_2/C$ (106 mg). The mixture was stirred under H_2 (1 atm) for 18 hours. After filtration, the filtrate was concentrated in vacuo to afford the title compound (300 mg). ^{1}H-NMR ($CDCl_3$): δ 3.29 (m, 1H), 2.74 (m, 1H), 2.37-1.66 (m, 9H), 1.40 (m, 1H), 0.78 (m, 2H), 0.51 (m, 2H). (M+1) 213.

[0304] The following compounds were prepared in a similar fashion to preparing 3-amino-4-cyclobutyl-N-cyclopropyl-2-hydroxybutanamide by using the appropriate amine:

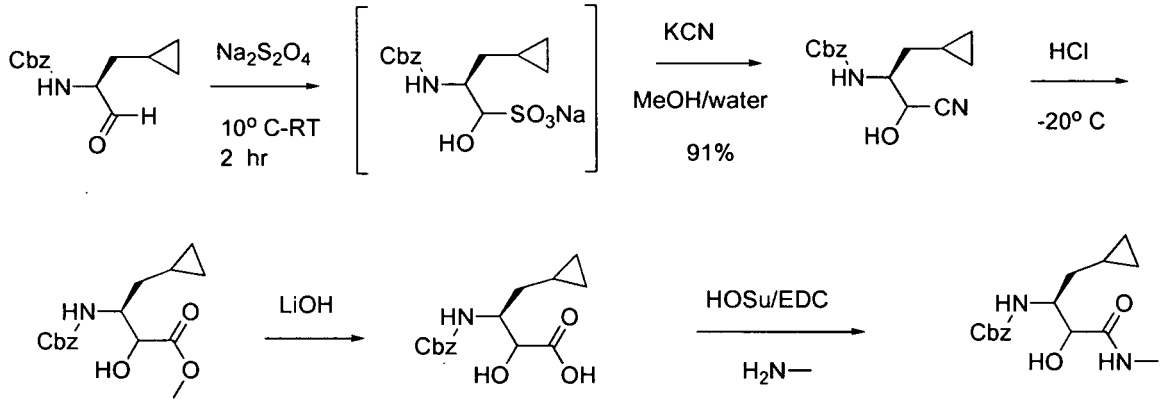


Preparation of 3-amino-N-cyclopropyl-2-hydroxyhept-6-ynamide

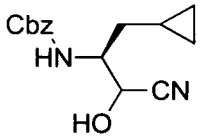


[0305] 3-Amino-N-cyclopropyl-2-hydroxyhept-6-ynamide was prepared as described by N. Kobayashi, et al. in US 2003/153788, which is incorporated herein by reference in its entirety. ^{1}H-NMR (500 MHz, DMSO-d_6): 8.18 (s), 6.34 (s), 4.22 (s), 3.45 (s), 3.17 (s), 2.84 (s), 2.69 (d, J = 3.2 Hz), 2.30 (m), 2.24 (m), 1.70 (m), 1.59 (m), 0.62 (d, J = 5.0 Hz), 0.53 (s) ppm; FIA m/z 197.01 ES^+.

Preparation of Cbz-protected (3S)-3-amino-4-cyclopropyl-2-hydroxy-N-methylbutanamide

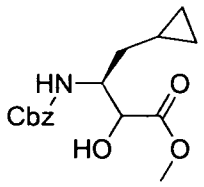


Step 1: Preparation of benzyl (2S)-1-cyano-3-cyclopropyl-1-hydroxypropan-2-ylcarbamate.



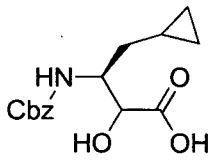
[0306] To a solution of the aldehyde (7.9 g, 32 mmol) in MeOH (50 mL) at 10 °C was added $Na_2S_2O_4$ (6.13 g, 35.2 mmol) and the resulting mixture was warmed to room temperature and stirred for 2 hours then cooled to 10 °C. To this reaction mixture, a solution of KCN in water (50 mL) was added. After stirring at room temperature for 18 hours, the mixture was extracted with TBME (100 mL, twice). The combined organic layers were washed with water and brine, dried and concentrated in vacuo to afford the title compound (8 g). (M+1) 275.

Step 2: Preparation of (3S)-methyl 3-(benzyloxycarbonylamino)-4-cyclopropyl-2-hydroxybutanoate.



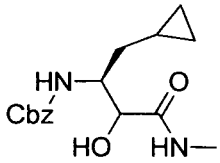
[0307] To a solution of the cyanohydrin (1 g, 3.65 mmol) in MeOH (15 mL) at -20°C was bubbled through a stream of dry HCl gas for 30 minutes. The resulting mixture was stirred at room temperature for 2 hours. The reaction mixture was purged with nitrogen gas for 30 minutes and then concentrated. The residue at 0°C was quenched with ice water and then stirred at room temperature for 1 hour. The product was extracted with EtOAc. The combined organic layer was washed with $NaHCO_3$, water, brine and concentrated in vacuo to afford the title compound (0.5 g). ^{1}H-NMR ($CDCl_3$) δ: 7.31-7.30 (m, 5H), 5.09 (d, 2H), 4.44-4.14 (m, 2H), 3.78 (d, 3H), 1.58-1.42 (m, 2H), 0.70 (m, 1H), 0.47 (t, 2H), 0.11-0.01 (m, 2H). (M+1) 308.

Step 3: Preparation of (3S)-3-(benzyloxycarbonylamino)-4-cyclopropyl-2-hydroxybutanoic acid



[0308] To a solution of the methyl ester of Step 2 (400 mg; 1.3 mmol) in THF (8 mL) and water (6.63 mL) was added LiOH (1 N; 1.37 mL). The reaction mixture was stirred for 30 minutes and then acidified with 1.0 N HCl to pH = 3≈4. The mixture was extracted with EtOAc (20 mL, twice). The combined organic layer was washed with water, brine, and then concentrated in vacuo to afford the title compound (370 mg). (M+1) 294.

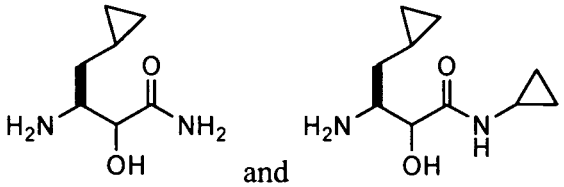
Step 4: Preparation of benzyl (2S)-1-cyclopropyl-3-hydroxy-4-(methylamino)-4-oxobutan-2-ylcarbamate.



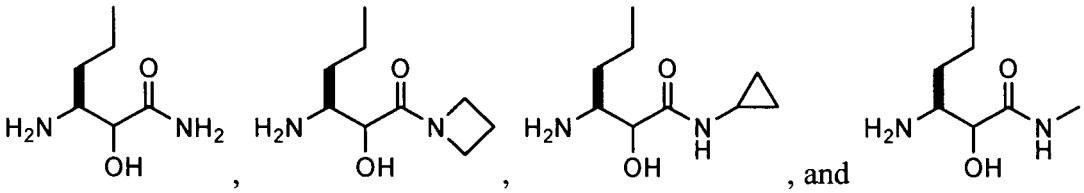
[0309] To a solution of (3S)-3-(benzyloxycarbonylamino)-4-cyclopropyl-2-hydroxybutanoic acid (180 mg, 0.26 mmol) in DCM (20 mL) was added HOSu (105 mg, 0.92 mmol), EDC (175 mg, 0.92 mmol). After stirred for 30 minutes, methylamine in THF

(2 N, 0.92 mL) was added to above mixture. The reaction mixture was stirred for 18 hours and then concentrated in vacuo. The residue was purified by Gilson Prep to afford title compound (50 mg). ^{1}H-NMR ($CDCl_3$): δ 7.53-7.26 (m, 5H), 6.83 (NH, 1H), 5.25 (NH, 1H), 5.05 (m, 2H), 4.25-3.89 (m, 3H), 2.70 (m, 3H), 1.4 (m, 1H), 0.86 (m, 1H), 0.61 (m, 1H), 0.38 (m, 2H), 0.33 (m, 2H). (M+1) 307.

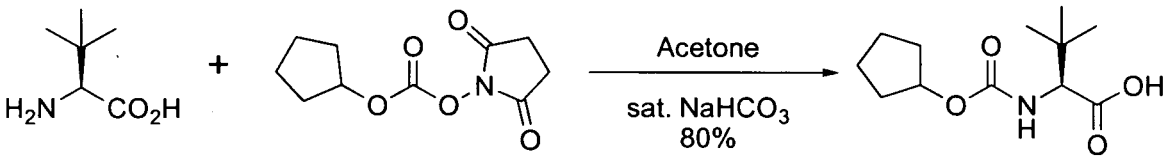
[0310] The following compounds can be prepared in the similar manner by using appropriate amines, followed by hydrogenation.


and

[0311] The following compounds can be prepared in the methods described by Perni, R. et al. in WO 01/74768, which is incorporated herein by reference in its entirety.

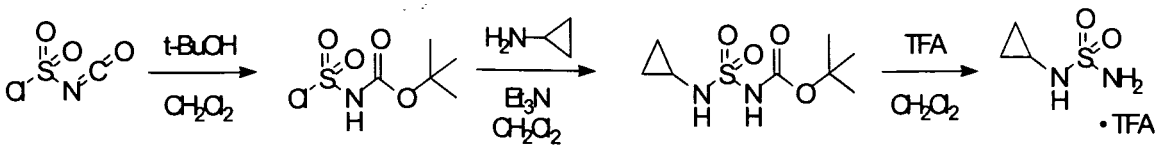

, and

Preparation of (S)-2-(cyclopentyloxycarbonylamino)-3,3-dimethylbutanoic acid



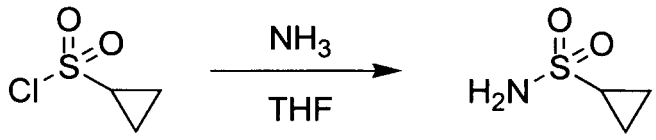
[0312] In a 5L RB flask dissolved t-butyl glycine (74 g, 0.56 mol, 1.02 eq.) in saturated sodium bicarbonate (11 vol). Cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate (126 g, 0.55 mol, 1 eq.) was dissolved in acetone (5.5 vol) and the solution slowly added via addition funnel at room temperature to the solution of the glycine. The reaction mixture was stirred at room temperature until complete (approximately 4 hours). The acetone was removed under reduced pressure and the remaining aqueous solution was extracted with 30% ethyl acetate in hexanes (thrice, 5.5 vol each). The organic layers were discarded. The pH of the aqueous layer was adjusted to 2 with 2 N HCl and then extracted with ethyl acetate (thrice, 5.5 vol). The combined organic layers were dried (Na_2SO_4), filtered, and the solvent removed under reduced pressure to provide a clear oil the slowly crystallized. The crude product was crystallized from hexanes/ethyl acetate to provide (S)-2-(cyclopentyloxycarbonylamino)-3,3-dimethylbutanoic acid as a white solid (82 g). The mother liquid was stripped and a second crop of crystals obtained (combined yield 105.54 g).

Preparation of Sulfonyl Compounds

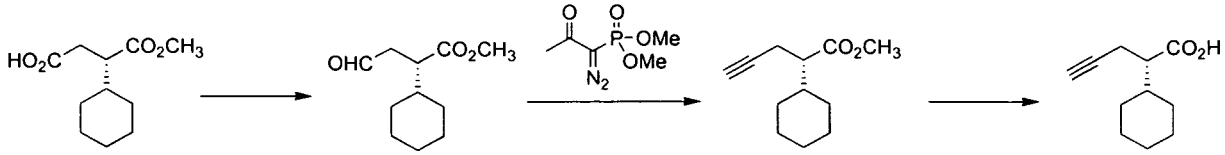


S1 **S2** **S3** **S4**

[0313] Compounds **S1**, **S2**, **S3**, and **S4**, shown above, were prepared according to procedures described in WO 2005/095403 and PCT/US2005/010494, hereby incorporated by references by their entireties. Specifically, to a solution of chlorosulfonylisocyanate (10 mL, 115 mmol) in CH_2Cl_2 (200 mL) at 0 °C was added t-BuOH (11 mL, 1 eq.). The mixture was stirred for 60 minutes, then added via cannula into a solution of cyclopropylamine (6.6 g) in CH_2Cl_2 (200 mL) with triethylamine (30 mL) at 0 °C concurrently with a solution of triethylamine (50 mL) in CH_2Cl_2 (100 mL) via addition funnel. Internal temperature was maintained below 8 °C. Stirred at room temperature after completion of addition for 4 hours. The reaction was then diluted with CH_2Cl_2 and transferred to a separatory funnel, washed with 1 N HCl (twice, 400 mL each), brine (300 mL), dried ($MgSO_4$), filtered and concentrated. The product was recrystallized from ethyl acetate/hexanes to yield 16.8 g (71.3 mmol) of **S3**. Compound **S3** was deprotected with trifluoroacetic acid in CH_2Cl_2 to give compound **S4**.



[0314] Ammonia gas was bubbled through a gas dispersion tube into THF (40 mL) cooled to 0 °C for 5 minutes. To this solution at 0 °C was added cyclopropylsulfonylchloride (1 gram, 7.1 mmol). The reaction was stirred at room temperature overnight, then filtered through a plug of silica gel, followed by elution with EtOAc to yield 750 mg (6.19 mmol) of cyclopropylsulfonamide. ^{1}H-NMR (500 MHz, Methanol-d_4): 4.79 (s, 2H), 2.59-2.54 (m, 1H), 1.06-0.96 (m, 4H).

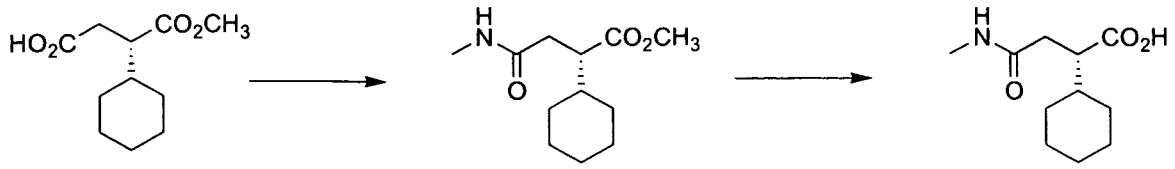


XX5 **XX6** **XX7** **XX8**

[0315] To a solution of compound **XX5** (1.37 g, 6.41 mmol) in THF (30 mL) at 0 °C was added dropwise borane-dimethylsulfide (3.85 mL, 7.8 mmol, 2.0 M in toluene). The reaction mixture was stirred for 1 h with gradual warming to room temperature, quenched with H_2O (20 mL), and extracted with ethyl acetate (thrice, 30 mL each). The combined organics were dried and concentrated under reduced pressure to provide 1.3 g of a colorless oil which was used without further purification. To oxalyl chloride (2.24 mL, 25.6 mmol) in CH_2Cl_2 (15 mL, anhydrous) at -78 °C under inert atmosphere was added dropwise a solution of DMSO (2.73 mL, 38.5 mmol) in CH_2Cl_2 (8 mL). After stirring for 10 min, a solution of the alcohol (1.3 g, 6.41 mmol) in CH_2Cl_2 (6 mL) was added dropwise. After an additional 10 min, triethylamine (7.15 mL, 51.3 mmol) in CH_2Cl_2 was added and the reaction was stirred another 30 min with gradual warming to 0 °C. The reaction mixture was washed with 1 M HCl (20 mL) followed by brine (20 mL). The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure. The resulting oil was purified via silica gel chromatography to afford 748 mg (over 2 steps) of aldehyde **XX6**. ^{1}H-NMR (500 MHz, CDCl3): 9.75 (s, 1H), 3.67 (s, 3H), 2.91-2.85 (m, 1H), 2.78-2.74 (m, 1H), 2.56-2.52 (m, 1H), 1.74-1.71 (m, 2H), 1.66-1.58 (m, 4H), 1.27-0.95 (m, 5H).

[0316] To a solution of compound **XX6** (581 mg, 2.9 mmol) and K_2CO_3 (811 mg, 5.9 mmol) in MeOH (15 mL) was added dimethyl 1-diazo-2-oxopropylphosphonate (676 mg, 3.5 mmol, Synlett 1996, p. 521). The reaction was stirred 1 h at room temperature, diluted wth Et_2O (20 mL), and washed with saturated $NaHCO_3$ solution (10 mL, aqueous). The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure to give 600 mg of alkyne **XX7** which was used without further purification. ^{1}H-NMR (500 MHz, $CDCl_3$): 3.69 (s, 3H), 2.48-2.37 (m), 1.95 (s, H), 1.73-1.60 (m), 1.30-0.94 (m).

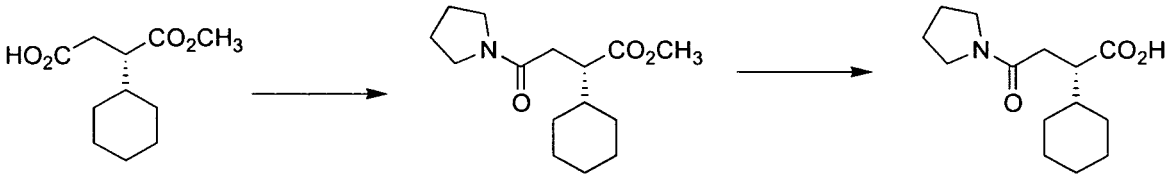
[0317] To a solution of compound **XX7** (600 mg, 2.9 mmol) in a solution of THF / H_2O / MeOH (25 mL, 2:1:2) was added LiOH monohydrate (850 mg, 20.3 mmol). The reaction mixture was stirred 2 h at room temperature, acidified using 1 N HCl (25 mL), and extracted with EtOAc (thrice, 15 mL each). The combined organics were dried over $MgSO_4$ and concentrated to yield 533 mg of carboxylic acid **XX8**, which was used without further purification.



XX5 XX9 XX10

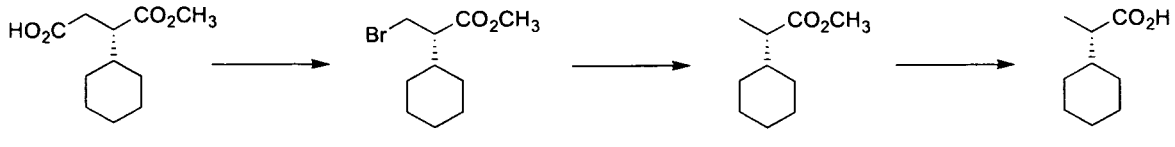
[0318] To a solution of compound **XX5** (100 mg, 0.5 mmol) in CH_2Cl_2 (2.5 mL) was added EDC (107 mg, 0.6 mmol), HOBt (76 mg, 0.6 mmol) and triethylamine (195 μL, 1.4 mmol). To the activated acid solution was added methylamine hydrochloride (38 mg, 0.6 mmol) and the reaction was stirred at room temperature for 12 h. The reaction mixture was washed with H_2O (2 mL), 1 N HCl (2 mL) and saturated $NaHCO_3$ solution (2 mL). The organic layer was dried over $MgSO_4$ and concentrated to give 100 mg of amide **XX9**, which was used without further purification. ^{1}H-NMR (500 MHz, $CDCl_3$): 3.61 (s, 3H), 2.75 - 2.70 (m, 4H), 2.48 - 2.42 (m, 1H), 2.28 - 2.24 (m, 1H), 1.66 -1.48 (m, 6H), 1.35 - 0.90 (m, 5H).

[0319] To a solution of compound **XX9** (100 mg, 0.5 mmol) in a solution of THF/H_2O/MeOH (3 mL, 2:1:2) was added LiOH monohydrate (124 mg, 3 mmol). The reaction mixture was stirred 2 h at room temperature, acidified using 1 N HCl (4 mL), and extracted with EtOAc (3 x 5 mL). The combined organics were dried over $MgSO_4$ and concentrated to yield 87 mg of carboxylic acid **XX10**, which was used without further purification. ^{1}H-NMR (500 MHz, $CDCl_3$) 11.32 (s, H), 2.75-2.64 (m, H), 2.52-2.46 (m, H), 2.37-2.33 (m, H), 2.25 (td, J = 8.7, 2.9 Hz, H), 1.97 (s, H), 1.79 (s, H), 1.74-1.62 (m, H), 1.59-1.49 (m, H), 1.23-1.12 (m, H), 1.08-0.81 (m, H).



XX5 XX11 XX12

[0320] Intermediate **XX12** was prepared according to the procedure for preparing intermediate **XX10** described above, except for using pyrrolidine as a reagent instead of methylamine hydrochloride. ^{1}H-NMR (500 MHz, CDCl3) 11.47 (s, 1H), 3.45 - 3.32 (m, 4H), 2.76 - 2.72 (m, 1H), 2.64 - 2.59 (m, 1H), 2.37 - 2.33 (m, 1H), 1.92 - 1.76 (m, 4H), 1.71 - 1.57 (m), 1.22 - 0.84 (m).

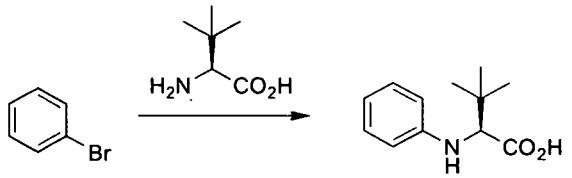


XX5 XX13 XX14 XX15

[0321] To a solution of compound **XX5** (1 g, 4.7 mmol) and HgO yellow (1.01 g, 4.7 mmol) in CCl_4 (23 mL) at reflux was added dropwise over 30 min a solution of bromine (264 μL, 5.1 mmol) in CCl_4 (5 mL). The reaction was stirred at reflux for 1 h, cooled to room temperature, diluted with CH_2Cl_2 (20 mL), washed with 1 N HCl (10 mL), H_2O (10 mL), and brine (10 mL). The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure to yield 1.3 g of compound **XX13** as a colorless oil that was used without further purification. ^{1}H-NMR (500 MHz, $CDCl_3$): 3.67 (s, 3H), 3.52 - 3.44 (m, 2H), 2.63 - 2.58 (m, 1H), 1.70 - 1.64 (m, 3H), 1.60 - 1.54 (m, 3H), 1.24 - 0.92 (m, 5H).

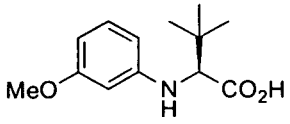
[0322] To a solution of compound **XX13** (578 mg, 2.3 mmol) in DMSO (12 mL) was added sodium borohydride (177 mg, 4.7 mmol). The reaction mixture was stirred at 90 °C for 1 h, diluted with H_2O (10 mL), and extracted with hexanes (3 x 15 mL). The combined organics were dried over $MgSO_4$ and concentrated under reduced pressure. Purification via silica gel chromatography, eluting with EtOAc / petroleum ether, afforded 204 mg of compound **XX14**. ^{1}H-NMR (500 MHz, $CDCl_3$): 3.59 (s, 3H), 2.18 (m, 1H), 1.69 - 1.43 (m, 6H), 1.21 - 0.83 (m, 8H).

[0323] Intermediate **XX15** was prepared according to the procedure for preparing intermediate **XX10**, step b, except for using substrate **XX14** instead of **XX9**.



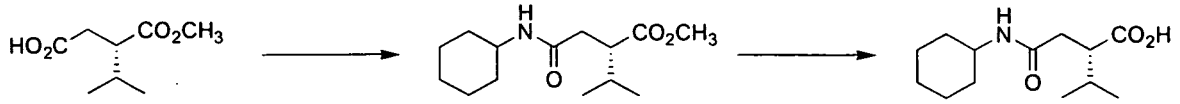
XX16

[0324] To a solution of (S)-2-amino-3,3-dimethylbutanoic acid (787 mg, 6.0 mmol), bromobenzene (632 μL, 6.0 mmol), K_2CO_3 (1.24 g, 9.0 mmol) and CuI (114 mg, 0.6 mmol) was added N,N-dimethylacetamide (7.5 mL). The contents were stirred for 16 h at 90 °C in a sealed pressure vessel. The reaction mixture was diluted with H_2O (15 mL), cooled to 0 °C, and acidified to pH ≈ 5 using 1 N HCl. The mixture was extracted with EtOAc (3 x 20 mL), and the combined organics were washed with brine (1 x 15 mL), dried over $MgSO_4$, and concentrated under reduced pressure. The resulting residue was purified via silica gel chromatography to provide 150 mg of compound **XX16**. ^{1}H-NMR (500 MHz, CDCl3): 7.11-7.09 (m, 2H), 6.69 (t, J = 7.3 Hz, 1H), 6.60- 6.59 (m, 2H), 3.69 (s, 1H), 1.02 (s, 9H).



XX17

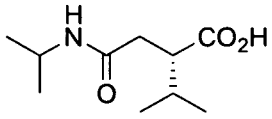
[0325] Intermediate **XX17** was prepared according to the procedure for preparing **XX16**, except for using 1-bromo-3-methoxybenzene as a reagent instead of bromobenzene. ^{1}H-NMR (500 MHz, $CDCl_3$): 6.98 (t, J = 8.1 Hz, 1H), 6.24-6.18 (m, 2H), 6.14 (s, 1H), 3.69 (s, 1H), 3.66 (s, 3H), 1.00 (s, 9H).



XX18 **XX19**

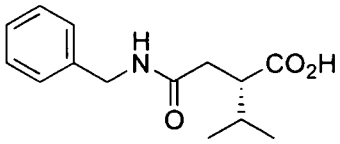
[0326] To a solution of (S)-3-(methoxycarbonyl)-4-methylpentanoic acid (200 mg, 1.2 mmol) in CH_2Cl_2 (6 mL) was added EDC (264 mg, 1.4 mmol), HOBt (186 mg, 1.4 mmol) and triethylamine (481 μL, 3.5 mmol). To the activated acid solution was added cyclohexylamine (158 μL, 1.4 mmol) and the reaction was stirred 4 hours. The reaction mixture was washed with H_2O (3 mL), 1 N HCl (3 mL), and saturated $NaHCO_3$ solution (3 mL). The organic layer was dried over $MgSO_4$, and concentrated under reduced pressure to afford 290 mg of compound **XX18** which was used without further purification. ^{1}H-NMR (500 MHz, $CDCl_3$): 5.78 (d, J=7.5 Hz, 1H), 3.69-3.61 (m, 4H), 2.73-2.69 (m, 1H), 2.45-2.40 (m, 1H), 2.24-2.20 (m, 1H), 1.85 (m, 1H), 1.82-1.76 (m, 2H), 1.63-1.60 (m, 2H), 1.54-1.50 (m, 1H), 1.31-1.22 (m, 2H), 1.12-1.00 (m, 3H), 0.90-0.85 (m, 6H).

[0327] Intermediate **XX19** was prepared according to the procedure for preparing compound **XX10** described above, except for using substrate **XX18** as a reagent instead of compound **XX9**. ES (+) MS: m/e 256 $(M + H)^+$.



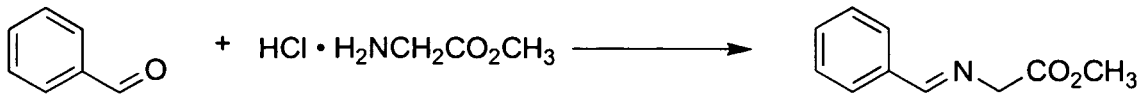
XX20

[0328] Intermediate **XX20** was prepared according to the procedure for preparing compound **XX18** or **XX19** described above, except for using isopropylamine as a reagent instead of cyclohexylamine. ES (+) MS: m/e 216 $(M + H)^+$.

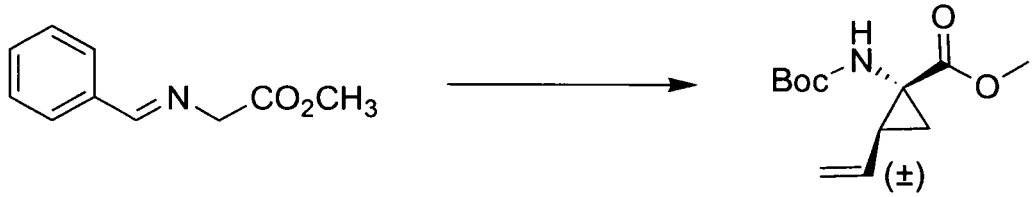


XX21

[0329] Intermediate **XX21** was prepared according to the procedure for preparing **XX18** or **XX19** described above, except for using benzylamine as a reagent instead of cyclohexylamine. ES (+) MS: m/e 264 $(M + H)^+$.

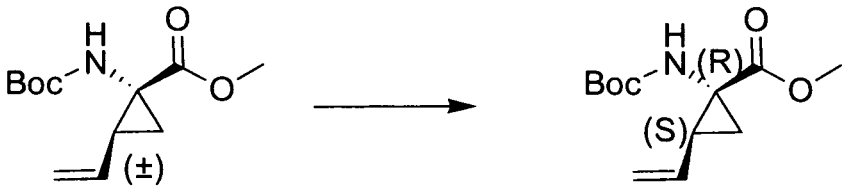


[0330] Glycine methyl ester hydrochloride (50.0 g) was suspended in MTBE (300 mL) at RT. To this was added benzaldehyde (40.5 mL) and anhydrous Na2SO4 (33.9 g). The suspension was cooled in an ice-water bath for 20 minutes, then triethylamine (80 mL) was added dropwise over 15 minutes. After 5 minutes, the reaction was removed from the ice-water bath, and stirred at RT for 24 hours. The reaction was quenched with 200 mL ice-water mixture and the organic layer was separated. The aqueous layer was extracted with MTBE (200 mL). The organic layers were combined, washed with a 1:1 mixture of brine and saturated NaHCO3 (aq.), dried ($MgSO_4$), and concentrated to yield 62.83 grams of the N-benzyl imine as a yellow oil. ^{1}H-NMR (500 MHz, $CDCl_3$): 8.30 (s, 1H), 7.78-7.77 (m, 2H), 7.45-7.40 (m, 3H), 4.42 (s, 2H), 3.78 (s, 3H).

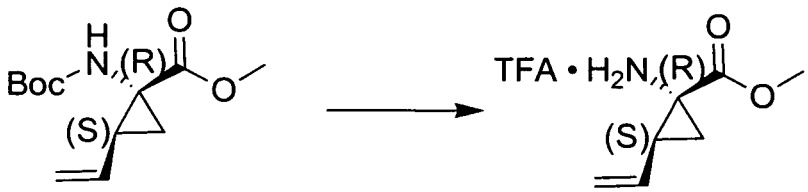


[0331] Lithium tert-butoxide (15.13 g) was suspended in dry toluene (200 mL) at room remperature. To this was added dropwise a solution of the N-benzyl imine of glycine methyl ester (16.89 g) and 1,4-dibromo-2-butene (19.28 g) in toluene (100 mL) over 40 minutes. The red solution was stirred for 100 minutes, then quenched with H_2O (200 mL). The contents were transferred to a separatory funnel and diluted with MTBE (200 mL). The layers were separated and the aqueous layer was extracted with MTBE. The combined organic layers were stirred with 1 N HCl (aq.) (500 mL) for 3 hours. The layers were separated and the organic layer was extracted with H_2O (100 mL). The aqueous layers were combined, NaCl (250 g) and MTBE (700 mL) were added and the pH was brought to ≈13 with 10 N NaOH (aq). The organic layer was separated and the aqueous layer was extracted with MTBE (twice, 300 mL each). The organic layers were combined, dried ($MgSO_4$), and concentrated to a volume of ≈400 mL. To the solution was added di-tert-butyl dicarbonate (25.0 g) and the reaction was stirred for 3 days. Additional di-tert-butyl dicarbonate (5.6 g) was added, followed by heating of the reaction in a 60 °C bath for 1 hour. The reaction was purified by flash silica gel column chromatography with EtOAc/hexane (1:9) as eluent to

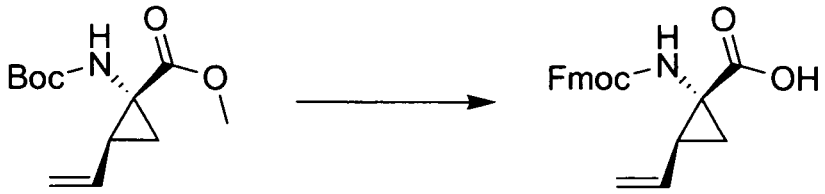
yield 10.89 g of racemic N-Boc-(1R,2S)/(1S,2R)-1-amino-2-vinylcyclopropane carboxylic acid methyl ester. See, e.g., WO00/09558 and Beaulieu, P. L.et al., J. Org. Chem., 70 (15), 5869 -5879, 2005. ^{1}H-NMR (500 MHz, $CDCl_3$): 5.78-5.71 (m, 1H), 5.29-5.26 (m, 1H), 5.11 (dd, J=1.2, 10.3 Hz, 1H), 3.71 (s, 3H), 2.14 (q, J=8.8 Hz, 1H), 1.79 (s, 1H), 1.53-1.45 (m, 10H).



[0332] Racemic N-Boc-(1R,2S)/(1S,2R)-1-amino-2-vinylcyclopropane carboxylic acid methyl ester (4.2 g) was dissolved in acetone (80 mL) and then diluted with water (160 mL). The pH was adjusted to 7.8 with 0.2N NaOH (aq). Subtilisin A (product P-5380 from Sigma, St. Louis, MO, USA) (4.5 g) was added to the solution. Its pH was maintained between 7.4 and 8.7 for 3 days by the dropwise addition of 0.1 N NaOH (aq.). When HPLC analysis (Chiralpak AD from Daicel Chemical Industries, Tokyo, 4.6 mm x 250 mm, 0.5 mL/min, 10-85% 2-propanol/hexanes over 10 minutes, monitor 215.4 nm) of the reaction indicated the presence of only the (1R,2S)-enantiomer (retention time of (1R,2S) = 6.2 min, (1S,2R) = 5.9 min) the pH was brought to 8.5 with 2 N NaOH (aq). The contents of the reaction were transferred to a separatory funnel and extracted with MTBE (3 X 400 mL). The extracts were washed with saturated NaHCO3 (aq) solution (3 X 150 mL), water (2 X 200 mL), and dried (MgSO4). The solution was filtered, concentrated, diluted with CH2Cl2, dried (MgSO4), filtered, and concentrated to yield 1.95 g of N-Boc-(1R,2S)-1-amino-2-vinylcyclopropane carboxylic acid methyl ester.

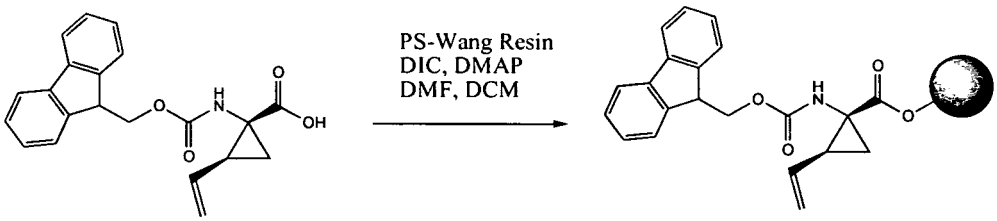


[0333] N-Boc-(1R,2S)-1-amino-2-vinylcyclopropane carboxylic acid methyl ester (125 mg, 0.52 mmol) stirred in CH2Cl2/TFA (1:1, 2 mL) at RT for 90 minutes. Solvents removed under vacuum to yield (1R,2S)-1-amino-2-vinylcyclopropane carboxylic acid methyl ester trifluoroacetic acid salt.



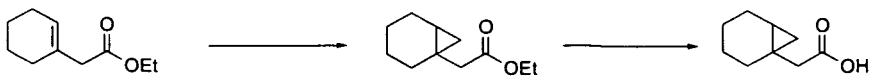
XX1 XX2

[0334] Compound **XX1** (2.34g, 9.71 mmol) was stirred with $LiOH \cdot H_2O$ (0.45 g, 10.7 mmol) in $THF/H_2O/THF$ (3:1:0.5, 22 mL) at room temperature overnight. The solvents were evaporated and the remaining solids were taken up in CH_2Cl_2/EtOAc and 1N HCl (aq). The aqueous layer was extracted with CH_2Cl_2 and the combined organic extracts were dried ($MgSO_4$), filtered, and concentrated. This material was dissolved in CH_2Cl_2 (10 mL) at room temperature and treated with trifluoroacetic acid (10 mL). HPLC analysis at 70 minutes showed no starting material was present. The solvents were removed in vacuo to yield a viscous light colored oil. This was taken up in additional CH2Cl2 (30 mL) and evaporated on a rotary evaporator to yield a tan solid. This solid was dissolved in saturated NaHCO3 (aq) and acetone (1:1, 50 mL) and treated with Fmoc-Cl (2.65 g, 10.2 mmol). After 4 hours, the contents of the flask were transferred to a separatory funnel with CH_2Cl_2 and acidified with 2N HCl (aq). The aqueous layer was extracted with CH_2Cl_2, the combined organic layers were dried (MgSO4), filtered, and concentrated to yield 1.86 g (5.3mmol) of **XX2** as a light yellow solid. (M+1) = 350.1



XX3 XX4

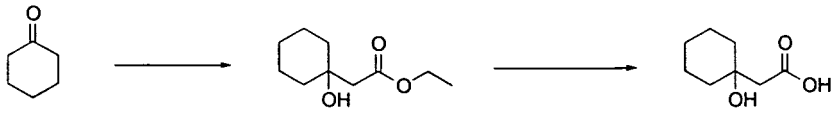
[0335] PS-Wang resin (2.0g, 1.0eq.) swelled in DMF (enough to cover). (1R,2S)-1-(((9H-fluoren-9-yl)methoxy)carbonyl)amino)-2-vinylcyclopropanecarboxylic acid (**XX3**) (922 mg, 1.1 eq.) was stirred in DCM. Diisopropylcarbodiimide (409uL, 1.1 eq.) was added to the DCM solution and stirred at 4 °C for 2 hours, then added to resin and DMF. Dimethylaminopyridine (29mg, 0.1eq.) in DMF was added to resin solution and shaken for 5 hours. Drained and washed with DMF (thrice) and DCM (thrice) to yield Compound **XX4**.



X1 X2

Preparation of 2-(bicyclo[4.1.0]heptan-1-yl)acetic acid X2:

[0336] Commercially available compound **X1** (Aldrich Chemical Co., Milwaukee, Wisconsin, USA) was converted to **X2** according to method described by E. J. Kantorowski et al. in J. Org Chem., 1999, 64, 570-580. ^{1}H-NMR ($CDCl_3$, 500 MHz): 9.2 (br s, 1H), 2.23 (m, 2H), 1.92 (m, 1H), 1.76 (m, 2H), 1.58 (m, 1H), 1.34 (m, 1H), 1.18 (m, 4H), 0.85 (m, 1H), 0.52 (dd, 1H), 0.31 (t, 1H) ppm.

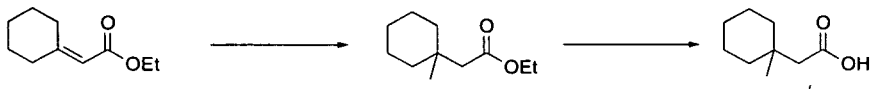


X3 **X4** **X5**

Preparation of 2-(1-hydroxycyclohexyl)acetic acid X5:

[0337] Compound **X4** was prepared using essentially the procedure described in Bull. Chem. Soc. Jpn., 1971, 44, 1090. Specifically, A solution of ethylbromoacetate (8.3 mL) (Aldrich Chemical Co., Milwaukee, Wisconsin, USA) in toluene was added dropewise at 80°C over 30 min. to a thoroughly stirred mixture of cyclohexanone **X3** (4.9 g) and zinc powder (4.9 g) in toluene. The addition was carefully monitored and the temperature was kept at 80 °C. After the addition was completed, the mixture was refluxed for 90 min., cooled, decomposed with 1N aqueous HCl, and extracted with Et_2O. The organics were washed with water, aq. $NaHCO_3$, dried ($MgSO_4$) and concentrated in vacuo to yield **X4** (5.9g). ^{1}H-NMR ($CDCl_3$, 500MHz) 4.16 (t, 2H), 3.0 (br s, 1H), 2.46 (s, 2H), 1.40-1.69 (m, 10H), 1.27 (t, 3H) ppm; FIA m/z 187.1 ES^+.

[0338] To a solution of **X4** (510 mg) in MeOH was added 1N aqueous NaOH. The reaction mixture was stirred at 60°C for 1h, and then concentrated in vacuo. The residue was diluted with water, washed with Et_2O and the aqueous layer acidified with 1N aqueous citric acid and extracted with EtOAc. The organics were dried ($MgSO_4$) and concentrated in vacuo to yield after recrystallization compound **X5** (220mg): ^{1}H-NMR ($CDCl_3$, 500MHz) 3.63 (s, 1H), 2.45 (s, 2H), 1.22-1.64 (m, 10H) ppm; FIA m/z 157.2 ES^-.



X6 **X7** **X8**

Preparation of 2-(1-methylcyclohexyl)acetic acid (X8)

[0339] Commercially available compound **X6** (Aldrich Chemical Co., Milwaukee, Wisconsin, USA) was converted to compound **X7** according to the method described by N.

Asao et al. in Tetrahedron Lett., 2003, 44, 4265. ^{1}H-NMR ($CDCl_3$, 500 MHz): 4.12 (q, 2H), 2.22 (s, 2H), 1.30-1.48 (m, 10H), 1.25 (t, 3H), 1.01 (s, 3H) ppm.

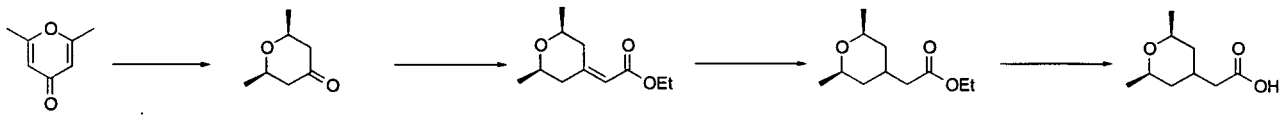
[0340] To a solution of compound **X7** in EtOH was added 1 N aqueous NaOH. The reaction mixture was stirred at 50 °C for 3 hours, and then concentrated in vacuo. The residue was diluted with water, washed with Et_2O and the aqueous layer acidified with 1 N aqueous citric acid and extracted with CH_2Cl_2. The organics were dried ($MgSO_4$) and concentrated in vacuo to yield compound **X8**. ^{1}H-NMR ($CDCl_3$, 500MHz): 11.7 (s, 1H), 2.26 (s, 2H), 1.32-1.49 (m, 10H), 1.05 (s, 3H) ppm.

X9 **X10** **X11** **X12**

Preparation of 2-(4-methyltetrahydro-2H-pyran-4-yl)acetic acid (X12)

[0341] To a solution of dihydro-2H-pyran-4(3H)-one (**X9**) (3.13 g, from Aldrich) in toluene was added (carbethoxymethylene)-triphenylphosphorane (12.0 g, Aldrich). The solution was stirred at 110 °C for 3 days. The resulting dark solution was concentrated in vacuo and the residue directly purified by column over silica gel to yield compound **X10** (4.54 g) as a clear liquid. ^{1}H-NMR ($CDCl_3$, 500MHz): 5.66 (s, 1H), 4.16 (q, 2H), 3.98 (s, 4H), 3.00 (t, 2H), 2.38 (m, 2H), 1.77 (m, 4H), 1.27 (t, 3H) ppm.

[0342] Compounds **X11** and **X12** were obtained in a similar manner as described for compounds **X7** and **X8**. ^{1}H-NMR ($CDCl_3$, 500 MHz): 3.64-3.73 (m, 4H), 2.35 (s, 2H), 1.65 (ddd, 2H), 1.50 (ddt, 2H), 1.17 (s, 3H) ppm.



X13 **X14** **X15** **X16**

Preparation of 2-(cis-2,6-dimethyltetrahydro-2H-pyran-4-yl)acetic acid (X16)

[0343] Intermediate **X13** was prepared from commercially available 2,6-dimethyl-g-pyrone (Aldrich Chemical Co., Milwaukee, Wisconsin, USA). A solution of the g-pyrone was dissolved in EtOH and hydrogenated (2 atm. H_2) with 10% Pd/C over 2h. The catalyst was subsequently filtered off and the solution was concentrated in vacuo to yield crude **X13** which was purified by column chromatography to yield pure compound **X13**. ^{1}H-NMR ($CDCl_3$, 500 MHz): 3.72 (m, 2H), 2.35 (m, 2H), 2.21 (dd, 2H), 1.32 (d, 6H) ppm.

[0344] Compound **X14** was then obtained from compound **X13** in a similar manner as described for compound **X10**. ^{1}H-NMR ($CDCl_3$, 500 MHz): 5.65 (s, 1H), 4.15 (q, 2H), 3.80

(dt, 1H), 3.49 (m, 2H), 2.17 (dt, 1H), 2.07 (dd, 1H), 1.79 (dt, 1H), 1.28 (m, 9H) ppm. LC-MS m/z 199.126 ES^+.

[0345] A solution of compound **X14** in EtOAc was then hydrogenated (1 atm. H_2) with 10% wet Pd/C over 1 hour. The catalyst was subsequently filtered off and the solution was concentrated in vacuo to yield crude compound **X15** which was used without further purification for the next step. Compound **X16** was then prepared from compound **X15** in a similar manner as described for compound **X8**. ^{1}H-NMR ($CDCl_3$, 500 MHz) major diastereomer: 3.50 (m, 2H), 2.27 (d, 2H), 2.07 (m, 1H), 1.71 (m, 2H), 1.19 (d, 6H) 0.92 (m, 2H) ppm; major diastereomer: 3.64 (m, 2H), 2.56 (d, 2H), 2.47 (m, 1H), 1.49 (m, 2H), 1.15 (d, 6H), 0.86 (m, 2H) ppm.

X17 **X18** **X19** **X20**

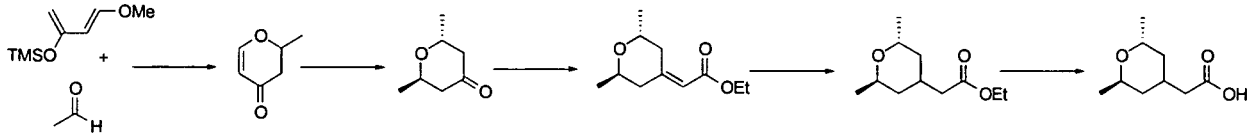
Preparation of 2-(1,4-dioxaspiro[4.5]decan-8-yl)acetic acid X20:

[0346] Compound **X20** was prepared from compound **X17** (from Aldrich) according to the procedures described above for preparing compound **X16**.

[0347] Compound **X18**: ^{1}H-NMR ($CDCl_3$, 500 MHz): 5.66 (s, 1H), 4.15 (q, 2H), 3.98 (s, 4H), 3.00 (m, 2H), 2.38 (m, 2H), 1.77 (m, 4H), 1.27 (t, 3H) ppm.

[0348] Compound **X19**: ^{1}H-NMR ($CDCl_3$, 500 MHz): 4.12 (q, 2H), 3.93 (s, 4H), (d, 2H), 1.83 (m, 1H), 1.72 (m, 4H), 1.56 (dt, 2H), 1.33 (m, 2H), 1.30 (m, 3H) ppm.

[0349] Compound **X20**: ^{1}H-NMR ($CDCl_3$, 500 MHz): 3.93 (s, 4H), 2.28 (d, 2H), 1.73-1.86 (m, 4H), 1.57 (dt, 2H), 1.35 (m, 2H) ppm.



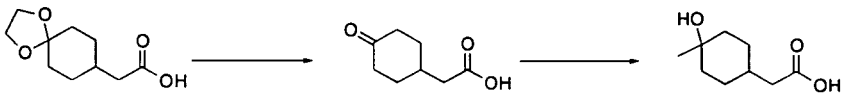
X21 **X22** **X23** **X24** **25**

Preparation of 2-(trans-2,6-dimethyltetrahydro-2H-pyran-4-yl)acetic acid 25:

[0350] Compounds **X21** and **X22** were prepared according to the method described by S. Danishefsky et al. in J. Org. Chem. 1982, 47, 1597-1598 and D. S. Reddy et al. in J. Org. Chem. 2004, 69, 1716-1719, respectively. Compound **X25** was prepared from compound **X22** according to the method described above for preparing compound **X16**.

[0351] Comopund **X23**. ^{1}H-NMR ($CDCl_3$, 500 MHz): 5.72 (s, 1H), 4.16 (q, 2H), 4.08 (q, 2H), 3.06 (dd, 1H), 2.75 (dd, 1H), 2.39 (dd, 1H), 2.05 (dd, 1H), 1.28 (t, 3H), 1.19 (m, 6H) ppm.

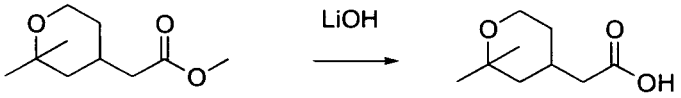
[0352] **X25**: ^{1}H-NMR ($CDCl_3$, 500 MHz) 4.24 (m, 1H), 3.78 (m, 1H), 2.25 (m, 3H), 1.71 (m, 1H), 1.53 (m, 1H), 1.46 (m, 1H), 1.29 (d, 3H), 1.13 (d, 3H), 0.90 (m, 1H) ppm.



X20 **X26** **X27**

Preparation of 2-(4-hydroxy-4-methylcyclohexyl)acetic acid X27:

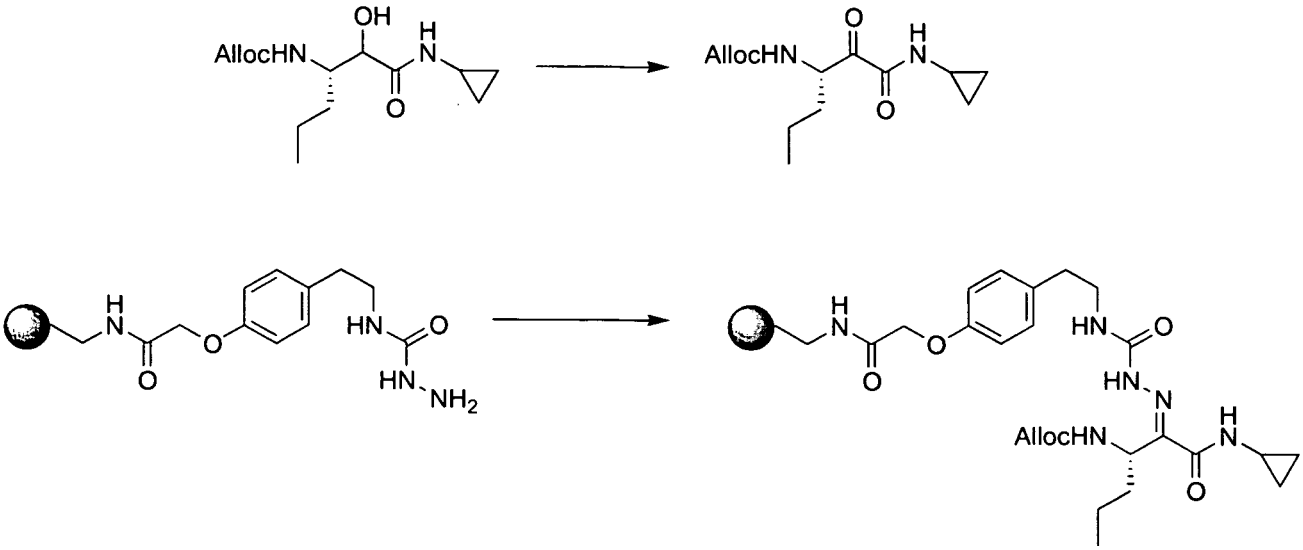
[0353] A solution of compound **X20** in dioxane was treated with 4N HCl in dioxane. The reaction solution was stirred at roomtemperature for 4 hours and concentrated in vacuo to give crude compound **X26** which was used without further purification for the next step. To a stirred solution of compound **X26** in THF was slowly added MeMgBr (3 N in THF). The resulting mixture was stirred at 40 °C for 3 hours, quenched with 1 N aqueous citric acid and diluted with EtOAc. The phase were separated and the organics were dried ($MgSO_4$), concentrated in vacuo and purified by chromatography over silica gel to give compound **X27** as a mixture of two diastereomers: isomer 1: ^{1}H-NMR ($CDCl_3$, 500 MHz): 4.50 (br s), 2.27 (m, 2H), 1.75 (m, 1H), 1.65 (m, 4H), 1.39 (m, 4H), 1.22 (s, 3H) ppm; isomer 2: ^{1}H-NMR ($CDCl_3$, 500 MHz): 2.12 (m, 2H), 1.69 (m, 3H), 1.56 (m, 2H), 1.39 (m, 2H), 1.12 (s, 3H), 1.05 (m, 2H) ppm.



Preparation of 2-(2,2-dimethyltetrahydro-2H-pyran-4-yl)acetic acid

[0354] To a solution of the methyl ester (500 mg; 2.69 mmol) in THF (21.5 mL), MeOH (21.5 mL) and water (10.75 mL) was added LiOH (1 N; 10.75 mL). The reaction mixture was stirred for 3 hours. The reaction was acidified with HCl (1 N, pH = 5). The product was extracted with EtOAc (twice, 20 mL each). The combined organic layer was then wash with water, brine and concentrated in vacuo to afford 420 mg of 2-(2,2-dimethyltetrahydro-2H-pyran-4-yl)acetic acid. ^{1}H-NMR ($CDCl_3$): δ 3.76-3.67 (m, 2H), 2.56-2.19 (m, 3H), 1.63 (m, 2H), 1.26-1.10 (m, 8H). (M+1) 173.

X30 X31



X32 X33

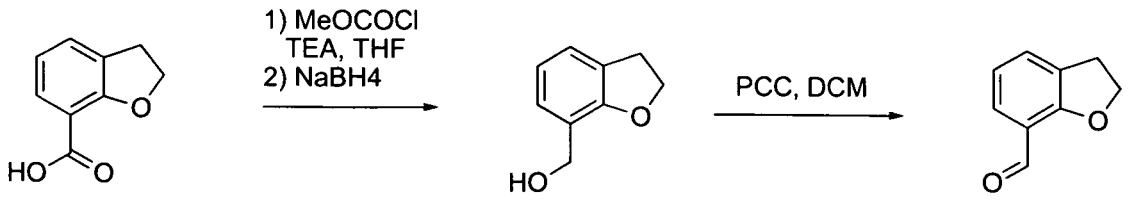
[0355] To a solution of compound **X30** (64 g, 237 mmol) and EDC (226 g, 1.19 mol) in EtOAc (1.5 L) was added DMSO (400 mL), and the resulting suspension was cooled to 0 °C. To this mixture was added a solution of dichloroacetic acid in EtOAc (1:1 v/v, 130 mL) keeping the internal reaction temperature below 25 °C. The reaction was warmed to room temperature, stirred for 15 minutes, cooled to 0 °C, and quenched with 1 N HCl (1 L). The organic layer was separated, washed with H_2O (2 x 500 mL), dried over $MgSO_4$, and concentrated under reduced pressure. The resulting oil was filtered through a plug of silica eluting with EtOAc / hexanes to afford 48 g of compound **X31** as a white solid.

[0356] To resin **X32** (prepared according to the procedure described in WO 00/23421) (100 g, 0.88 mmol/g) was added a solution of **X31** (48 g, 179 mmol) in THF (650 mL), followed by AcOH (30 mL). The mixture was shaken for 16 hours, and the resin was filtered, washed with THF (4 times, 400 mL each) and CH_2Cl_2 (4 times, 400 mL each) and dried in vacuo. The filtrate and washes were combined and concentrated, and the above procedure was repeated to afford resin **X33** with a loading of approximately 0.4 mmol/g.

Preparation of Aldehyde Compounds

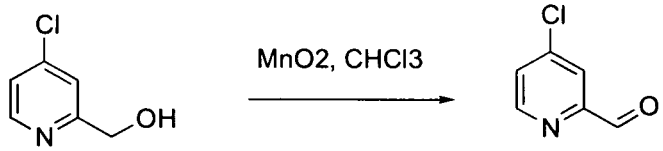
[0357] 5-chloronicotinaldehyde was prepared according to methods described by D.L. Comins et al. in Hetereocycles, 1987, 26 (8), pp. 2159-2164.

[0358] Some other aldehydes such as 2-fluoro-5-chlorobenzaldehyde, 2-methoxy-3-methyl benzaldehyde, 2-methoxynicotinaldehyde, 2,3-dihydrobenofuran-7-carbaldehyde can be made from corresponding acid based on following procedure:



Preparation of 2,3-dihydrobenzofuran-7-carbaldehyde

[0359] 2,3-Dihydrobenzofuran-7-carboxylic acid (820 mg, 5 mmol) was dissolved in THF (10 mL). To the solution was added TEA (0.7 mL, 5 mmol) and methylchloroformate (0.43 mL, 5 mmol). The solution was stirred for 0.5 hour. The white precipitates were removed by filtration, the filtrate was added to a solution of $NaBH_4$ (437 mg, 12.5 mmol) in H_2O (5 mL). The resulting solution was stirred overnight. The reaction mixture was neutralized with 2 M aqueous HCl solution and then extracted with EtOAc. The organic layer was washed with brine, dried over anhydrous Na_2SO_4 and concentrated in vacuo. The crude alcohol was dissolved in DCM. To the solution was added PCC (1.83 g, 7.5 mmol). The mixture was stirred for 2 hours at room temperature and diluted with diethyl ether, then ether layers were decanted. Combined organic layer was filtered though a layer of Celite®. The filtrate was concentrated to give crude product. The crude was purified from column with 10% EtOAc/hexane to afford 450 mg of 2,3-dihydrobenzofuran-7-carbaldehyde as a slightly yellow solid. HPLC 4.3 min.



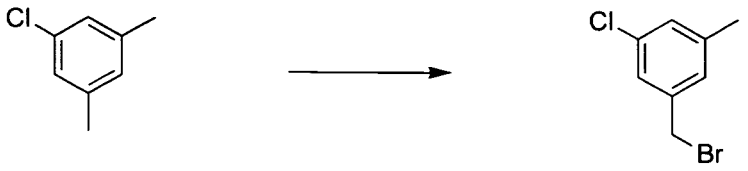
Preparation of 4-chloropicolinaldehyde

[0360] A suspension of MnO_2 (7.3 g, 84 mmol) and (4-chloro-pyrindin-2-yl)methanol (1 g, 7 mmol) in $CHCl_3$ was heated to refulx for 90 minutes. The mixture was filtered though a layer of Celite® and concentrated in vacuo to afford 520 mg of 4-chloropicolinaldehyde as a white solid. HPLC 1.8 minutes and MS 142 as M=1 peak.

Preparation of 3-chloro-5-methoxybenzaldehyde

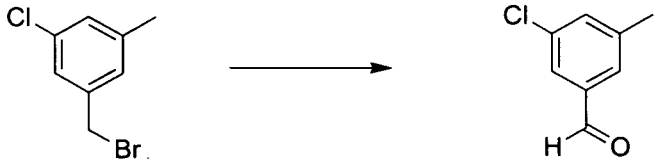
[0361] A mixture of 3-chloro-5-methoxybenzyl alcohol (5.0g, 28.9 mmol) and pyridinium chlorochromate (20% on alumina, 40g, 37.8 mmol) was allowed to stir for 1.25 hr. Diethyl ether (200ml) was then added followed by filtration of precipitate. The filtrate

was concentrated under reduced pressure and the resulting residue was purified via silica gel chromatography using 40% dichloromethane, 60% petroleum ether as eluant, to give 3.8g of 3-chloro-5-methoxybenzaldehyde. ^{1}H-NMR ($CDCl_3$): 3.84 (s, 3H) 7.13 (s, 1H), 7.28 (s, 1H), 7.41 (s, 1H), 9.89 (s, 1H).



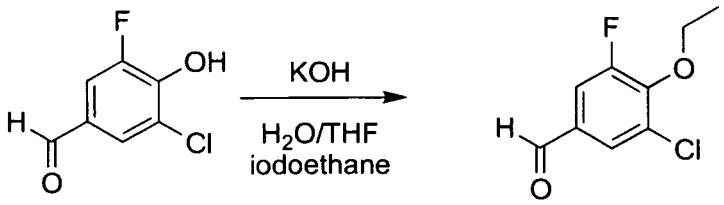
Preparation of 1-(bromomethyl)-3-chloro-5-methylbenzene

[0362] To a solution of m-chloroxylene (0.96g, 6.8mmol) in carbon tetrachloride at reflux was added N-bromosuccinmide (1.4g, 7.5 mmol) followed by benzoyl peroxide (1.6g, 6.8 mmol). The reaction was allowed to stir for 20 minutes and cooled to room temperature, filtered off precipitate and the filtrate was concentrated under reduced pressure and the resulting residue was purified via silica gel chromatography using petroleum ether as eluant to give 0.89g of 1-(bromomethyl)-3-chloro-5-methylbenzene. NMR ($CDCl_3$): 2.31 (s,3H) 4.37 (s,2H) 7.09 (s,1H) 7.12 (s,1H) 7.20 (s,1H).

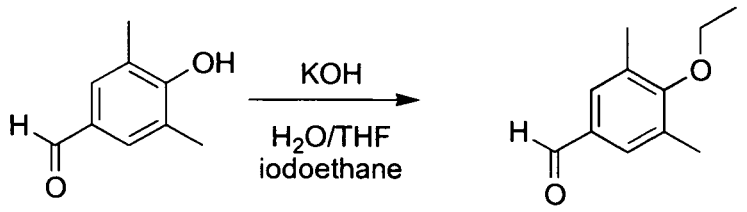


Preparation of 3-chloro-5-methylbenzaldehyde

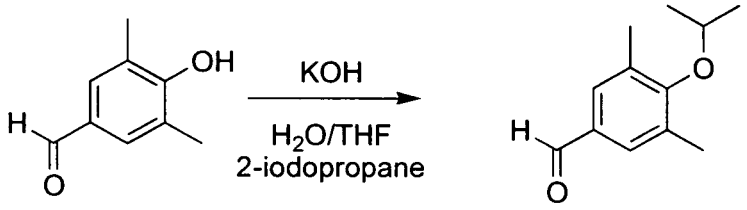
[0363] To a solution of sodium metal (52 mg, 2.3mmol) in ethanol was added 2-nitropropane (0.23g, 2.4 mmole) followed by the addition of 3-chloro-5-methybenzylbromide (0.5g, 2.3 mmol). The reaction was allowed to stir for 3 hours and the precipitate formed was filtered off. The filtrate was concentrated under reduced pressure, redissolved in diethylether and washed with 1N sodium hydroxide (twice), water, and dried over sodium sulfate, filtered and the filtrate was concentrated under reduced pressure. The resulting residue was purified via silica gel chromatography using 10% dichloromethane and 90% petroleum ether, to give 0.15g of 3-chloro-5-methylbenzaldehyde. ^{1}H-NMR ($CDCl_3$): 2.46 (s, 3H) 7.43(s, 1H) 7.56 (s, 1H) 7.68(s, 1H), 9.92 (s, 1H).



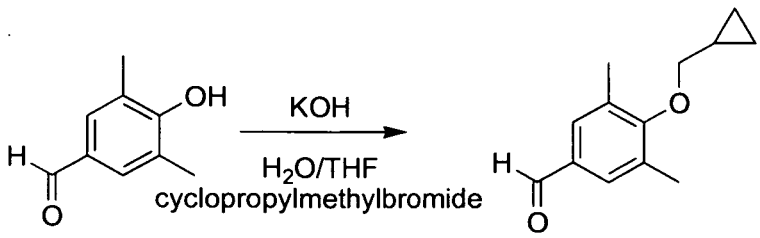
[0364] 3-Chloro-5-fluoro-4-hydroxybenzaldehyde (1.0 gram, 5.7 mmol) in THF (40mL) was heated at reflux for 17 hours with KOH (534 mg, 9.5 mmol, 1.7 eq) in water (5 mL) and iodoethane (1 mL, 2.2 eq). The reaction was then transferred to a separatory funnel with water and extracted with methylene chloride (thrice, 150 mL each). The combined organic layers were washed with 10 % aqueous HCl (40 mL), dried ($MgSO_4$), and concentrated to a viscous orange liquid to yield 1.13 g of 3-chloro-4-ethoxy-5-flurobenzaldehyde. ^{1}H-NMR (500 MHz, $CDCl_3$): 9.84 (d, J=1.9 Hz, 1H), 7.71 (t, J=1.6 Hz, 1H), 7.53 (dd, J=1.9, 10.7 Hz, 1H), 4.37-4.32 (m, 2H), 1.47-1.40 (m, 3H).



[0365] 4-Ethoxy-3,5-dimethylbenzaldehyde was prepared in a manner similar to that of 3-chloro-4-ethoxy-5-flurobenzaldehyde. ^{1}H-NMR (300 MHz, $CDCl_3$): 9.89 (s, 1H), 7.56 (s, 2H), 3.91 (q, 7 Hz, 1H), 2.34 (s, 6H), 1.44 (t, J=7 Hz, 6H).

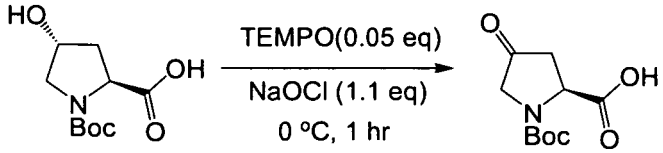


[0366] 4-Isopropoxy-3,5-dimethylbenzaldehyde was prepared in a manner similar to that of 4-Ethoxy-3,5-dimethylbenzaldehyde. ^{1}H-NMR (300 MHz, $CDCl_3$): 9.88 (s, 1H), 7.55 (s, 2H), 4.31 (q, J=6 Hz, 1 H), 2.32 (s, 6H), 1.32 (d, J=6 Hz, 6H).



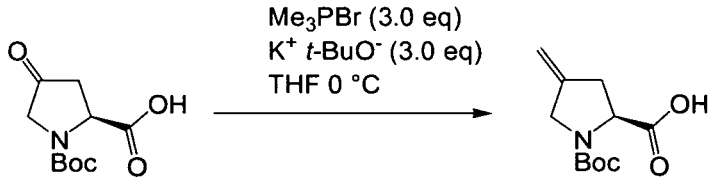
[0367] 4-(Cyclopropylmethoxy)-3,5-dimethylbenzaldehyde was prepared in a manner similar to that of 4-Ethoxy-3,5-dimethylbenzaldehyde. ^{1}H-NMR (300 MHz, $CDCl_3$): 9.87 (s, 1H), 7.55 (s, 2H), 3.69 (d, J=7 Hz, 2H), 2.35 (s, 6H), 1.35-1.23 (m, 1H), 0.67-.060 (m, 2H), 0.35-0.30 (m, 2H).

Preparation of (S)-1-(tert-butoxycarbonyl)-4-oxopyrrolidine-2-carboxylic acid.



[0368] A solution of (2S,4R)-1-(tert-butoxycarbonyl)-4-hydroxypyrrolidine-2-carboxylic acid (1.0 eq.) in isopropyl acetate (5 vol) was cooled to 0 °C and TEMPO (0.05 eq.) was added. A solution of bleach (12.5 wt %, 1.2 eq., 2.6 vol) was then slowly added over 1 hour while maintaining the temperature at 0-5 °C. The mixture was stirred and monitored by HPLC for completion, then aqueous 10% $KHSO_4$ (2.5 vol) was added, stirred for 10 minutes, and then the phases were separated. The organic phase was washed with aqueous 5% Na_2SO_3 (2 vol) then brine (1 vol) then dried azeotropically and concentrated to afford the title compound as a solid. The solid was triturated with acetonitrile (1.0 vol) to remove residual color and impurities. ^{1}H-NMR (400 MHz, DMSO): δ 4.54 (m, 1H), 3.82 (m, 1H), 3.67 (m, 1H); 3.15 (m, 1H); ≈ 2.50 (m, 1H, coincides with DMSO); 1.42 and 1.39 (2 s rotamers, 9H).

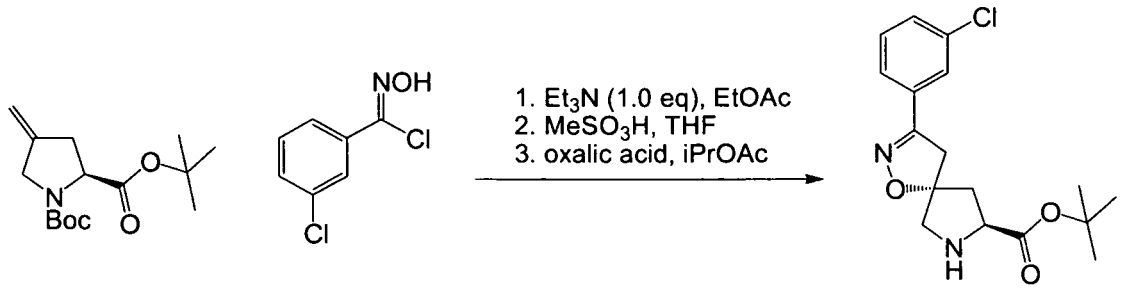
Preparation of (S)-1-(tert-butoxycarbonyl)-4-methylenepyrrolidine-2-carboxylic acid



[0369] To a suspension of methyltriphenylphosphonium bromide (2.2 eq.) in 2-methyl tetrahydrofuran (3 vol) was added rapidly solid potassium tert-butoxide (2.3 eq.) maintaining the temperature around 0 °C. The temperature was kept at +20 °C for 2 hours (a suspension remained) and re-cooled to 0 °C. Keeping the temperature below 6 °C, (S)-1-(tert-butoxycarbonyl)-4-oxopyrrolidine-2-carboxylic acid (1 eq.) was added over 40 minutes. The reaction was warmed to room temperature and stirred for 16 h and then cooled to 0 °C. The reaction was quenched with saturated $NaHCO_3$ (5 vol) and water (2 vol) and the aqueous layer was separated. The organic layer was extracted with saturated $NaHCO_3$/water (1.8 vol/1.8 vol) and the combined aqueous layers were filtered through Celite®. The aqueous layer was acidified with 6 N HCl (2.6 vol) at ambient temperature and extracted twice with isopropyl acetate (16 vol, then 8 vol). The organic phase was dried ($MgSO_4$) and the solvent removed. The crude product was dissolved in isopropyl acetate (10 vol) and extracted with 0.5 M NaOH (10 vol, then 1 vol). The combined aqueous layers were acidified at ambient

temperature with 6 N HCl to pH = 3, and extracted twice with ethyl acetate (10 vol, then 8 vol). The combined extracts were dried (Na_2SO_4), the solvent removed and the crude product was recrystallized from cyclohexane (5 vol) to afford the title compound. ^{1}H-NMR (400 MHz, DMSO): δ 12.9, (broad, 1H); 5.00 (m, 2H); 4.24 (dt, J=1.9 H, J=7.3 Hz, 1H), 3.91 (m, 2H); 2.98 (m, 1H); ≈ 2.50 (m, 1H, coincides with DMSO); 1.41 and 1.36 (2 s rotamers, 9H).

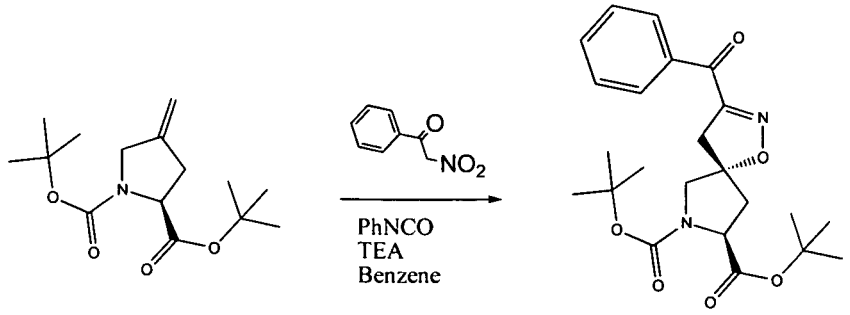
Preparation of (5S,8S)-tert-butyl 3-(3-chlorophenyl)-1-oxa-2,7-diazaspiro[4.4]non-2-ene-8-carboxylate.



[0370] A solution of 3-chloro-N-hydroxybenzimidoyl chloride (175 g, 0.919 moles) in EtOAc (2.1 L) was added to a solution of (S)-di-tert-butyl 4-methylenepyrrolidine-1,2-dicarboxylate (200 g, 0.707 moles) in EtOAc (2.0 L) at room temperature. The mixture was cooled below 10 °C in an ice bath, then triethylamine (128 mL, 0.919 moles) was added slowly. The resultant mixture was stirred overnight then quenched with water (3 L). The phases were separated and the organic phase washed with water (2 x 1.0 L), dried over $MgSO_4$, and the solvent removed to afford a mixture of the syn- and anti- spiroisoxazolines as an oil.

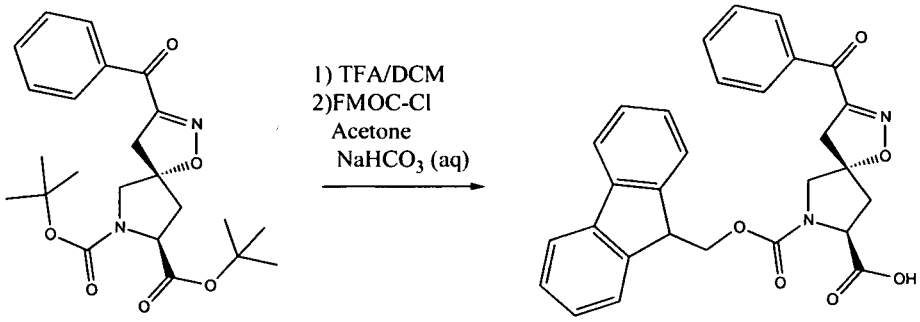
[0371] The mixture of isomers was dissolved in THF (0.72 L) and cooled to 20 °C. Methanesulfonic acid (150 mL) was slowly added maintaining 20 to 30 °C. The mixture was stirred at 25 °C and quenched after 7 hours by carefully adding a solution K_2CO_3 (300 g) in water (1 L). The phases were separated and the aqueous phase was extracted with isopropyl acetate (1 L). The organic phases were combined and approximately half of the solvent removed under vacuum. The solution was washed with a 1:1 mixture of saturated brine (250 mL) and water (250 mL). The aqueous phase was extracted with isopropyl acetate (200 mL) and the organic phases combined then dried over K_2CO_3 and filtered to afford a homogeneous solution. The solution volume was made up to 3 L by adding isopropyl acetate and then a solution of oxalic acid (20 g) in isopropyl acetate (400 mL) was slowly added. The solid was isolated by filtration and dried in a vacuum oven. The solid was suspended in isopropyl acetate (1.5 L) and water (1.0 L) then K_2CO_3 was added slowly until the solids fully dissolved. The organic layer was isolated, dried over K_2CO_3, filtered then a solution of

oxalic acid (12.5 g) in isopropyl acetate (250 mL) was added slowly. The solid was isolated by filtration and dried in a vacuum oven to give the spiroisoxazolines as a 98:2 anti-:syn- mixture of diastereomers. ^{1}H-NMR (400 MHz, DMSO-d_6): δ 7.67-7.48 (m, 4H), 4.08 (dd, J=7.9, 8.9 Hz, 1H), 3.55 (s, 2H), 3.27 (d, J=4.0 Hz, 2H), 2.46 (dd, J=7.8, 13.8 Hz, 1H), 2.19 (dd, J=9.1, 13.8 Hz, 1H), 1.46 (d, J=7.5 Hz, 9H).



M2B **X37**

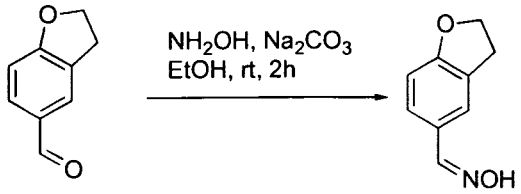
[0372] Compound **M2B** (1.0g, 1.0eq) was stirred in 20 mL benzene with benzoylnitromethane (583 mg, 1.0 eq.) and catalytic triethylamine. Phenyl isocyanate (880uL) was added slowly and stirred for 40 hours. Dark colored precipitate was filtered off and to the filtrate was added 2mL water and the mixture was stirred for 2 hours. Organics were separated and concentrated, purified by silica gel chromatography (10-90% ethyl acetate/hexanes gradient) to give 350mg of Compound **X37**. (M+H=431.2.) ^{1}H-NMR (500 MHz, $CDCl_3$): 8.19 (d, 2H), 7.61 (t, 1H), 7.56-7.46 (m, 2H), 4.45-4.36 (m, 1H), 3.99-3.88 (m, 1H), 3.61 (d, 1H), 3.39-3.33 (m, 2H), 2.77 (m, 1H), 2.17-2.12 (m, 1H), 1.49 (s, 9H) 1.46 (s, 9H).



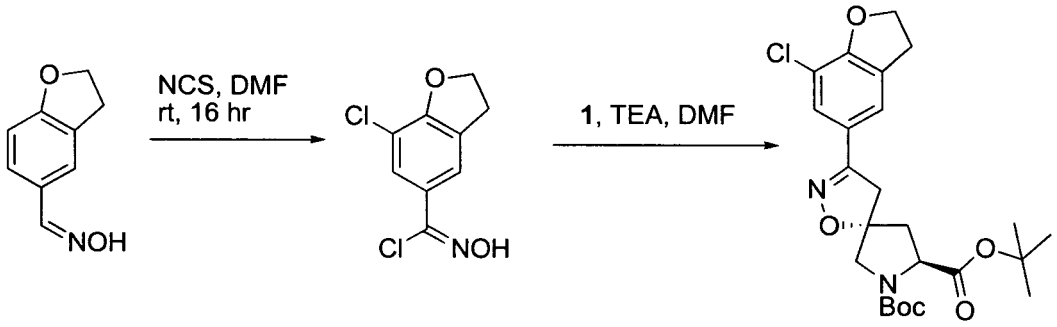
X37 **X38**

[0373] Compound **X37** (1.35 g. 1.0 eq.) was stirred in 20 mL 1/1 TFA/DCM for 2 hours. The mixture was concentrated and to it was added 20mL acetone, 20mL saturated sodium bicarbonate solution, and FMOC-Cl (1.22 g, 1.5 eq.). The mixture was stirred for 3 hours and diluted with ethyl acetate and a 2 N HCl solution until aqueous became acidic. The mixture was stirred, aqueous extracted with ethyl acetate, combined organics, dried over

magnesium sulfate, filtered, and concentrated. The concentrate was purified by silica gel chromatography (100% DCM-10%MeOH/DCM gradient) to give compound **X38**. (M+H=497.1).



[0374] To a solution of 2,3-dihydrobenzofuran 5-carboxaldehyde (1g, 6.75 mmol) in ethanol (5 mL) was added a 2.4 M of NH_2OH (3.3 mL, 8.1 mmol) solution and then 1.2 M of Na_2CO_3 (3.3 mL, 4.05 mmol). The resulting solution was stirred for 2 hours at room temperature (HPLC showed no starting material left). The reaction mixture was diluted with EtOAc, washed with brine, dried over Na_2SO_4 and concentrated under vacuum. This afforded 1.0 g of the product as a white solid. ES-MS 164 as M+1 peak.

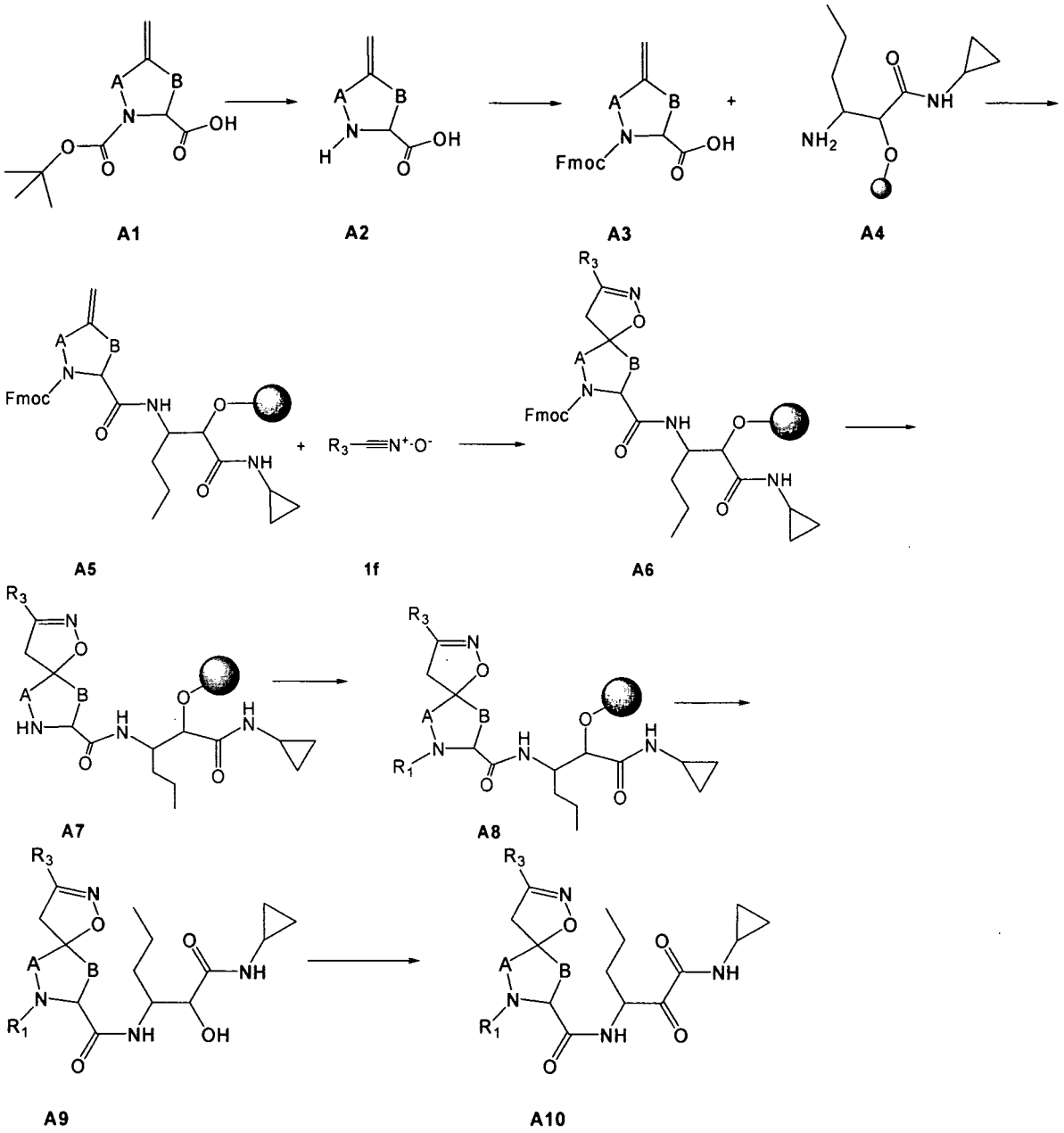


[0375] To a solution of aldoxime (426 mg, 2.6 mmol) in DMF (5 mL) was added NCS (697 mg, 5.2 mmol). The resulting mixture was stirred for overnight at room temperature. To the solution was added (S)-di-tert-butyl 4-methylenepyrrolidine-1,2-dicarboxylate, compound **1** (600 mg, 2.1 mmol) and then a solution of TEA (0.37 mL, 2.6 mmol) in DMF (2 mL) was added over 10 minutes. The reaction mixture was stirred for 4 hr at room temperature and then heated to 50-60 °C for 2 hours. The reaction mixture was diluted with EtOAc (20 mL) and washed with H_2O, brine, dried over Na_2SO_4, concentrated in vacuo. The crude products were purified from flash column chromatography eluted with 30% EtOAc/Hexane, to afford S (500-600 mg) (Rf = 0.3) and R isomer (150 mg) (Rf= 0.2). ES-MS 479 as M+1 peak.

B. Synthesis of Exemplary Compounds of Formula I

[0376] Certain exemplary compounds of Formula I may be prepared by Method 1 as illustrated below.

METHOD 1:



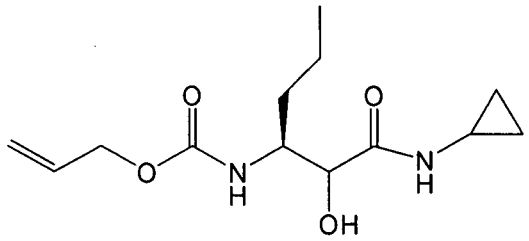
[0377] Referring to Method 1, the exomethylene compound **A1** is deprotected to **A2**, which is converted to the corresponding Fmoc derivative **A3**. Reaction of the resin bound aminoalcohol **A4** with **A3** in the presence of a coupling reagent provides the resin bound product **A5**. A dipolar addition reaction of **A5** with the nitrile oxide **1f**, generated in situ, provides the resin bound spiroisoxazoline **A6**, which is deprotected to provide the resin bound spiroisoxazoline **A7**. Reaction of **A7** with an R_1-carboxylic acid in the presence of a coupling agent provides **A8**, wherein R_1 is $R_4C(O)$-. Cleavage of the spiroisoxazoline from the resin provides the alcohol **A9**. Oxidation of **A9** with an oxidizing reagent such as Dess-

Martin periodinane or sodium hypochlorite in the presence of TEMPO provides the final compound **A10**.

[0378] In some instance, R_4 may contain an amine functionality. Where R_4 contains a protected amine, deprotection of the protected amine to give a free amine, following by a reaction with an activated acid, provides a further elaborated R_4. Alternatively, a free amine in R_4 may be converted to the corresponding p-nitrophenylcarbamate followed by ractions with an amine or alcohol to provide R4 compounds containing carbamate or urea functionarity.

Preparation of Allyl 1-(cyclopropylamino)-2-(6-(hydroxymethyl)tetrahydro-2H-pyran-2-yloxy)-1-oxohexan-3-ylcarbamate (M1B).

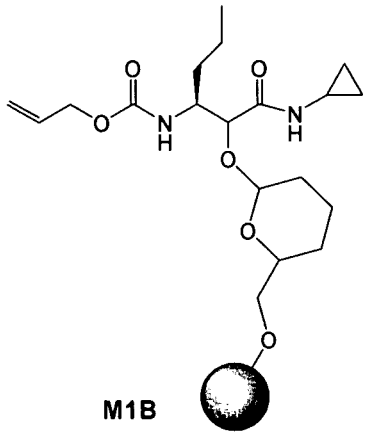
Step 1: Allyl 1-(cyclopropylamino)-2-hydroxy-1-oxohexan-3-ylcarbamate (M1A).



M1A

[0379] To a solution of (3S)-3-amino-N-cyclopropyl-2-hydroxyhexanamide (10 g, 53.7 mmol), DIEA (28 mL, 161 mmol, 3 eq.) in methylene chloride (250 mL) was added dropewise at 0 °C to a solution of allylchloroformate (6.8 mL, 64.4 mmol, 1.2 eq.) in DCM (50 mL). The reaction solution was warmed to room temperature and stirred for 4 hours. Water (300 mL) was then slowly added followed by aqueous HCl (1.0 N, 300 mL). The phases were separated and the organics washed with saturated aqueous $NaHCO_3$ (300 mL), brine (300 mL), dried with $MgSO_4$, filtered, and concentrated in vacuo. The resulting off-white solid was recrystallized from 30% hexanes in EtOAc (120 mL) to yield the title compound **M1A** as a white solid. The mother liquor was concentrated, in vacuo, and recrystallized from 50% hexanes in EtOAc to yield another 4.04 g of **MIA**. The mother liquor from the second recrystallization was concentrated in vacuo on Celite®, and the resulting Celite® plug was purified by flash chromatography (Isco Companion®, SiO_2, DCM to 70%EtOAc in DCM) to give 1.46 g of **MIA**. The total amount of compound **MIA** was 13.4 g. (Rf ≈ 0.40 in 1:1 DCM:EtOAc, CAM detection).

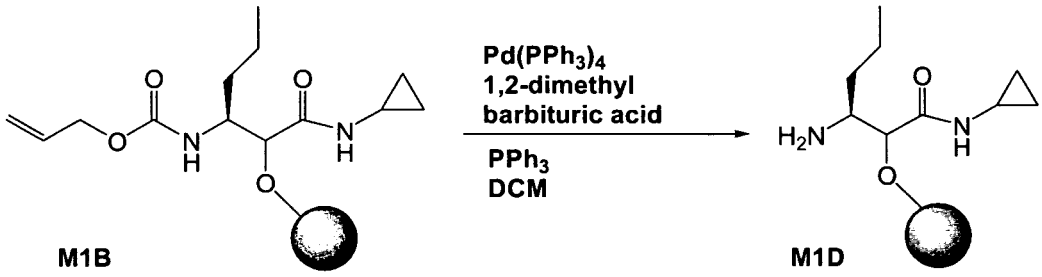
Step 2: Allyl 1-(cyclopropylamino)-2-(6-(hydroxymethyl)tetrahydro-2H-pyran-2-yloxy)-1-oxohexan-3-ylcarbamate bound resin (M1B).



[0380] A 500 mL two neck round bottom flask equipped with an overhead mechanical stirrer and a reflux condenser was charged with **M1A** (9.08g, 33.6 mmol, 3 eq.), pyridinium p-toluenesulfonate (5.6g, 22.4 mmol, 2 eq.), DHP-resin (10.2g, 11.2 mmol, Novabiochem, Cat# 01-64-0192, loading: 1.1 mmol/g), and dichloroethane (84 mL, $[0.4]_I$). The mixture was gently stirred at 80 °C for 3 days, before being cooled to 50 °C and filtered. The resin was washed with DCM (200 mL) and the combined filtrate were concentrated in vacuo to give the resin **M1B**, which was additionally washed with DCM (twice), DMF (thrice), DCM-MeOH (thrice in succession), Et_2O, and dried under vacuum overnight to yield a light brown resin. The loading of the resin **M1B** was determined by cleavage of an aliquot (176mg) of the resin with 90% aq. TFA. Loading: 0.48 mmol/g.

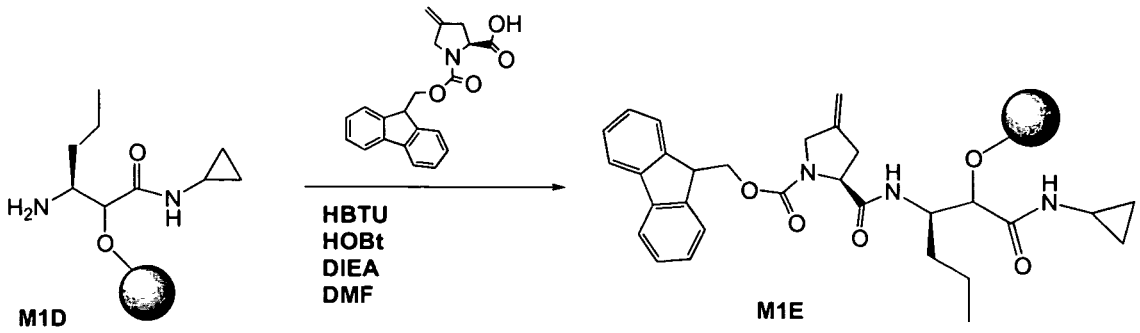
Preparation of (9H-fluoren-9-yl)methyl 2-(1-(cyclopropylamino)-2-hydroxy-1-oxohexan-3-ylcarbamoyl)-4-methylenepyrrolidine-1-carboxylate bound resin (M1E)

Step 1: 3-Amino-N-cyclopropyl-2-hydroxyhexanamide bound resin (M1D).



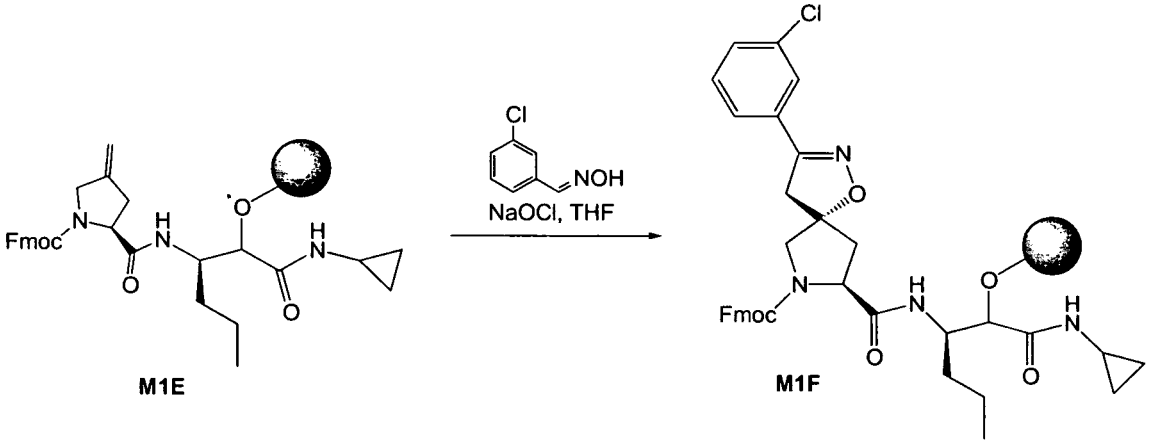
[0381] Allyl 1-(cyclopropylamino)-2-hydroxy-1-oxohexan-3-ylcarbamate bound resin **M1B** (30 g, 1.0eq.) was swollen with DCM. 1,3-Dimethylbarbituric acid (24.17 g, 12 eq.) and tetrakis(triphenylphosphine)palladium (1.49 g, 0.1 eq.) were added and the mixture shaken overnight. The mixture was filtered and washed with DMF and DCM to yield the resin **M1D**.

Step 2: (9H-Fluoren-9-yl)methyl 2-(1-(cyclopropylamino)-2-hydroxy-1-oxohexan-3-ylcarbamoyl)-4-methylenepyrrolidine-1-carboxylate bound resin (M1E).



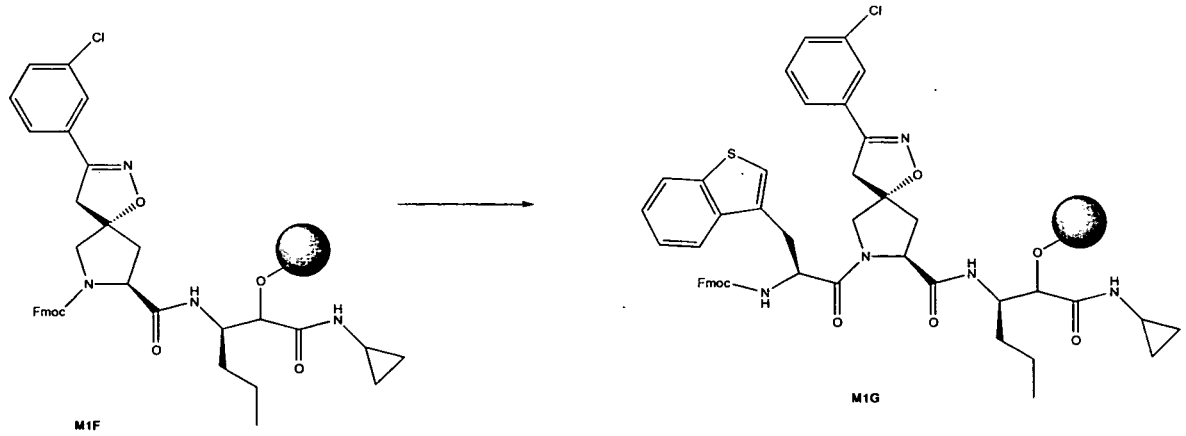
[0382] Resin **M1D** (1.0 g, 1.0 eq.) was stirred in DMF with FMOC-4-exomethyleneproline carboxylic acid (248 mg, 1.1 eq.), HBTU (4.8 mL of 0.5 M DMF solution, 5.0 eq.), HOBt (2.4 mL of 1.0 M DMF solution, 5.0 eq.), DIEA (836 uL, 10.0 eq.) for 3 hours. The resulting mixture was drained and washed with DMF (thrice) and DCM (thrice) to give title compound **M1E**.

Preparation of Fmoc-protected isoxazoline compound bound resin (M1F).



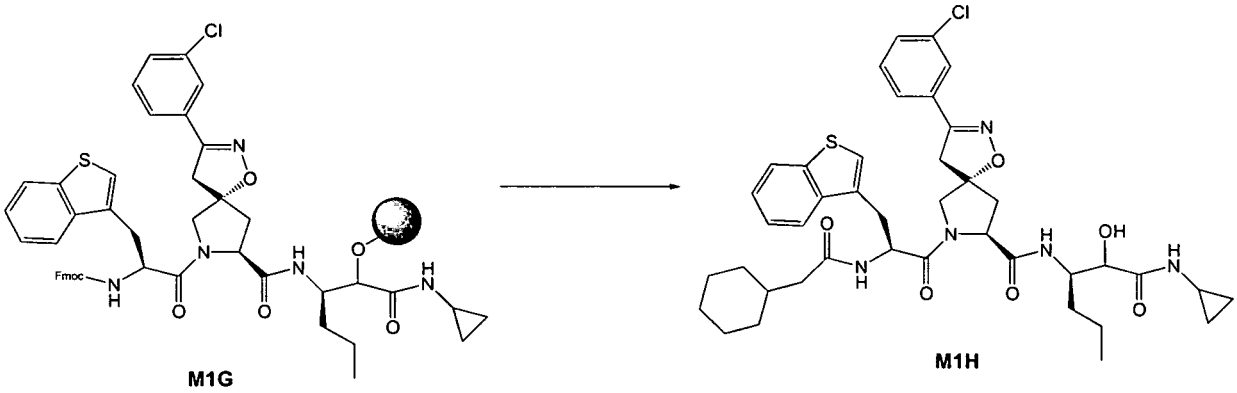
[0383] The resin **M1E** (2 g, 0.94 mmol) in THF was shaken with 3-chlorobenzaldoxime (5 eq.) and bleach (5% NaOCl) (15 eq.) for 18 hours. The resin was then filtered and washed with water, DMF, and DCM to yield the resin compound **M1F**. An aliquot of the resin was cleaved to provide a sample for LC-mass analysis (M+1 = 671).

Preparation of Fmoc Protected Isoxazoline bound resin compound (M1G)



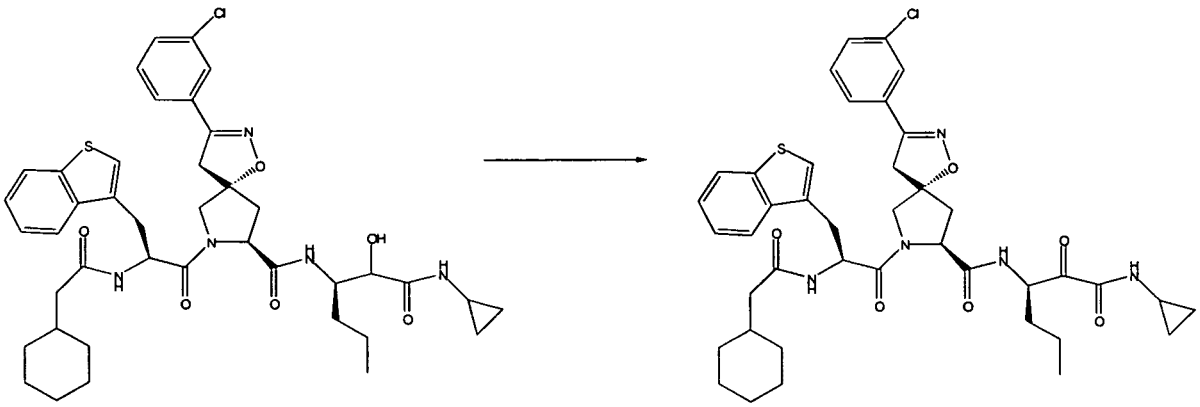
[0384] The resin **M1F** was shaken in 20% piperidine/DMF for 10 minutes, filtered, and washed with DMF and DCM. The THP resin bound spiroisoxazoline proline (0.14 mmol, 0.3g) was mixed with FMOC-L-3-benzothienyl-ALA(0.56 mmol, 0.25g), HOBT (0.56 mmol, 0.075g), N,N-diisopropylethylamine (0.56 mmol, 0.072g), HBTU (0.56 mmol, 0.21g) in DMF 2.3 mL and was agitated for 48 hours. The resin was filtered and washed with DMF, dichloromethane, and ether to yield the resin compound **M1G**.

. **Preparation of 7-((S)-3-(benzo[b]thiophen-3-yl)-2-(2-cyclohexylacetamido)propanoyl)-3-(3-chlorophenyl)-N-((3R)-1-(cyclopropylamino)-2-hydroxy-1-oxohexan-3-yl)-1-oxa-2,7-diazaspiro[4.4]non-2-ene-8-carboxamide (M1H)**



[0385] To the THP-resin bound FMOC protected spiroisoxazoline **M1G** was added 20% piperidine in DMF (3 mL). The mixture was agitated for 1hour, filtered, and washed with DMF and dichloromethane. The resin was them mixed with cyclohexylacetic acid (0.56 mmol, 80mg), HOBT (0.56 mmol, 0.075g), N,N-diisopropylethylamine (0.56 mmol, 0.072g), HBTU (0.56 mmol, 0.21g) in DMF 2.3 mL and was agitated for 48 hr. The resin was filtered and washed with DMF, dichloromethane, and ether. The resin obtained was then mixed with a solution of (50:45:5) trifluoroacetic acid, dichloromethane, and triisopropyl silane (3 mL) and was agitated overnight. The reaction was filtered and washed with dichloromethane. The filtrate was concentrated under vacuum and purified via silica gel chromatography using a gradient of 40% ethyl acetate/60% dichloromethane to 100% ethyl acetate to produce the alcohol **M1H**.

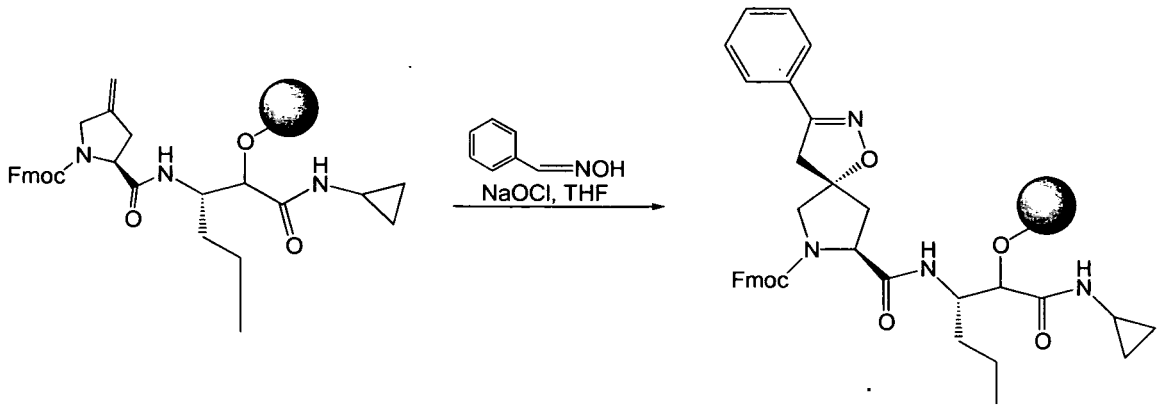
Example 1: Compound No. 336



M1H **Compound No. 336**

[0386] To a solution of the hydroxyamide **M1H** (14 mg, 0.018 mmol) in 0.38 mL of ethyl acetate was added EDC (35 mg, 0.18 mmol) followed by DMSO (0.070 mL). The mixture was cooled in an ice bath and dichloroacetic acid (15 mg, 0.12 mmol) in ethyl acetate (0.15 mL) was added. The reaction was warmed to room temperature and allowed to stir for 15 minutes and then cooled in an ice bath and quenched with 1.0 N HCl (0.21 mL). The solution was partitioned between ethyl acetate and water. The organic phase was washed with water and dried over sodium sulfate and evaporated solvent under vacuum. The resulting residue was purified by chromatography over silica gel using ethyl acetate and hexanes (3:1) as eluant to give Compound No. **336** as a white solid.

Preparation of (9H-fluoren-9-yl)methyl 8-((3S)-1-(cyclopropylamino)-2-hydroxy-1-oxohexan-3-ylcarbamoyl)-3-phenyl-1-oxa-2,7-diazaspiro[4.4]non-2-ene-7-carboxylate (M1N).

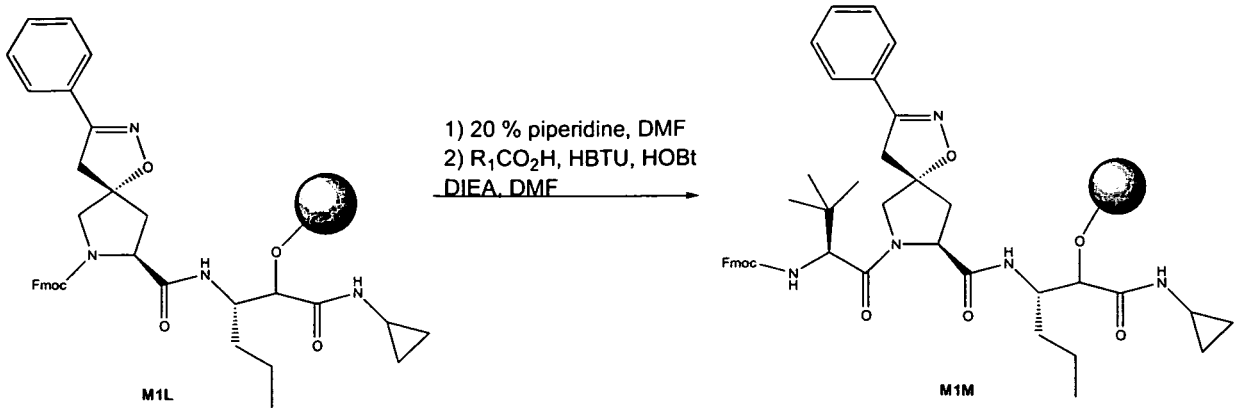


M1E **M1L**

Step 1: Fmoc Protected Phenyl-Substituted Isoxazoline bound resin (M1L).

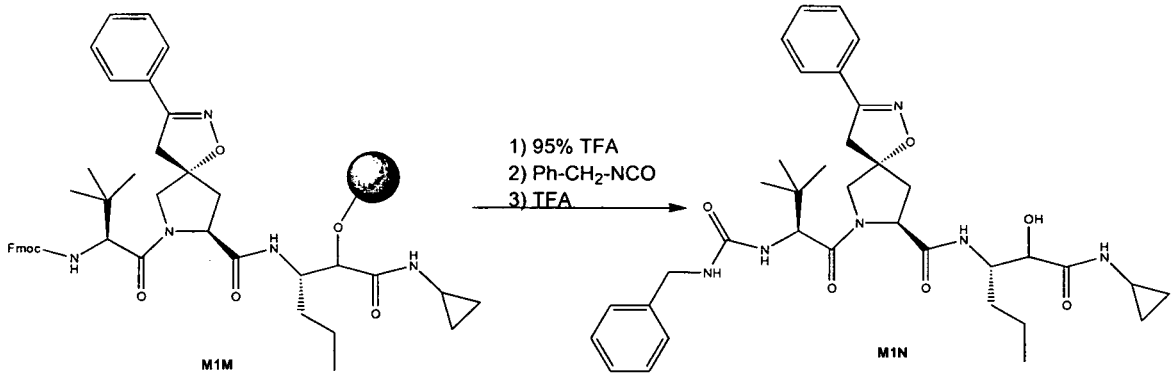
[0387] The resin **M1E** (2 g, 0.94 mmol) in THF was shaken with the oxime (5eq.) and bleach (5% NaOCl) (15 eq.) for 18 hours. The resin was then filtered and washed with water,

DMF, and DCM to give the Fmoc protected phenyl-substituted isoxazoline bound resin **M1L**. An aliquot of resin was cleaved for LC-mass analysis (M+1 = 637).



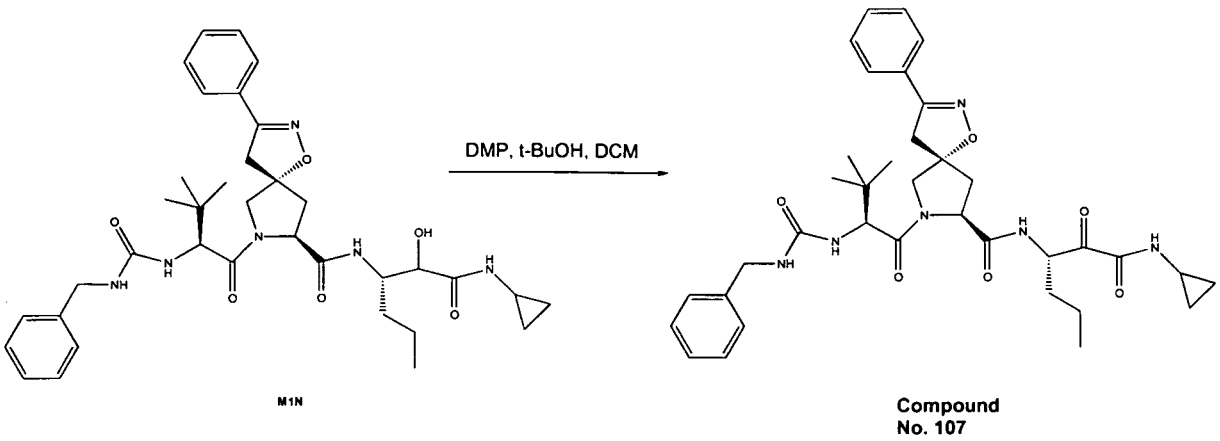
[0388] The resin **M1L** (0.47 mmol) was shaken in 20% piperidine/DMF for 10 minutes, and then filtered and washed with DMF and DCM. The resulting resin was shaken overnight with a solution of Fmoc-tBG-OH (480 mg 3.0 eq.), HOBT (2.82 mL of 0.5 M in DMF, 3.0 eq.), HBTU (2.82 mL of 0.5 M in DMF, 3.0 eq.), and DIEA (0.493 mL, 6.0 eq.). The resin was then filtered and washed with DMF and DCM to give the resin compound **M1M**, which was used in next reaction without further purification.

Step 2: Compound M1N



[0389] The resin **M1M** (0.47 mmol) was shaken in 20% piperidine/DMF for 10 minutes. The resin was filtered, washed with DMF and DCM. The resulting resin (140 mg, 0.065 mmol) was shaken overnight with benzylisocyanate (176 mg 20.0 eq.), then filtered and washed with DMF and DCM. The resin was shaken with 90% TFA in water for 30 min. The resulting solution was concentrated in vacuo to give the compound **M1N** (0.065 mmol), (M+1) 661, which was used in next reaction without further purification.

Example 2: Compound No. 107

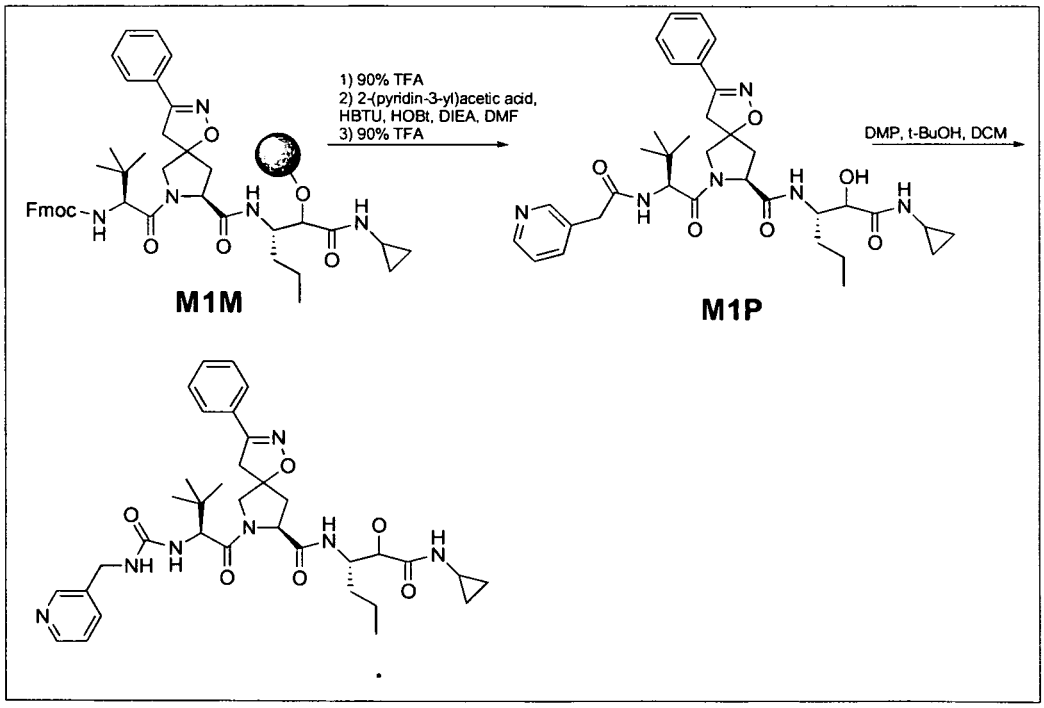


M1N

Compound No. 107

[0390] A solution of amide compound **M1N** in DCM (3 mL) was stirred with Dess-Martin Periodinane (54 mg, 2 eq.) and t-BuOH (54 uL) for 1 hour, and then sodium thiosulfate was added to the mixture. The product was extracted with EtOAc and the combined organic layer was then washed with water, $NaHCO_3$, brine and concentrated in vacuo and purified by Gilson Prep HPLC to afford Compound No. **107**. (M+1) 659.

Example 3: Compound No. 283



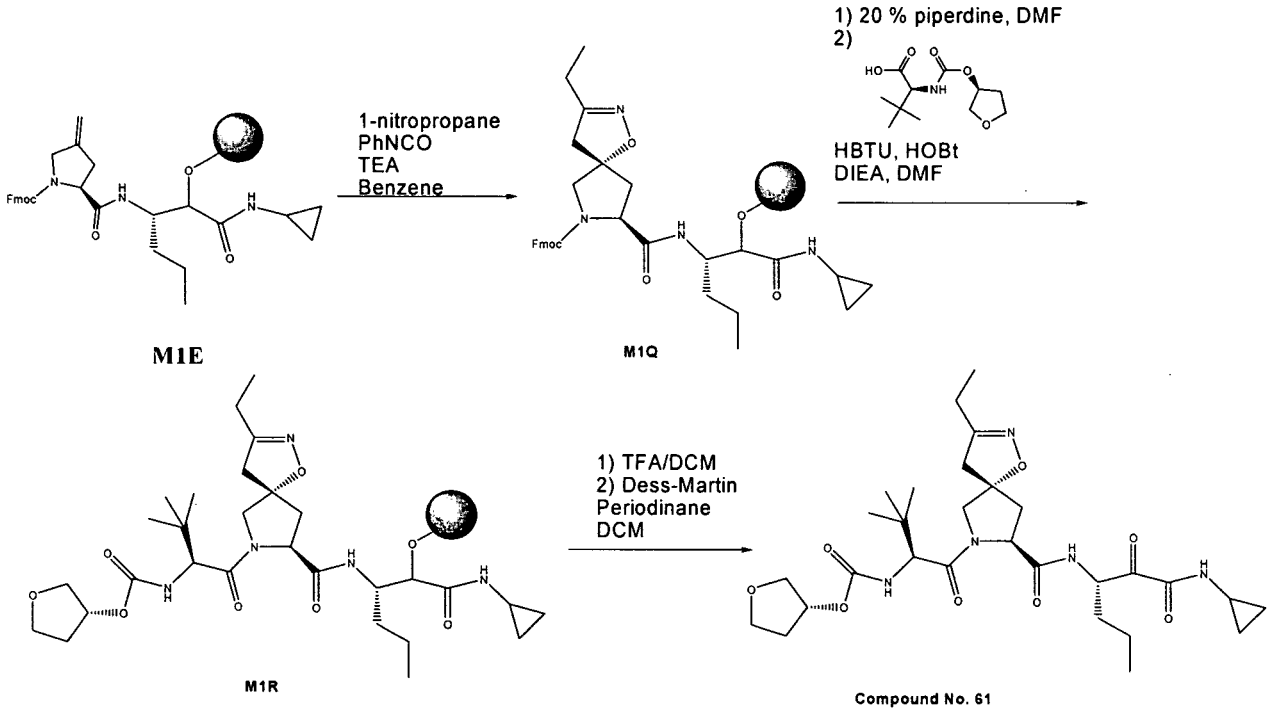
Compound 283

[0391] The THP resin **M1M** (0.065 mmol) was shaken in 20% piperidine/DMF for 10 minutes, and then filtered and washed with DMF and DCM. The resulting resin was shaken overnight with a solution of 2-(pyridin-3-yl)acetic acid (0.25 mmol 3.0 eq.), HOBT (0.5 mL of 0.5 M in DMF, 3.85 eq.), HBTU (0.5 mL of 0.5 M in DMF, 3.85 eq.), and DIEA (0.5 mmol, 7.69 eq.). The resin was then filtered and washed with DMF and DCM and was

shaken with 90% TFA in water for 30 minutes. The resulting solution was concentrated in vacuo to give the hydroxyl amide compound **M1P** (0.065 mmol) which was used in the next reaction without further purification. (M+1) 647.

[0392] A solution of the hydroxyl amide **M1P** (0.065 mmol) in DCM (3 mL) was stirred with Dess-Martin Periodinane (41 mg, 1.5 eq.) and t-BuOH (41 uL). After stirred for 1 hour, sodium thiosulfate was added to above mixture. The product was extracted with EtOAc. The combined organic layer was then washed with water, $NaHCO_3$, brine and concentrated in vacuo and purified by Gilson Prep HPLC to afford Compound No. **283** (4 mg). (M+1) 645.

Example 4: Compound No. 61



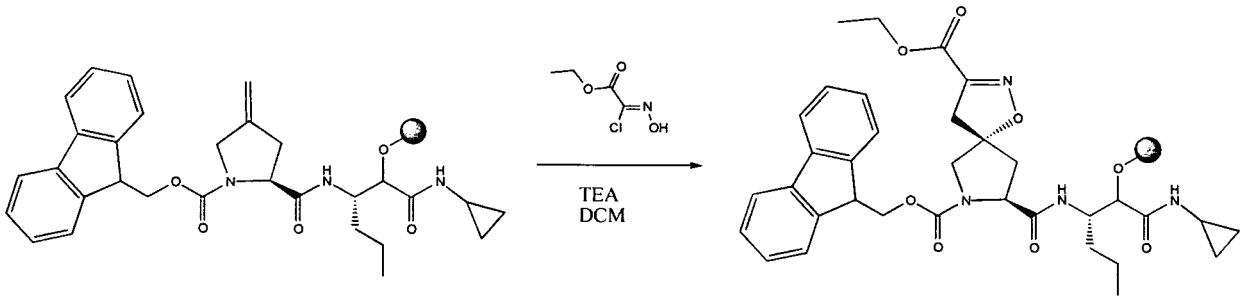
[0393] Compound **M1E** (750 mg, 1.0 eq.) was stirred in benzene with 1-nitropropane (315 uL, 10.0 eq.), and phenylisocyanate (385 uL, 10.0 eq.). Triethylamine (5 uL) was added, and the resulting mixture was shaken overnight, drained, and washed with DMF (thrice) and DCM (thrice). This process was repeated to yield compound **M1Q**. (M+H=589.0)

[0394] Compound **M1Q** (750 mg, 1.0 eq.) was then shaken in 20% piperidine/DMF for 10 minutes. The resin was filtered and washed with DMF (thrice) followed by DCM (thrice). This process was repeated. The resulting resin was shaken overnight with a solution of (S)-3,3-dimethyl-2-(((S)-tetrahydrofuran-3-yloxy)carbonylamino)butanoic acid (216 mg, 2.5 eq.), HBTU (1.76 mL of 0.5 M in DMF, 3.0 eq.), HOBt (0.88 mL of 1.0 M in DMF, 2.5 eq.), and

DIEA (307 uL, 5.0 eq.) in DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice) to give compound **M1R**. (M+H=593.9)

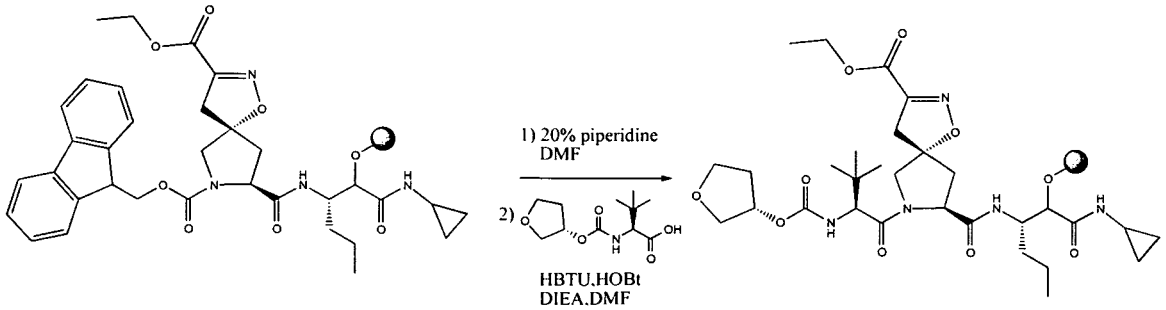
[0395] Compound **M1R** (750 mg, 1.0 eq.) stirred in 1/1 TFA/DCM for 3 hours. The resin was drained and washed with DCM (thrice). All of the organics were concentrated and DCM was added followed by Dess-Martin Periodinane (50 mg, 3.0 eq.). The resulting mixture was stirred for 1 hour, 1 N $Na_2S_2O_3$ was added, and stirred again. A racemic mixture of Compound No. 61 was purified by silica gel chromatography (10-90% ethyl acetate/hexanes gradient) to yield Compound No. **61** as one diastereomer. (M+H=591.8) ^{1}H-NMR (500 MHz, $CDCl_3$): 7.12 (d, 1H), 6.91 (d, 1H), 5.48 (d, 1H), 5.34 (td, 1H), 5.24 (s, 1H), 4.69 (t, 1H), 4.28 (d, 1H), 4.13 (s, 2H), 3.93-3.82 (m, 4H), 3.60 (d, 1 H), 3.06 (s, 0.5H), 3.03 (s, 0.5H), 2.95 (d, 1H), 2.90 (d, 1H), 2.78 (td, 1H), 2.51-2.47 (m, 1H), 2.44-2.34 (m, 3H), 2.14-2.10 (m, 1H), 1.94-1.88 (m, 1H), 1.63-1.57 (m, 1H), 1.46-1.36 (m, 2H), 1.17 (t, 3H), 0.98 (s, 9H), 0.95-0.83 (m, 5H), 0.59 (dd,2 H)

Example 5: Compound No. 146



M1E **M1S**

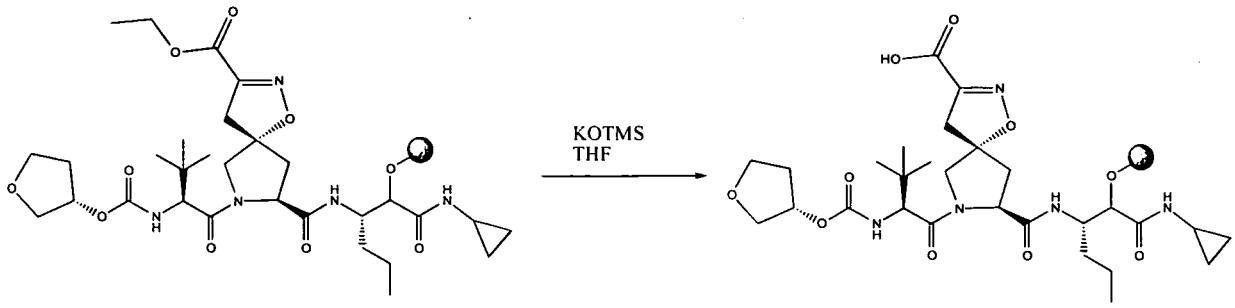
[0396] Compound **M1E** (50 mg, 1.0 eq.) was stirred in DCM with (Z)-ethyl 2-chloro-2-(hydroxyimino)acetate (7.1 mg, 2.0 eq.). To this mixture was slowly added TEA (6.6 uL, 2.0 eq.) in DCM and the mixture was shaken for 3 hours, then drained and washed with DMF (thrice) and DCM (thrice). This process was repeated to give compound **M1S** (M+H=632.4).



M1S **M1T**

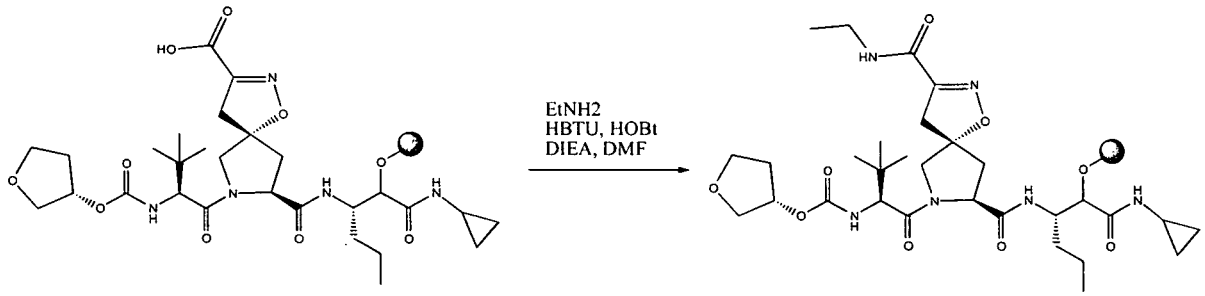
[0397] Compound **M1S** (1.0 g, 1.0 eq.) was shaken in 20% piperidine/DMF for 10 minutes. The resin was filtered and washed with DMF (thrice) followed by DCM (thrice).

This process was repeated. The resulting resin was shaken overnight with a solution of (S)-3,3-dimethyl-2-(((S)tetrahydrofuran-3-yloxy)carbonylamino)butanoic acid (230 mg 2.0 eq.), HBTU (1.88 mL of 0.5 M in DMF, 2.0 eq.), HOBt (0.94 mL of 1.0 M in DMF, 2.0 eq.), and DIEA (327 uL, 4.0 eq.) in 2mL DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice) to give compound **M1T** (M+H=638.0).



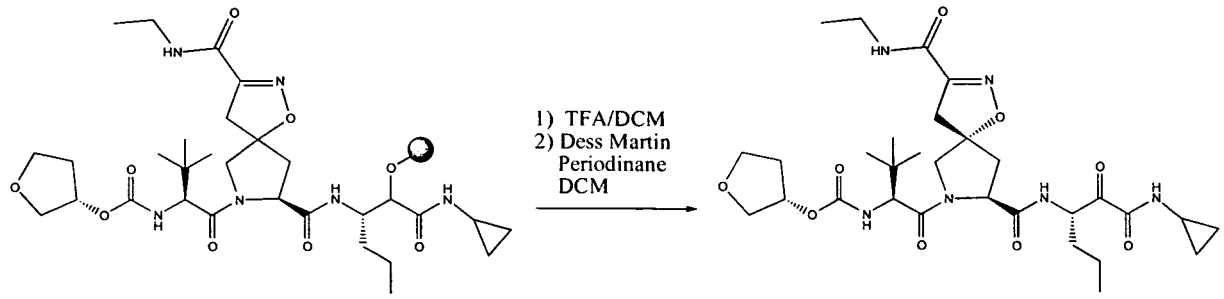
M1T **M1U**

[0398] Compound **M1T** (750 mg, 1.0 eq.) was shaken in THF with KOTMS (133 mg, 3.0 eq.) for 3 hours. The mixture was then drained and washed with THF/water (1/1), THF, DMF, and DCM (thrice each) to give compound **M1U**. (M+H=609.5).



M1U **M1V**

[0399] Compound **M1U** (250 mg, 1.0 eq.) was shaken overnight with a solution of ethylamine (22 mg 3.0 eq.), HBTU (0.54 mL of 0.5 M in DMF, 3.0 eq.), HOBt (0.27 mL of 1.0 M in DMF, 3.0 eq.), and DIEA (47 uL, 3.0 eq.) in DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice) to give compound **M1V.** (M+H=637.2).



M1V **Compound No. 146**

[0400] Compound **M1V** (0.4 g, 1.0 eq.) was stirred in 1/1 TFA/DCM for 2 hours and then drained and washed with DCM (thrice). The organic phases were combined and dried, and to it was added DCM followed by Dess-Martin Periodinane (97 mg, 3.0 eq.). The solution was stirred for 1 hour and to it was added 1 N $Na_2S_2O_3$ and the mixture was further stirred. The solution was purified by silica gel chromatography (10-90% ethyl acetate/hexanes gradient) to yield 6.1 mg of Compound No. **146**. (M+H=635.0) ^{1}H-NMR ($CDCl_3$): 5.5-5.2 (m, 2H), 5.1-5.0 (m, 1H), 4.9-4.7 (m, 2H), 4.5-4.2 (m, 3H), 4.1 (m, 1H), 3.9-3.7 (m, 3H), 3.6-3.5 (m, 2H), 3.5-3.2 (m, 2H), 2.8-2.4 (m, 2H), 2.1 (m, 1H), 2.0-1.8 (m, 3H), 1.8-1.5 (m, 3H), 1.5-1.3 (m, 3H), 1.3-1.2 (m, 2H), 1.0 (s, 9H), 0.9 (t, 3H), 0,8 (m, 2H), 0.6 (m, 2H).

[0401] The following compounds of Formula I were also produced according to Method 1 and the preparations described thereunder.

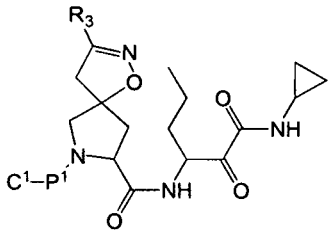


Table 2: Additional compounds of formula I produced by method 1.

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
7	N-FMOC-L-tert-butylglycine	3-(4-fluorophenyl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
12	N-FMOC-L-tert-butylglycine	Acetic acid	3-chlorobenzenecarbaldehyde oxime
14	N-FMOC-L-tert-butylglycine	cyclopentylmethanol	3-chlorobenzenecarbaldehyde oxime
24	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Fluorobenzenecarbaldehyde oxime
27	N-FMOC-L-tert-butylglycine	cyclobutylmethanol	3-chlorobenzenecarbaldehyde oxime
29	N-FMOC-L-tert-butylglycine	2-(1-methylcyclohexyl)acetic acid	3-chlorobenzenecarbaldehyde oxime
30	N-FMOC-L-tert-butylglycine	(S)-5-oxo-1-(thiophen-2-ylmethyl)pyrrolidine-2-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
33	N-FMOC-L-tert-butylglycine	cyclohexyl isocyanate	3-chlorobenzenecarbaldehyde oxime
34	N-FMOC-L-tert-butylglycine	5-hydroxypentan-2-one	3-chlorobenzenecarbaldehyde oxime
37	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
39	N-FMOC-L-tert-butylglycine	2-chlorobenzyl chloroformate	3-chlorobenzenecarbaldehyde oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
44	N-FMOC-L-tert-butylglycine	4-oxo-pentanoic acid	3-chlorobenzenecarbaldehyde oxime
53	N/A	2-(1-(2,6-dichlorobenzyl)piperidin-4-yl)acetic acid	Benzaldoxime
61	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Nitropropane
71	N-FMOC-L-tert-butylglycine	(R)-2,3-dihydrobenzo[b][1,4]dioxine-2-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
72	N-FMOC-L-tert-butylglycine	2-cyclopentylacetic acid	3-chlorobenzenecarbaldehyde oxime
75	N-FMOC-L-tert-butylglycine	2-(2,4-dimethylthiazol-5-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
76	N-FMOC-L-tert-butylglycine	Cyclopropyl isocyanate	3-chlorobenzenecarbaldehyde oxime
85	N-FMOC-L-tert-butylglycine	2-Fluoroethyl chloroformate	3-chlorobenzenecarbaldehyde oxime
92	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
93	N-FMOC-L-tert-butylglycine	cyclohexanecarboxylic acid	3-chlorobenzenecarbaldehyde oxime
94	N-FMOC-L-tert-butylglycine	2-aminoacetamide	3-chlorobenzenecarbaldehyde oxime
102	N-FMOC-L-tert-butylglycine	2-(3-fluoro-4-methylphenyl)acetic acid	3-chlorobenzenecarbaldehyde oxime
107	N-FMOC-L-tert-butylglycine	Benzyl isocyanate	Benzaldoxime
108	N-FMOC-L-tert-butylglycine	cis-4-methoxycyclohexanecarboxylic acid	3-chlorobenzenecarbaldehyde oxime
110	N-FMOC-L-tert-butylglycine	Benzyl chloroformate	3-Chloro-4,6-dimethoxybenzaldoxime
112	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	2-nitro-1-phenyl ethanone
118	N-FMOC-L-tert-butylglycine	2-(4-fluorophenyl)ethanol	3-Chlorobenzene carbaldehyde oxime
119	N-FMOC-L-tert-butylglycine	tert-Butyl isocyanate	2-nitro-1-phenyl ethanone
122	N-FMOC-L-tert-butylglycine	3-Fluorobenzyl isocyanate	3-Chlorobenzene carbaldehyde oxime
123	N-FMOC-L-tert-butylglycine	Ethyl isocyanate	3-Chlorobenzene carbaldehyde oxime
124	N-FMOC-O-Methyl-L-Threonine	2-cyclohexylacetic acid	3-Chlorobenzene carbaldehyde oxime
125	(2R,3S)-N-FMOC-2-Amino-3-phenyl-butyric acid	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
128	N-FMOC-L-tert-butylglycine	4-(1H-pyrrole-2,5-dione)phenyl isocyanate	3-chlorobenzenecarbaldehyde oxime
135	N-FMOC-L-tert-butylglycine	1-isopropyl-4-oxo-1,4-dihydroquinoline-3-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
139	N-FMOC-L-tert-butylglycine	(R)-2-hydroxy-2-phenylpropanoic acid	3-chlorobenzenecarbaldehyde oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
146	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2-Chloro-2-hydroximinoacetic aicd ethyl ester (chlorooxime)
152	N-FMOC-L-tert-butylglycine	(tetrahydrofuran-3-yl)methanol	3-chlorobenzenecarbaldehyde oxime
154	N-Alloc-L-tert-butylglycine	N/A	4-Fluorobenzenecarbaldehyde oxime
155	N-FMOC-L-tert-butylglycine	2-(5-fluoro-2-methylphenyl)acetic acid	3-chlorobenzenecarbaldehyde oxime
156	N-FMOC-L-tert-butylglycine	Isobutylamine	3-chlorobenzenecarbaldehyde oxime
159	N-FMOC-L-tert-butylglycine	2-(thiophen-3-yl)ethanol	3-chlorobenzenecarbaldehyde oxime
160	N-CBZ-L-tert-butylglycine	N/A	4-Fluoro benzene carbaldehyde oxime
161	N-FMOC-L-tert-butylglycine	5-acetamido-2-acetylthiophene-3-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
164	N-CBZ-L-tert-butylglycine	N/A	2-Chlorobenzenecarbaldehyde oxime
167	N-FMOC-L-tert-butylglycine	(2-methylpyridin-3-yl)methanol	3-chlorobenzenecarbaldehyde oxime
173	N-FMOC-L-tert-butylglycine	2,2-difluoroethylamine	3-chlorobenzenecarbaldehyde oxime
174	N-FMOC-L-tert-butylglycine	m-tolylmethanol	3-chlorobenzenecarbaldehyde oxime
180	N-FMOC-L-tert-butylglycine	Acetic acid	Nitroethane
183	N-CBZ-L-tert-butylglycine	N/A	3-Fluorobenzenecarbaldehyde oxime
185	N-FMOC-L-3-Thienyl-Alanine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
193	N-FMOC-L-tert-butylglycine	Isopropyl chloroformate	3-chlorobenzenecarbaldehyde oxime
199	N-CBZ-L-tert-butylglycine	N/A	9-Anthraldehyde oxime
201	N-FMOC-L-tert-butylglycine	(3-methoxyphenyl)methanol	3-chlorobenzenecarbaldehyde oxime
203	N-FMOC-L-tert-butylglycine	(3,5-difluorophenyl)methanol	3-chlorobenzenecarbaldehyde oxime
205	N-FMOC-L-tert-butylglycine	Benzyl isocyanate	3-chlorobenzenecarbaldehyde oxime
207	N-FMOC-L-Glycine	Ethyl chloroformate	3-chlorobenzenecarbaldehyde oxime
208	N-CBZ-L-tert-butylglycine	N/A	2-Naphthaldehyde oxime
209	N-FMOC-L-tert-butylglycine	(3-fluorophenyl)methanol	3-chlorobenzenecarbaldehyde oxime
210	N-FMOC-L-tert-butylglycine	2-chlorobenzyl chloroformate	3-Chloro-4,6-dimethoxybenzaldoxime
213	N-FMOC-4-Methoxy-L-Phenylalanine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
216	N-FMOC-L-tert-butylglycine	5-oxo-1-(thiophen-2-ylmethyl)pyrrolidine-3-carboxylic acid	3-chlorobenzenecarbaldehyde oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
235	N-CBZ-L-tert-butylglycine	N/A	nitrobutane
237	N-FMOC-L-tert-butylglycine	3-(2-methyl-1H-imidazol-1-yl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
241	N-FMOC-L-tert-butylglycine	(S)-1-isopropyl-5-oxopyrrolidine-2-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
242	N-FMOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	Piperonal oxime
243	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	nitrobutane
249	N-FMOC-L-tert-butylglycine	cyclohexanemethyl isocyanate	Benzaldoxime
254	N-FMOC-L-tert-butylglycine	1-(thiophen-2-yl)propan-2-ol	3-chlorobenzenecarbaldehyde oxime
259	N-FMOC-L-tert-butylglycine	3,4,5-trimethoxybenzyl isocyanate	3-chlorobenzenecarbaldehyde oxime
260	N-FMOC-L-tert-butylglycine	2-methoxyethyl chloroformate	3-chlorobenzenecarbaldehyde oxime
261	N-FMOC-L-tert-butylglycine	benzyl 4-isocyanatopiperidine-1-carboxylate	3-chlorobenzenecarbaldehyde oxime
262	N/A	4-nitrophenyl choroformate	3-chlorobenzenecarbaldehyde oxime
276	2-((3S,4aS,8aS)-3-(tert-butylcarbamoyl)octahydroisoquinolin-2(1H)-yl)acetic acid	N/A	3-chlorobenzenecarbaldehyde oxime
278	N-FMOC-L-tert-butylglycine	benzyl chloroformate	2-(4-Methoxyphenoxy)benzenecarbaldehyde oxime
283	N-FMOC-L-tert-butylglycine	2-(pyridin-3-yl)acetic acid	Benzaldoxime
287	N-FMOC-L-tert-butylglycine	2-(3-methoxyphenyl)acetic acid	3-chlorobenzenecarbaldehyde oxime
288	N-FMOC-L-tert-butylglycine	1-Naphthyl isocyanate	Benzaldoxime
289	N-FMOC-2-Trifluoromethyl-L-Phenylalanine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
291	N-FMOC-L-tert-butylglycine	spiro[indene-1,4'-piperidin]-3(2H)-one	3-chlorobenzenecarbaldehyde oxime
294	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
308	N-FMOC-L-tert-butylglycine	(tetrahydro-2H-pyran-2-yl)methanol	3-chlorobenzenecarbaldehyde oxime
311	N-FMOC-L-tert-butylglycine	2-(pyrrolidine-1-carbonyl)cyclohexanecarboxylic acid	3-chlorobenzenecarbaldehyde oxime
313	N/A	benzyl isocyanate	3-Chloro-4,6-dimethoxybenzaldoxime
317	N-FMOC-L-tert-butylglycine	2-(1-oxoisoindolin-2-yl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
324	N-FMOC-L-tert-butylglycine	(R)-3-(1-cyanoethyl)benzoic acid	3-chlorobenzenecarbaldehyde oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
329	N-FMOC-L-tert-butylglycine	cyclohexylacetic acid	Nitropropane
331	N-FMOC-L-tert-butylglycine	acetic acid	4-Fluorobenzene carbaldehyde oxime
333	N-FMOC-L-tert-butylglycine	(S)-1-methylbenzylamine	3-Chlorobenzene carbaldehyde oxime
334	N-FMOC-L-tert-butylglycine	(S)-2-methyl-3-phenylpropanoic acid	3-chlorobenzenecarbaldehyde oxime
336	N-FMOC-L-3-Benzothienyl-Alanine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
338	N-FMOC-2-Fluoro-L-Phenylalanine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
340	N-CBZ-L-tert-butylglycine	N/A	4-Phenylbenzenecarbaldehyde oxime
341	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2-chloro-2-hydroximinoacetic acid ethyl ester (chlorooxime) followed by ester hydrolysis and coupling of ethylamine
342	N/A	pyridine 3-methanol	3-chlorobenzenecarbaldehyde oxime
345	N-(5-methyl-3-nitroprydinyl)-L-tert-butylglycine	N/A	3-chlorobenzenecarbaldehyde oxime
349	N-FMOC-L-tert-butylglycine	2-(4-fluorophenyl)ethanol	3-chlorobenzenecarbaldehyde oxime
352	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Pyridine-4-aldoxime
357	N-FMOC-L-tert-butylglycine	pyridin-4-ylmethanol	3-chlorobenzenecarbaldehyde oxime
358	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Trifluoromethoxybenzenecarbaldehyde oxime
365	N-CBZ-L-tert-butylglycine	N/A	4-Trifluoromethoxybenzenecarbaldehyde oxime
367	N/A	3,4,5-trimethoxybenzyl isocyanate	Benzaldoxime
373	N-FMOC-L-tert-butylglycine	3-(pyridin-2-yl)propan-1-ol	3-chlorobenzenecarbaldehyde oxime
374	N-FMOC-L-tert-butylglycine	tetrahydro-2H-pyran-4-ol	3-chlorobenzenecarbaldehyde oxime
377	N-FMOC-L-tert-butylglycine	(S)-1-(3-chlorobenzyl)-5-oxopyrrolidine-2-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
378	N-FMOC-L-tert-butylglycine	pyridin-2-ylmethanol	3-chlorobenzenecarbaldehyde oxime
379	N-FMOC-L-tert-butylglycine	isopropyl isocyanate	3-chlorobenzenecarbaldehyde oxime
381	N-FMOC-L-tert-butylglycine	4-oxo-3,4-dihydrophthalazine-1-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
383	N-CBZ-L-tert-butylglycine	N/A	Nitropropane
387	N-FMOC-L-tert-butylglycine	(3R,3aS,6aR)-hexahydrofuro[2,3-b]furan-3-ol	3-chlorobenzenecarbaldehyde oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
389	N-FMOC-L-tert-butylglycine	3-(pyridin-3-yl)propan-1-ol	3-chlorobenzenecarbaldehyde oxime
390	N-FMOC-L-tert-butylglycine	cyclohexylacetic acid	2-nitro-1-phenyl ethanone
398	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Fluorobenzenecarbaldehyde oxime
400	N-FMOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	nitrobutane
402	N-FMOC-L-tert-butylglycine	2-(5-oxo-2-(thiophen-2-yl)cyclopent-1-enyl)acetic acid	3-chlorobenzenecarbaldehyde oxime
407	N-FMOC-L-tert-butylglycine	ethyl chloroformate	3-chlorobenzenecarbaldehyde oxime
417	N-FMOC-L-tert-butylglycine	N-FMOC-L-tert-butylglycine followed by 2-pyrazine carboxylic acid	4-Fluorobenzenecarbaldehyde oxime
427	N-FMOC-L-Phenylalanine	ethyl chloroformate	3-chlorobenzenecarbaldehyde oxime
431	N-FMOC-L-tert-butylglycine	2-o-tolylacetic acid	3-chlorobenzenecarbaldehyde oxime
432	N-FMOC-L-tert-butylglycine	N-methyl ethylamine	3-chlorobenzenecarbaldehyde oxime
437	N-FMOC-S-tert-Butyl-L-Cysteine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
450	N-FMOC-L-tert-butylglycine	N-FMOC-L-tert-butylglycine followed by 2-pyrazine carboxylic acid	Piperonal oxime
454	N-FMOC-L-tert-butylglycine	2-(quinolin-8-ylthio)acetic acid	3-chlorobenzenecarbaldehyde oxime
459	N-FMOC-L-Norleucine	2-cyclohexylacetic acid	3-chlorobenzenecarbaldehyde oxime
462	N-FMOC-L-tert-butylglycine	cyclohexylacetic acid	Piperonal oxime
463	N-FMOC-L-tert-butylglycine	2-phenylethanol	3-chlorobenzenecarbaldehyde oxime
465	N-(Cyclopentylformoyl)-L-tert-butylglycine	N/A	4-Fluorobenzenecarbaldehyde oxime
467	N-FMOC-L-tert-butylglycine	2-(bicyclo[2.2.1]heptan-2-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
471	N-FMOC-L-tert-butylglycine	p-tolylmethanol	3-chlorobenzenecarbaldehyde oxime
474	N-FMOC-L-tert-butylglycine	2-methyl-3-(3-methyl-1H-pyrazol-1-yl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
477	N-FMOC-L-tert-butylglycine	(S)-1-methoxy-3,3-dimethylbutan-2-amine	3-chlorobenzenecarbaldehyde oxime
484	N-FMOC-L-tert-butylglycine	Succinic Anhydride	3-chlorobenzenecarbaldehyde oxime
487	N-FMOC-L-tert-butylglycine	2-(6-methoxy-3-oxo-2,3-dihydro-1H-inden-1-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
487	N-FMOC-L-tert-butylglycine	2-(3-oxo-2,3-dihydro-1H-inden-1-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
492	N-FMOC-L-tert-butylglycine	tert-Butyl isocyanate	3-chlorobenzenecarbaldehyde oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
497	N-FMOC-L-tert-butylglycine	pyridin-3-ylmethylamine	3-chlorobenzenecarbaldehyde oxime
503	N-FMOC-L-tert-butylglycine	trans-4-methoxycyclohexanecarboxylic acid	3-chlorobenzenecarbaldehyde oxime
504	N-FMOC-L-tert-butylglycine	3-(pyridin-3-yl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
505	N-FMOC-L-tert-butylglycine	3-(2,5-dioxoimidazolidin-4-yl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
512	N-FMOC-L-tert-butylglycine	(2-fluorophenyl)methanol	3-chlorobenzenecarbaldehyde oxime
515	N-FMOC-L-tert-butylglycine	tetrahydro-2H-pyran-3-ol	3-chlorobenzenecarbaldehyde oxime
517	N-FMOC-L-tert-butylglycine	N/A	Nitroethane
518	N-FMOC-L-tert-butylglycine	(S)-1-(3-methylbenzyl)-5-oxopyrrolidine-2-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
520	N-FMOC-L-tert-butylglycine	benzyl chloroformate	Benzaldoxime
523	N-FMOC-L-tert-butylglycine	tetrahydro-2H-pyran-4-carboxylic acid	3-chlorobenzenecarbaldehyde oxime
526	N-FMOC-L-tert-butylglycine	benzyl chloroformate	3-chlorobenzenecarbaldehyde oxime
528	N-FMOC-L-tert-butylglycine	3-(1H-indazol-1-yl)propanoic acid	3-chlorobenzenecarbaldehyde oxime
532	N-FMOC-L-tert-butylglycine	3-methylbutanoic acid	3-chlorobenzenecarbaldehyde oxime
533	N/A	N/A	4-Fluorobenzenecarbaldehyde oxime
538	N-FMOC-L-tert-butylglycine	2-cyano-2-methyl-3-phenylpropanoic acid	3-chlorobenzenecarbaldehyde oxime
544	N-FMOC-L-tert-butylglycine	3-(1H-benzo[d]imidazol-1-yl)-2-methylpropanoic acid	3-chlorobenzenecarbaldehyde oxime
547	N-FMOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	Nitropropane
553	N-FMOC-L-tert-butylglycine	2-(2,6-dioxo-1,2,3,6-tetrahydropyrimidin-4-yl)acetic acid	3-chlorobenzenecarbaldehyde oxime
557	N-FMOC-L-tert-butylglycine	(1R,6S)-6-(methoxycarbonyl)cyclohex-3-enecarboxylic acid	3-chlorobenzenecarbaldehyde oxime
558	N-FMOC-L-tert-butylglycine	phenyl isocyanate	3-chlorobenzenecarbaldehyde oxime
559	N-FMOC-L-tert-butylglycine	tert-Butyl isocyanate	Nitropropane
561	N-FMOC-L-tert-butylglycine	(2,5-difluorophenyl)methanol	3-chlorobenzenecarbaldehyde oxime
563	N-FMOC-L-tert-butylglycine	Pyridine 3-methanol	3-chlorobenzenecarbaldehyde oxime
566	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	Nitropropane
576	N-FMOC-L-tert-butylglycine	3-pyridyl isocyanate	3-chlorobenzenecarbaldehyde oxime
580	N-FMOC-L-tert-butylglycine	ethyl isocyanate	Benzaldoxime

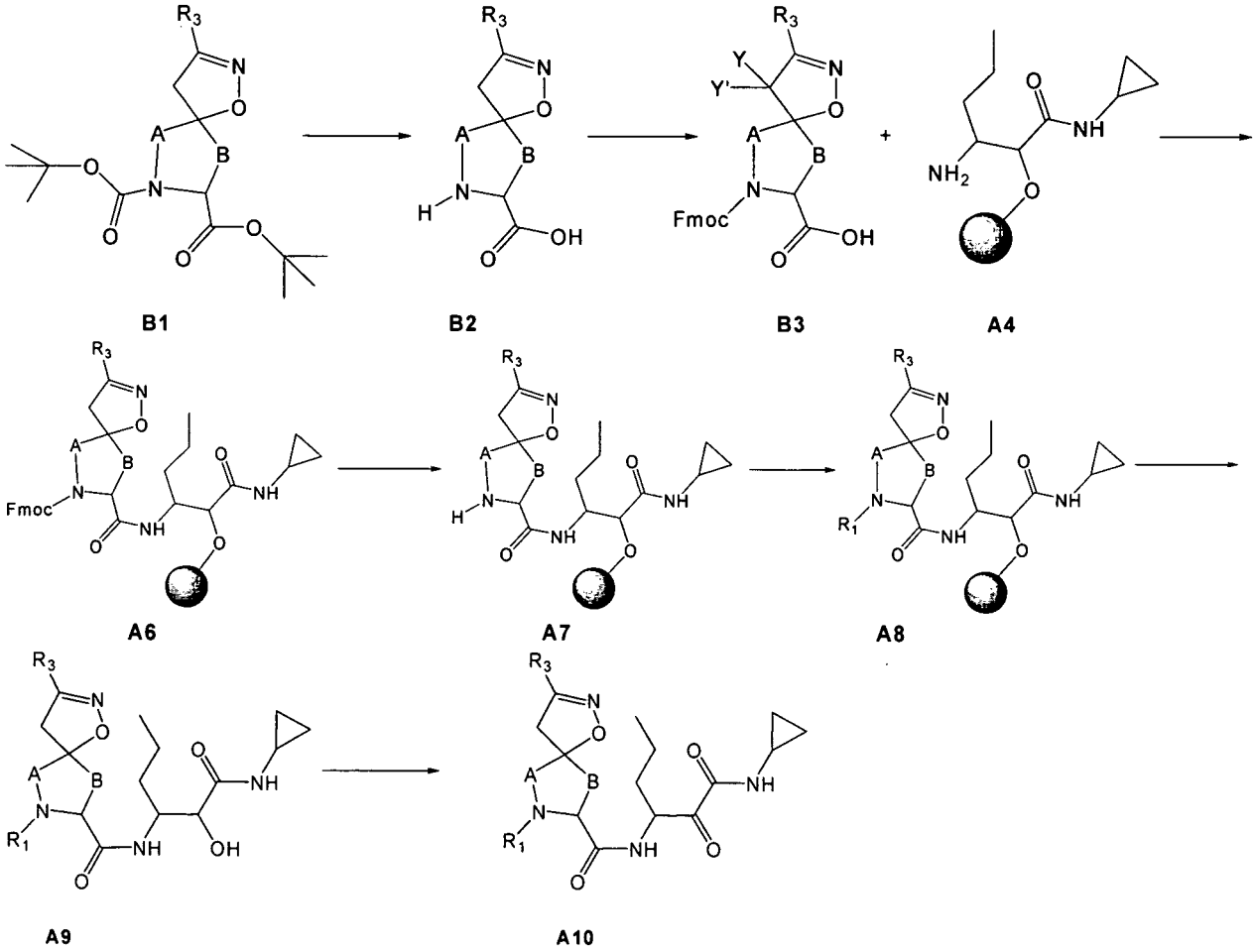
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
582	N-FMOC-L-tert-butylglycine	2-(thiophen-2-yl)ethanol	3-chlorobenzenecarbaldehyde oxime
583	N-FMOC-L-tert-butylglycine	benzyl isocyanate	3-chlorobenzenecarbaldehyde oxime

[0402] All starting materials for R_3 listed in Table 2 and all other tables herein were either commercially available (nitro or oxime) or readily prepared from corresponding aldehyde precursors.

[0403] Additionally, Compound Nos. 20, 22, 53, 81, 103, 116, 166, 187, 189, 194, 197, 200, 220, 223, 226, 245, 252, 271, 204, 307, 319, 339, 354, 360, 361, 371, 392, 393, 435, 449, 506, 514, 531, and 585 were also produced by using Method 1.

[0404] Certain other compounds of the invention may be prepared as illustrated by Method 2.

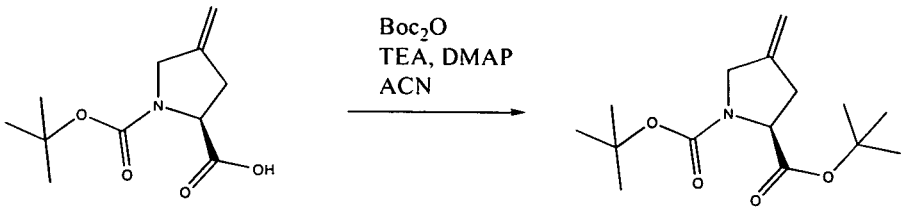
METHOD 2:



[0405] Referring to Method 2, the protected spiroisoxazoline **B1** is deprotected to **B2** which in turn is converted to the Fmoc derivative **B3**. Reaction of **B3** with the resin bound

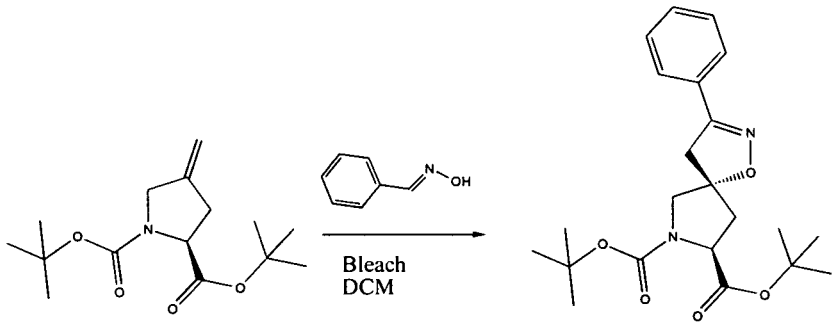
aminoalcohol **A4** provides the resin bound spiroisoxazoline **A6** which is converted to **A10** as described in Method 1.

Example 6: Compound No. 281



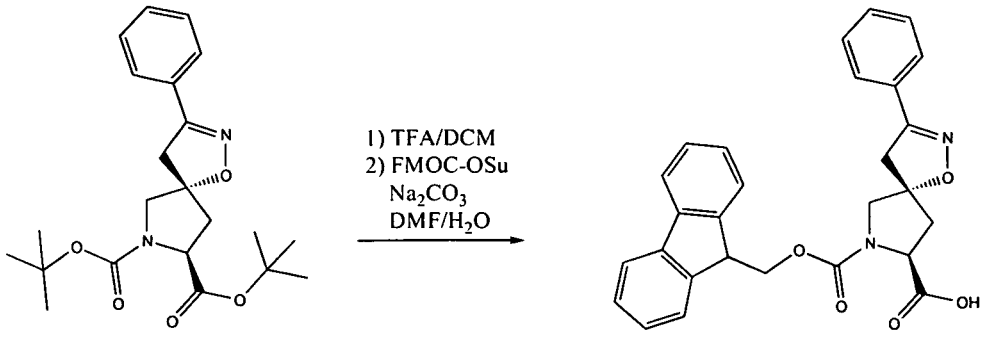
M2A **M2B**

[0406] Compound **M2A** (5.0 g, 1.0 eq.) was stirred in 100 mL acetonitrile and to this mixture was added ditertbutyldicarbonate (9.6 g, 2.0 eq.), dimethylaminopyridine (537 mg, 0.2 eq.), and triethylamine (6.13 mL, 2.0 eq.) and stirred overnight. The resulting mixture was concentrated, ethyl acetate was added, and the mixture was washed with 1.0 N HCl, dried over sodium sulfate, concentrated, and purified by silica gel chromatography (10-30% ethyl acetate/hexanes gradient) to yield compound **M2B**. (M+H=284.0) ^{1}H-NMR ($CDCl_3$): 5.0 (m, 2H), 4.3-4.5 (m, 1H), 4.0-4.1 (m, 2H), 2.9-3.0 (m, 1H), 2.5-2.6 (d, 1H), 1.5 (s, 3/9 of 18H), 1.4 (s, 6/9 of 18H).



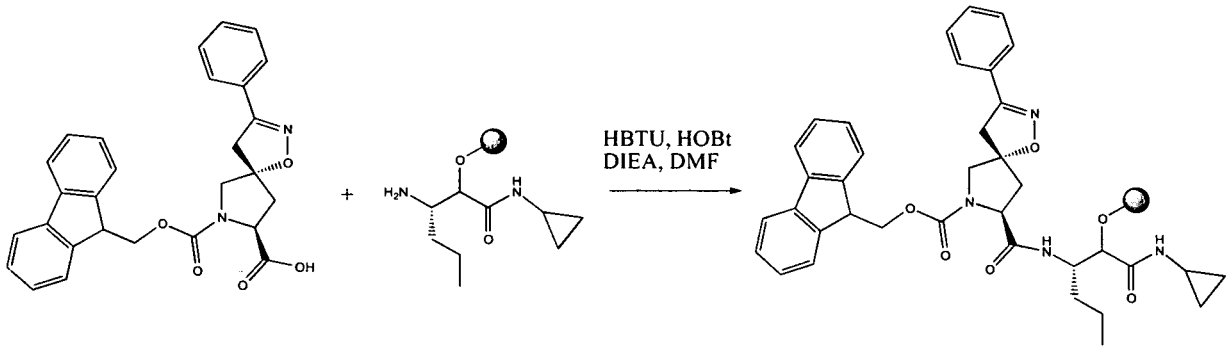
M2B **M2C**

[0407] Compound **M2B** (2.0 g, 1.0 eq.) stirred in 35 mL DCM with benzaldoxime (2.67 g, 2.0 eq.). The solution was cooled on an ice bath and to this bleach (5% NaOCl) (34.9 mL) was slowly added. The mixture was then warmed to room temperature and stirred for 2 hours. The aqueous layer was separated and extracted with DCM twice. The organics were combined and dried over magnesium sulfate, filtered and concentrated. Purified via silica gel chromatography (5-30% ethyl acetate/hexanes gradient) yielded compound **M2C**. (M+H=403.1) ^{1}H-NMR (500 MHz, CDCl3): 7.64-7.63 (m, 2H), 7.41-7.40 (m, 3H), 4.43-4.37 (t, 1H), 3.94-3.85 (dd, 1H), 3.62 (t, 1H), 3.44-3.38 (m, 1H), 3.29-3.24 (m, 1H), 2.74 (m, 1H), 2.14-2.10 (m, 1H), 1.49 (s, 9H), 1.46 (s, 9H).



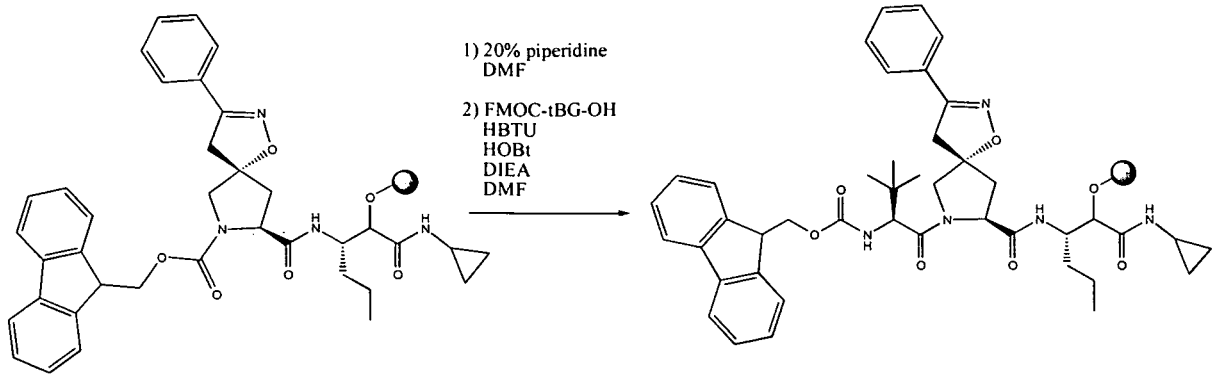
M2C **M2D**

[0408] Compound **M2C** was stirred in 1/1 TFA/DCM for 3 hours. The mixture was concentrated. To the concentrated mixture was added 17 mL DMF, 5 mL water, sodium carbonate (713 mg, 2.5 eq.), FMOC-OSu (951 mg, 1.05 eq.) and stirred 3 hours. Then, ethyl acetate was added and the resulting mixture was washed with 1.0 N HCl followed by brine. It was dried over magnesium sulfate, filtered and concentrated to yield compound M2D. (M+H=468.9).



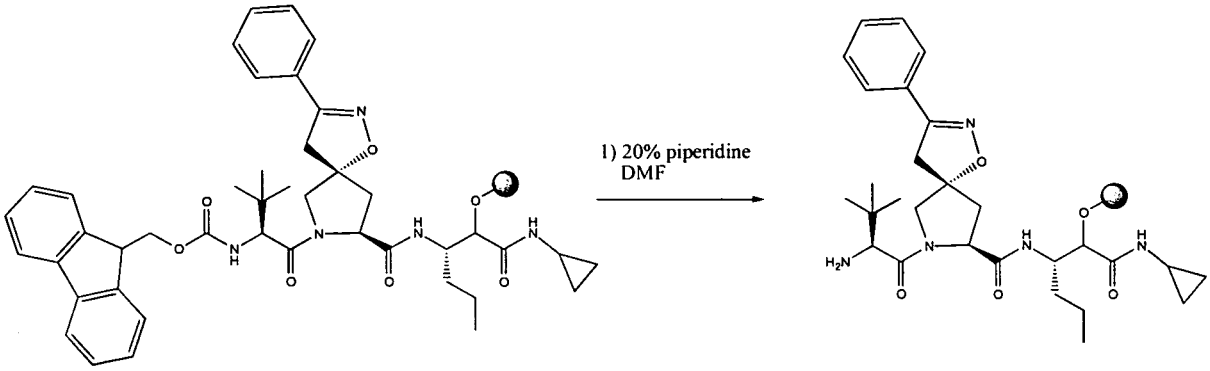
M2D **M1D** **M1L**

[0409] Compound **M2D** (1.26 g, 2.0 eq.) was stirred in DMF with **M1D** (2.5 g, 1.0 eq.), HBTU (12 mL of 0.5 M in DMF, 5.0 eq.), HOBt (6 mL of 1.0 M in DMF, 5.0 eq.), and Hünig's base (2.09 mL. 10.0 eq.) overnight. The mixture was drained and washed with DMF (thrice) and DCM (thrice) to yield compound **M1L**. (M+H=637.0).



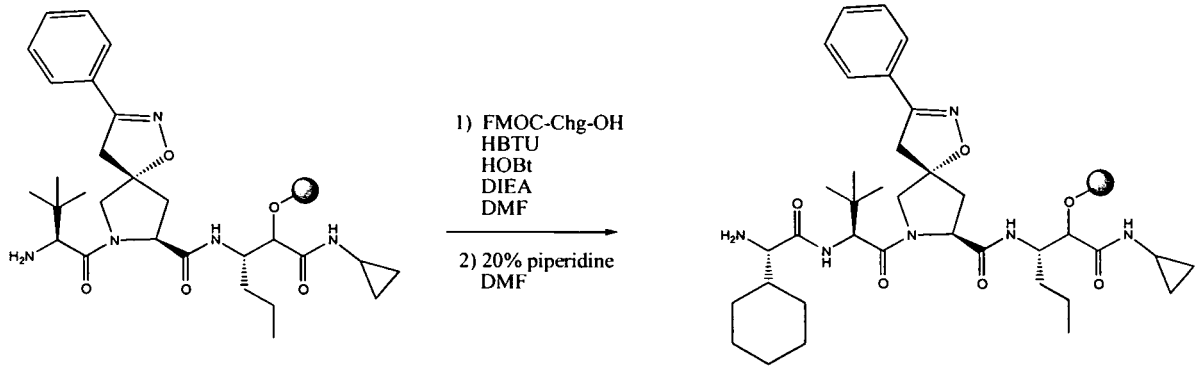
M1L **M1M**

[0410] Compound **M1L** (0.4 g, 1.0 eq.) was shaken in 20% piperidine/DMF for 10 minutes before being filtered and washed with DMF (thrice) followed by DCM (thrice). This process was repeated. The resulting resin was shaken overnight with a solution of FMOC-tert-butylglycine (200 mg 3.0 eq.), HBTU (1.15 mL of 0.5 M in DMF, 3.0 eq.), HOBt (0.58 mL of 1.0 M in DMF, 3.0 eq.), and DIEA (167 uL, 5.0 eq.) in 2 mL DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice) to give compound **M1M**. (M+H=750.1).



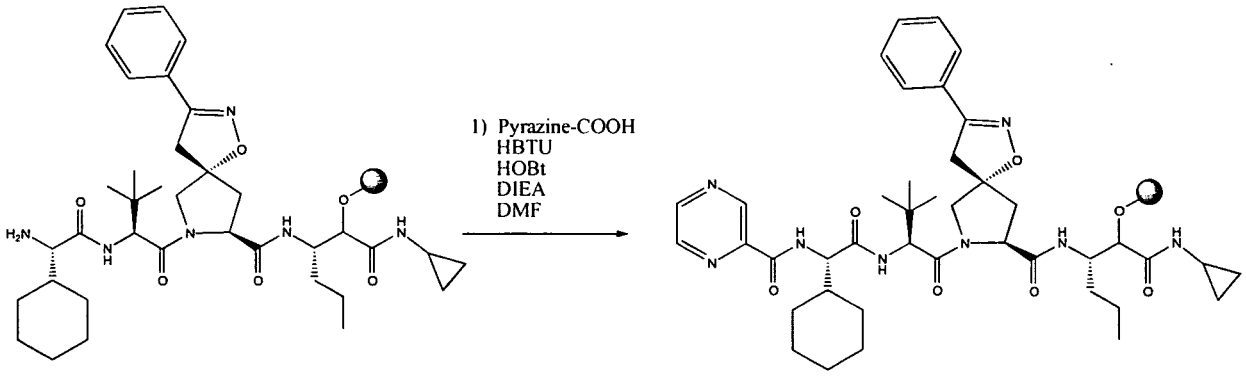
M1M **M2H**

[0411] Compound **M1M** (0.4 g, 1.0 eq.) was shaken in 20% piperidine/DMF for 10 minutes and the resin was filtered and washed with DMF (thrice) followed by DCM (thrice). This process was repeated to give Compound **M2H**.



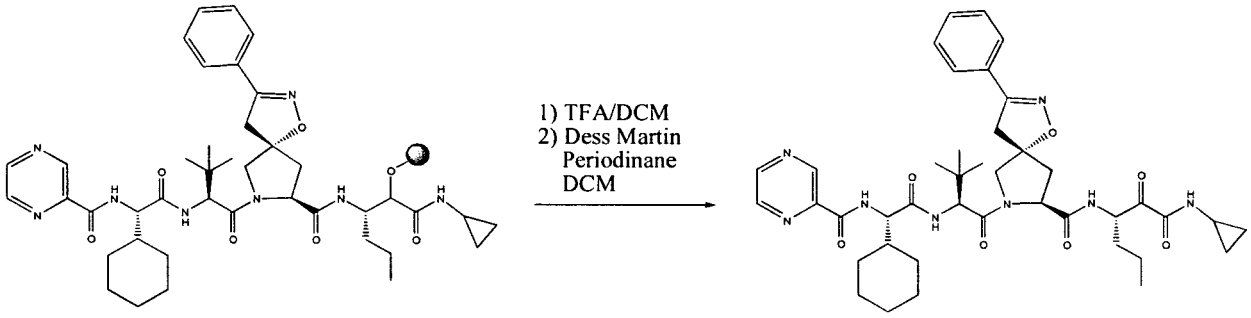
M2H **M2I**

[0412] Compound **M2H** (0.4 g, 1.0 eq.) was shaken overnight with a solution of FMOC-cyclohexylglycine (218 mg 3.0 eq.), HBTU (1.15 mL of 0.5 M in DMF, 3.0 eq.), HOBt (0.58 mL of 1.0 M in DMF, 3.0 eq.), and DIEA (167 uL, 5.0 eq.) in 2 mL DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice). The resin was then treated with 20% piperidine/DMF for 10 minutes. The resin was filtered and washed with DMF (thrice) followed by DCM (thrice). This process was repeated to give Compound **M2I**.



M2I **M2J**

[0413] Compound **M2I** (0.4 g, 1.0 eq.) was shaken overnight with a solution of pyrazine carboxylic acid (71 mg, 3.0 eq.), HBTU (1.15 mL of 0.5 M in DMF, 3.0 eq.), HOBt (0.58 mL of 1.0 M in DMF, 3.0 eq.), and DIEA (167 uL, 5.0 eq.) in 2 mL DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice) to give compound **M2J**. (M+H=772.9).



M2J **Compound No. 281**

[0414] Compound **M2J** (0.4 g, 1.0 eq.) was stirred in 1/1 TFA/DCM for 2 hours. The resin was drained and washed with DCM (thrice). The result was concentrated all organics and added DCM followed by Dess Martin Periodinane (97 mg, 3.0 eq.). Stirred for 1 hour and added 1N $Na_2S_2O_3$ and stirred. The solution was purified by silica gel chromatography (10-90% ethyl acetate/hexanes gradient) to yield 42 mg of Compound No. **281**. (M+H=771.0). ^{1}H-NMR (500 MHz, $CDCl_3$): 9.38 (d, 1H), 8.75 (d,1H), 8.56 (t, 1 H), 8.31 (d, 1H), 7.64-7.62 (m, 2H), 7.42-7.38 (m, 3H), 7.33 (d, 1H), 7.15 (s, 1H), 6.89 (d, 1H), 5.45-5.41 (m, 1H), 4.85 (t, 1H), 4.69 (d, 1H), 4.57-4.54 (m, 1H), 4.26 (d, 1H), 3.76 (d, 1H), 3.46 - 3.35 (m, 2H), 2.82 (td, 1H), 2.56 (d, 2H), 1.96-1.87 (m, 2H), 1.76 (m, 4H), 1.65-1.59 (m, 2H), 1.48-1.42 (m, 2H), 1.24 (m, 2H), 1.09 (m, 2H), 0.97 (s, 9H), 0.93 (t, 2H), 0.88-0.84 (m, 2H), 0.65 (t, 2H).

[0415] Listed below in Table 3 are additional compounds of Formula I prepared by Method 2.

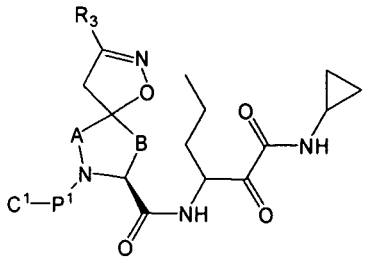


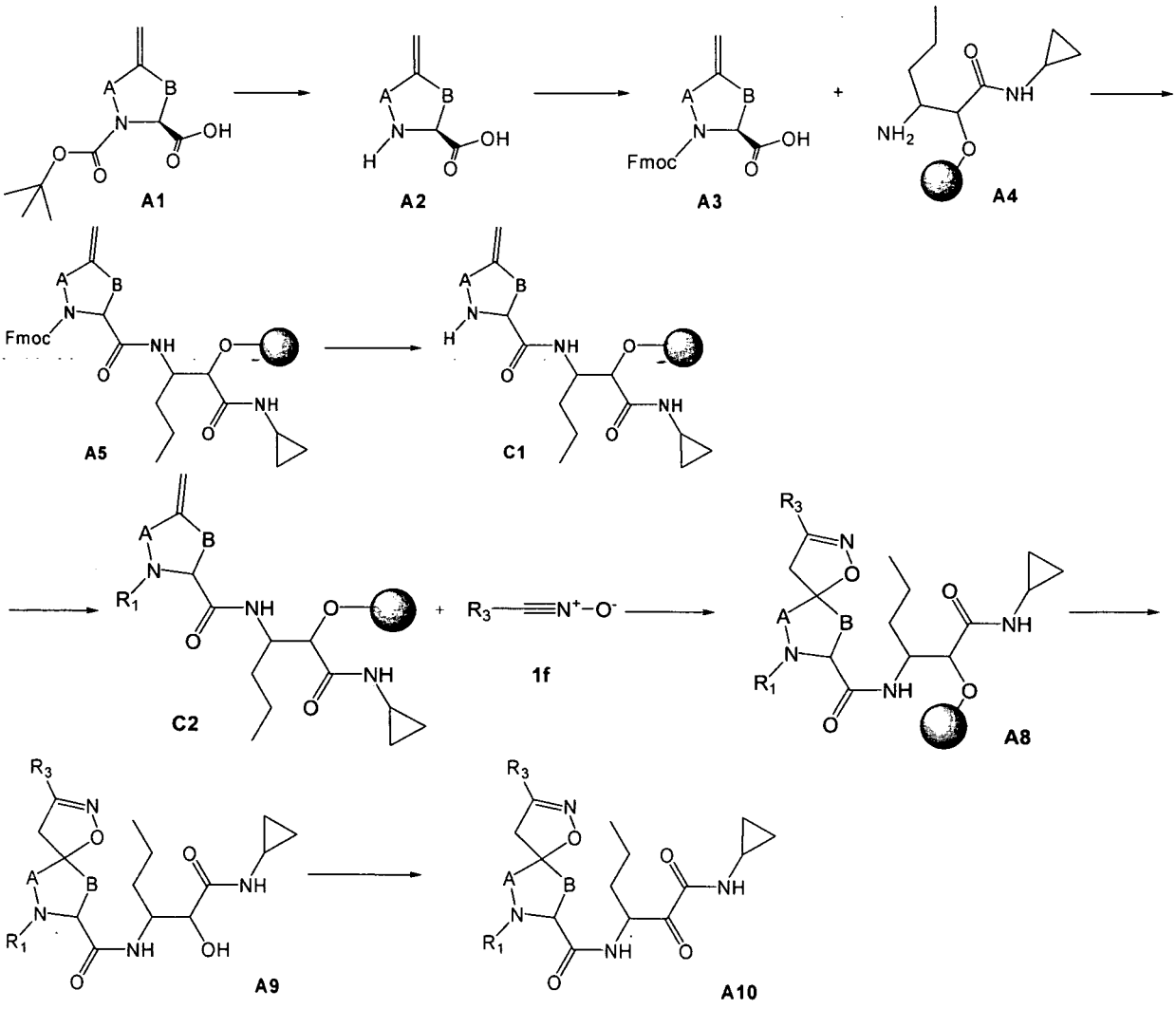
Table 3: Additional compounds of formula I produced by method 2.

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
40	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2,6-Dichlorobenzaldoxime
51	N/A	1H-pyrrole-2-carboxylic acid	Piperonal oxime
80	N/A	1H-pyrrole-2-carboxylic acid	Benzaldoxime
101	N-FMOC-L-tert-butylglycine	1-Naphthylsulfonyl chloride	Benzaldoxime
147	N-FMOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	2,6-Dichlorobenzaldoxime
151	N-Alloc-L-tert-butylglycine	N/A	Benzaldoxime
202	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Benzaldoxime
228	N-FMOC-L-tert-butylglycine	Acetic acid	Benzaldoxime
281	N-FMOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	Benzaldoxime
325	N-FMOC-L-tert-butylglycine	Acetic acid	Piperonal oxime
327	N-Alloc-L-tert-butylglycine	N/A	Piperonal oxime
343	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Piperonal oxime
428	N-FMOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	Benzaldoxime
464	N/A	1H-pyrrole-2-carboxylic acid	Piperonal oxime
491	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Benzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
527	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Piperonal oxime
536	N-FMOC-L-tert-butylglycine	1-Naphthylsulfonyl chloride	Piperonal oxime
570	N-FMOC-L-tert-butylglycine	Acetic acid	Piperonal oxime
578	N-FMOC-L-tert-butylglycine	Acetic acid	Benzaldoxime
584	N/A	1H-pyrrole-2-carboxylic acid	Benzaldoxime

[0416] Certain other compounds of Formula I may be prepared as illustrated by Method 3.

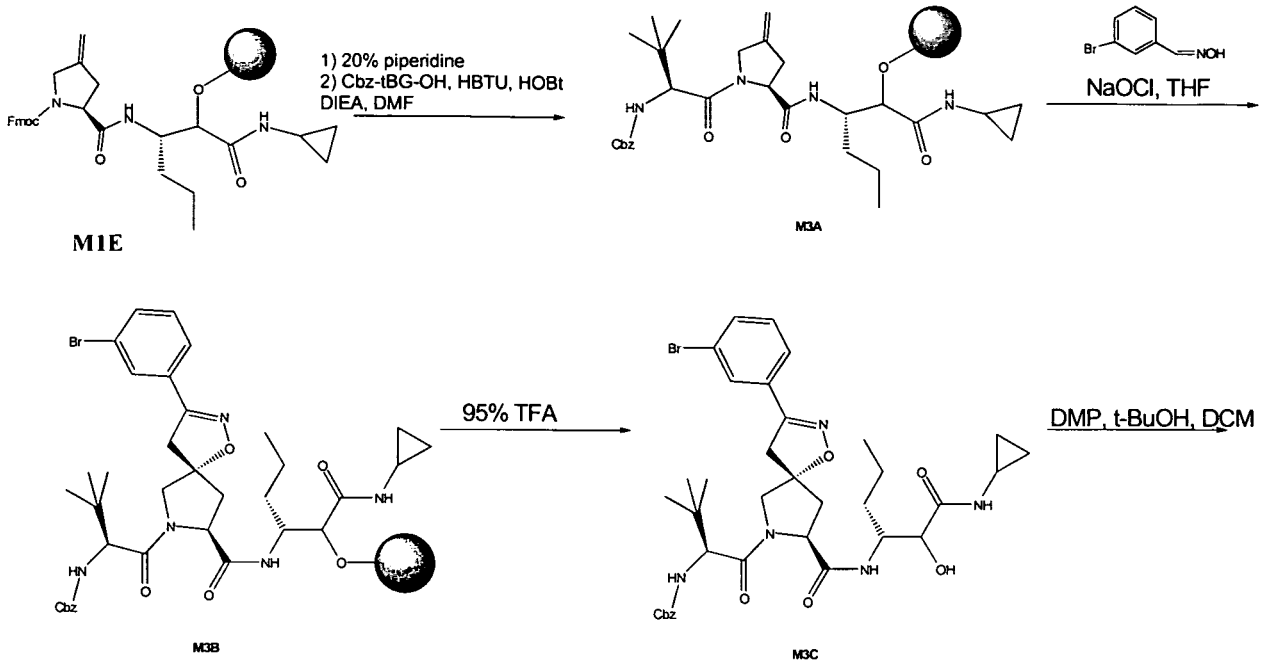
METHOD 3:



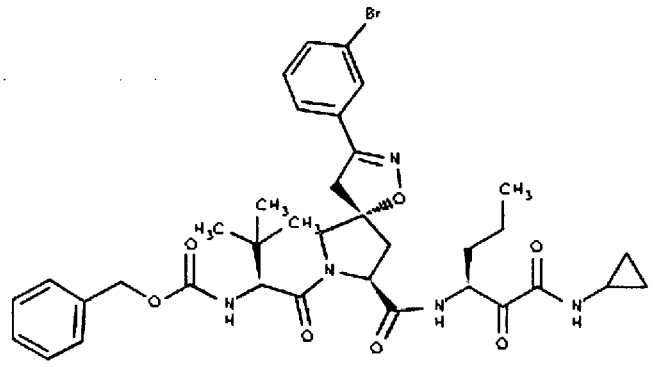
[0417] Referring to Method 3, the resin bound Fmoc exomethylene compound **A5**, prepared as in Method 1, is deprotected to give **C1**. Reaction of **C1** with an R_1 carboxylic

acid in the presence of a coupling reagent provides **C2** wherein R_1 is $R_4C(O)$-. Reaction of **C2** with the nitrile oxide **1f** leads to **A8** which is converted to **A10** as illustrated in Method 1.

Example 7: Compound No. 239



M1E M3A M3B M3C



Compound 239

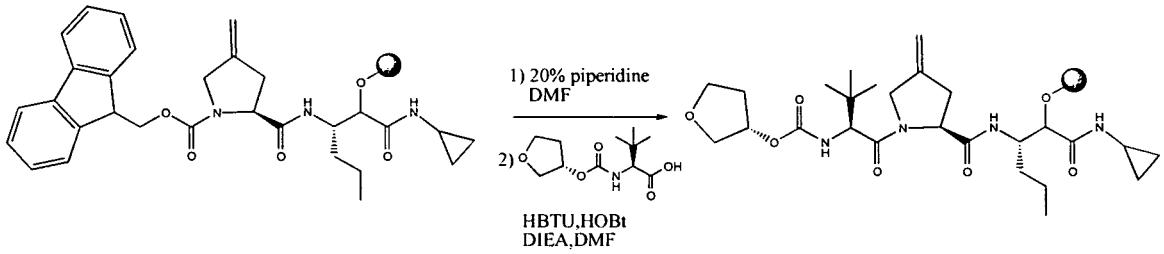
[0418] The resin **M1E** (0.47 mmol) was shaken in 20% piperidine/DMF for 10 minutes and then filtered and washed with DMF and DCM. The resulting resin was shaken again overnight with a solution of Cbz-tBG-OH (374 mg, 3.0 eq.), HOBT (2.82 mL of 0.5 M in DMF, 3.0 eq.), HBTU (2.82 of 0.5 M in DMF, 3.0e q.), and DIEA (0.493 mL, 6.0 eq.). The resin was then filtered and washed with DMF and DCM to give the resin compound **M3A** (0.47 g), which was used in next reaction without further purification.

[0419] The Cbz resin **M3A** (0.0611 mmol) in THF was shaken with 3-bromo-phenyl oxime (10 eq.) and bleach (5% NaOH) (20 eq.) for 12 hours. The resin was then filtered and washed with water, DMF, DCM to give the resin **M3B**.

[0420] The resin **M3B** was shaken with 95%TFA in water for 30 minutes and the resulting solution was concentrated in vacuo to give the compound **M3C** (0.031 mmol), (M+1) 740, which was used in next reaction without further purification.

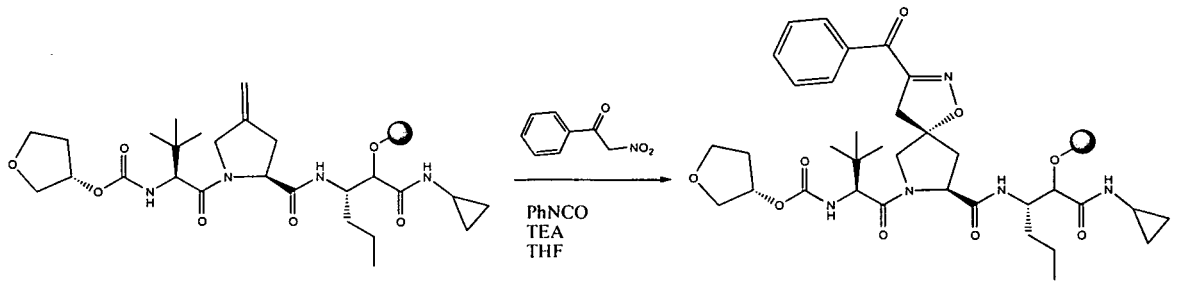
[0421] A solution of the compound **M3C** (0.031 mmol) in DCM (3 mL) was stirred with Dess-Martin Periodinane (26 mg, 2 eq.) and t-BuOH (26 uL). After stirring for 1 hour, sodium thiosulfate was added to above mixture. The product was extracted with EtOAc and the combined organic layer was then washed with water, $NaHCO_3$, brine and concentrated in vacuo and purified by Gilson Prep HPLC to afford Compound No. **239**. (M+1) 738.

Example 8: Compound No. 535



M1E **M3E**

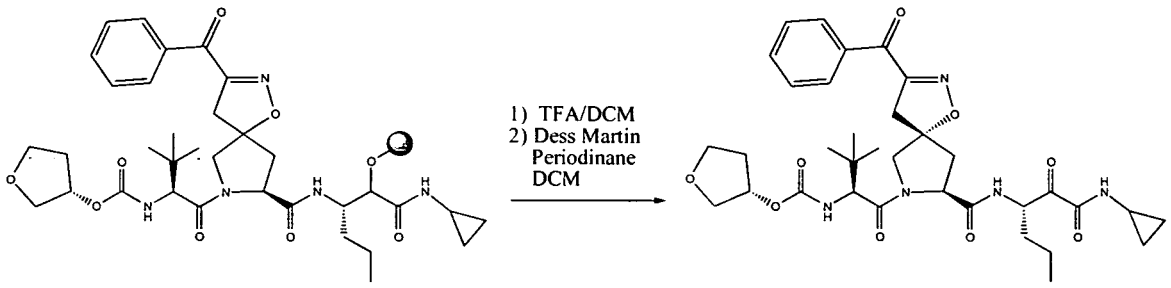
[0422] Compound **M1E** (10.0 g, 1.0 eq.) was shaken in 20% piperidine/DMF for 10 minutes. The resin was filtered and washed with DMF (thrice) followed by DCM (thrice). This process was repeated. The resulting resin was shaken overnight with a solution of (S)-2,3-dimethyl-2-(((S)-tetrahydrofuran-3-yloxy)carbonylamino)butanoic acid (3.46 g, 3.0 eq.), HBTU (28.2 mL of 0.5 M in DMF, 3.0 eq.), HOBt (14.1 mL of 1.0M in DMF, 3.0 eq.), and DIEA (4.91 mL, 6.0 eq.) in DMF. The resin was then filtered and washed with DMF (thrice) and DCM (thrice) to give compound **M3E**. (M+H=523.1)



M3E **M3F**

[0423] Compound **M3E** (300 mg, 1.0 eq.) was stirred in THF and 2-nitro-1-phenylethanone (272 mg, 10.0 eq.) was added to the mixture followed by phenyl isocyanate

(179 uL, 10.0 eq.) and catalytic TEA (10 uL). The resulting mixture was shaken overnight, drained, and washed with DMF, THF, and DCM (thrice each) to give compound **M3F** (M+H=669.8).



M3F **Compound No. 535**

[0424] Compound **M3F** (0.4 g, 1.0 eq.) was stirred in 1/1 TFA/DCM for 2 hours. The resin was drained and washed with DCM (thrice), all organics were concentrated, and DCM was added followed by Dess-Martin Periodinane (97 mg, 3.0 eq.). The resulting mixture was stirred for 1 hour, 1 N $Na_2S_2O_3$ was added and again, stirred. The reaction mixture was purified via silica gel chromatography (10-90% ethyl acetate/hexanes gradient) to yield Compound No. **535**. M+H=668.1. ^{1}H-NMR (500 MHz, $CDCl_3$): 8.19 (d, 2H), 7.61 (t, 1H), 7.47 (t, 2H), 7.19 (d, 1H), 6.93 (d, 1H), 5.52 (d, 1H), 5.37-5.33 (m, 1H, 5.24 (s, 1H), 4.78 (t, 1H), 4.32-4.29 (m, 2H), 3.93-3.79 (m, 4H), 3.70 (d, 1H), 3.48-3.36 (m, 2H), 2.79 (td,1 H), 2.68-2.63 (m, 1H), 2.55-2.50 (m, 1H), 2.12-2.04 (m, 1H), 1.96-1.89 (m, 1H), 1.66-1.59 (m, 1H), 1.47-1.37 (m, 2H), 1.00 (s, 9H), 0.94-0.81 (m, 6H), 0.63-0.57 (m, 2H).

[0425] Listed below in Table 4 are additional compounds of Formula I prepared by Method 3.

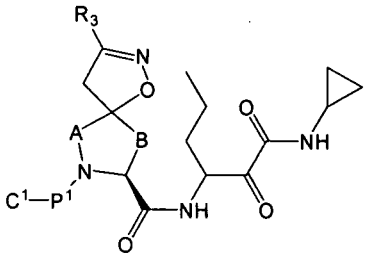


Table 4: Additional compounds of formula I produced by method 3.

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
4	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Chloro-5-fluorobenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
8	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-(Cyclopentyloxy)-4-methoxybenzaldehyde
9	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Di(trifluoromethyl)benzaldoxime
11	N/A	(S)-2,5-dioxopyrrolidin-1-yl tetrahydrofuran-3-yl carbonate	3-fluoro-4-methylbenzaldoxime
15	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Chloro-4-methoxybenzaldoxime
16	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	4-Methoxybenzaldoxime
25	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Difluorobenzaldoxime
32	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,4-Dichlorobenzaldoxime
36	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,4-Dimethylbenzaldoxime
47	N/A	(S)-2,5-dioxopyrrolidin-1-yl tetrahydrofuran-3-yl carbonate	4-Ethylbenzaldoxime
52	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Trifluoromethylbenzaldoxime
55	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	4-Chlorobenzaldoxime
56	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3,5-Dichlorobenzaldoxime
64	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Trifluoromethylbenzaldoxime
66	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chloro-4-fluorobenzaldoxime
70	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Cyclopentanecarboxaldehyde

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
78	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3,4-Dichlorobenzaldoxime
82	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	Piperonal oxime
83	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Chlorobenzaldoxime
95	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chloro-5-fluorobenzaldoxime
106	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Difluorobenzaldoxime
109	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Methyl-4-chlorobenzaldoxime
142	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Cyclohexanecarboxaldehyde
149	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Trifluoromethoxybenzaldoxime
150	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2,2-Dimethylchromane-6-carbaldehyde
171	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Cyanobenzaldoxime
177	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	4-Cyanobenzaldoxime
191	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Dimethyl-4-methoxybenzaldoxime
196	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,4-Dimethoxybenzaldoxime
198	N/A	(S)-2,5-dioxopyrrolidin-1-yl tetrahydrofuran-3-yl carbonate	3,5-Dimethyl-4-methoxybenzaldoxime
215	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,4,5-Trifluorobenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
222	N/A	(S)-2,5-dioxopyrrolidin-1-yl tetrahydrofuran-3-yl carbonate	2,2-Difluoro-1,3-benzodioxole-5-carboxaldehyde
224	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Dimethyl-4-methoxybenzaldoxime
229	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Methyl 4-nitrobutyrate
234	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chloro-5-fluorobenzaldoxime
236	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Chloro-4-methoxybenzaldoxime
239	N-CBZ-L-tert-butylglycine	N/A	3-Bromobenzaldoxime
240	N/A	(S)-tetrahydrofuran-3-yl-carbonate	4-Trifluoromethylbenzaldoxime
244	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Trifluoromethoxybenzaldoxime
251	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Phenylnitroethane
257	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Phenylbenzaldoxime
258	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-fluoro-5-trifluoromethylbenzaldoxime
270	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chloro-4-fluorobenzaldoxime
274	N/A	(S)-tetrahydrofuran-3-yl-carbonate	3,5-Dichlorobenzaldoxime
279	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chloro-4-methoxybenzaldoxime
285	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3,5-Dichlorobenzaldoxime
299	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	4-Chlorobenzaldoxime
301	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Chloro-4-fluorobenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
306	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Dichlorobenzaldoxime
314	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Methyl 4-formylbenzoate
316	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2,2-Difluoro-1,3-benzodioxole-5-carboxaldehyde
318	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Chlorobenzaldoxime
322	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Chloro-5-fluorobenzaldoxime
323	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3,4-Dichlorobenzaldoxime
330	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Di(trifluoromethyl)benzaldoxime
348	N/A	(S)-tetrahydrofuran-3-yl-carbonate	3-fluoro-5-trifluoromethylbenzaldoxime
353	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Methyl 3-formylbenzoate
362	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3,5-Dimethyl-4-methoxybenzaldoxime
363	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2,2-Difluoro-1,3-benzodioxole-5-carboxaldehyde
364	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Chlorophenylglyoxylohydroxamyl chloride
385	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Methylbenzaldoxime
391	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Dichlorobenzaldoxime

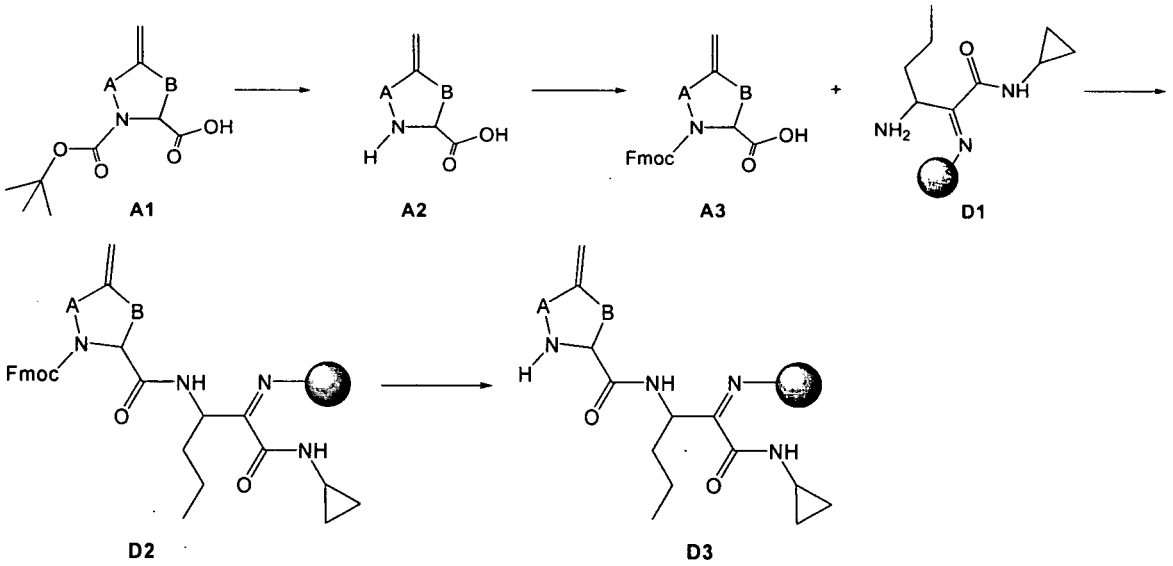
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
403	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2-Chloro-6-fluorobenzaldoxime
405	N/A	(S)-tetrahydrofuran-3-yl-carbonate	4-Isopropylbenzaldoxime
413	N/A	(S)-tetrahydrofuran-3-yl-carbonate	3-Methyl-4-fluorobenzaldoxime
414	N/A	(S)-tetrahydrofuran-3-yl-carbonate	3,4,5-Trifluorobenzaldoxime
423	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-fluoro-4-methylbenzaldoxime
425	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-(4-Pyridyl)benzaldehyde
434	N-CBZ-L-tert-butylglycine	N/A	2,3-Dimethoxybenzaldoxime
436	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-Methyl-4-chlorobenzaldoxime
444	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Methylbenzaldoxime
448	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-(4-chlorophenyl(-2,1-benzisoxazole-5-carbaldehyde oxime
451	N/A	(S)-tetrahydrofuran-3-yl-carbonate	3-Trifluoromethyl-4-fluorobenzaldoxime
455	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,4,5-Trifluorobenzaldoxime
456	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	1,4-benzodioxan-6-carboxaldehyde
472	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2-Furanaldoxime
480	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Methyl 3-formylbenzoate
481	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-(Carboxy)benzaldoxime

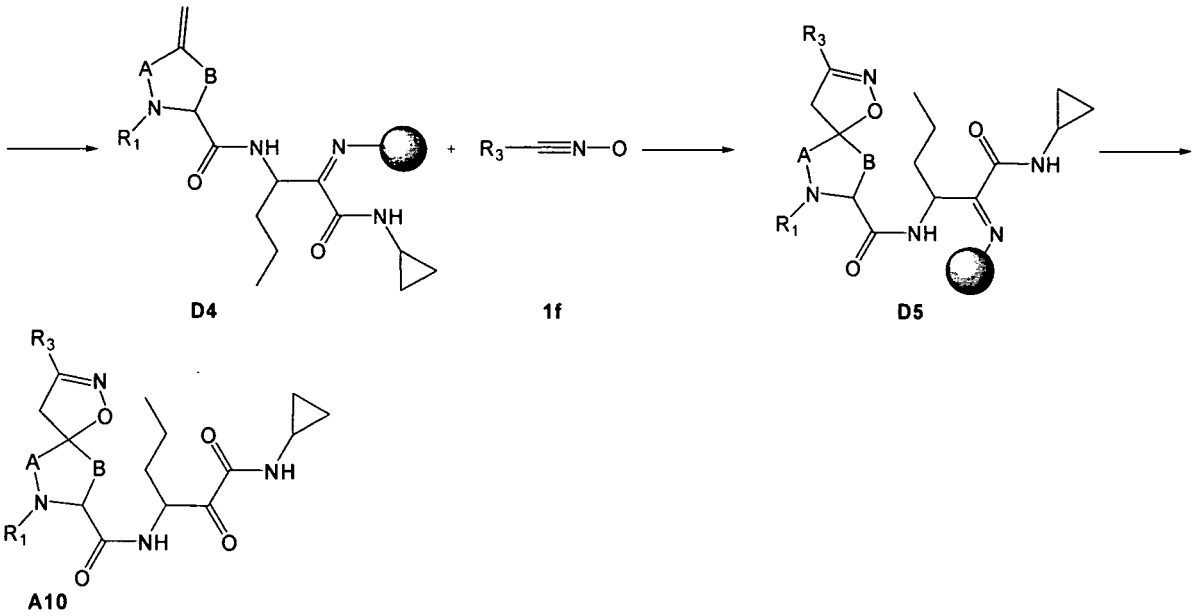
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
482	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2-Chloro-6-fluorobenzaldoxime
486	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chloro-4-methoxybenzaldoxime
490	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Methyl-4-chlorobenzaldoxime
498	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-fluoro-5-trifluoromethylbenzaldoxime
509	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Trifluoromethylbenzaldoxime
511	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Nitrobenzaldoxime
519	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3,5-Dimethylbenzaldoxime
524	N/A	(S)-tetrahydrofuran-3-yl-carbonate	4-Trifluoromethoxybenzaldoxime
525	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Methoxybenzaldoxime
530	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-Hydroxybenzaldoxime
535	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2-nitro-1-phenyl ethanone
539	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Methyl-4-fluorobenzaldoxime
543	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	4-(Carboxy)benzaldoxime
545	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Trifluoromethyl-4-fluorobenzaldoxime
548	N-FMOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chloro-4-fluorobenzaldoxime
550	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-fluoro-4-methylbenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
551	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Methyl 4-formylbenzoate
555	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Methyl-4-fluorobenzaldoxime
560	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	3-Trifluoromethylbenzaldoxime
572	N-FMOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3,5-Dimethyl-4-methoxybenzaldoxime

[0426] Certain other compounds of Formula I may be prepared as illustrated by Method 4.

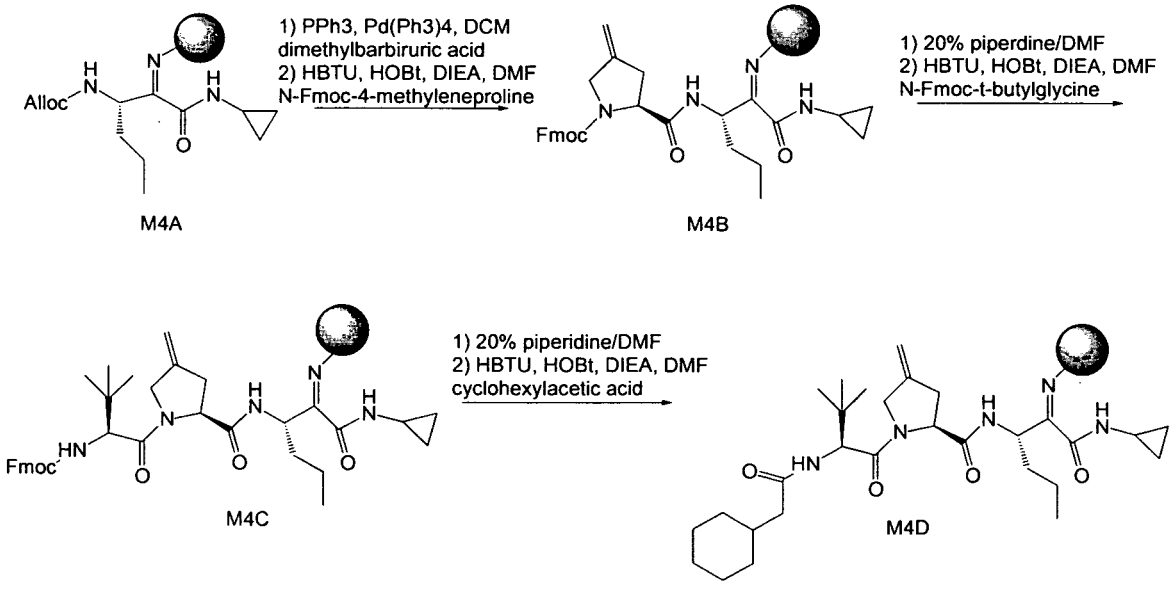
METHOD 4:





[0427] Referring to Method 4, the Fmoc derivative **A3** is prepared as described in Method 1. Reaction of **A3** with the resin bound imino amide **D1** in the presence of a coupling reagent provides the compound bound resin **D2**. The resin bound imino amide **D1** may be prepared from the diketo compound **X31** by reaction with an amino resin such as, for example, a derivatized aminomethylated polystyrene, e.g., **X32**. Deprotection of **D2** provides **D3** which reacts with an R_1 carboxylic acid in the presence of a coupling reagent to provide **D4** wherein R_1 is $R_4C(O)$-. Reaction of **D4** with the nitrile oxide **1f** provides **D5** which on hydroysis from the resin provides **A10**.

Example 9: Compound No. 303

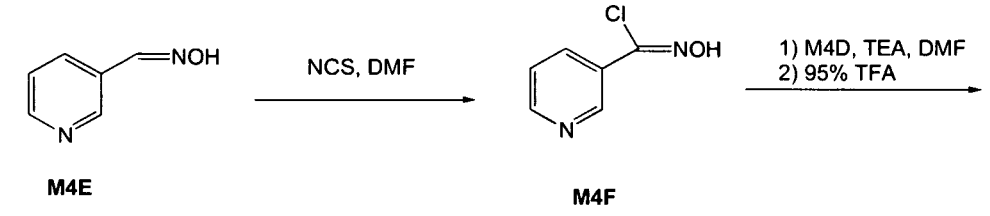


[0428] To a suspension of resin **M4A**, which has the same structure as **X33**, (20 g, 0.4 mmol/g, 8 mmol) in DCM (100 mL) was added PPh_3 (21 g, 80 mmol), dimethyl barbituric

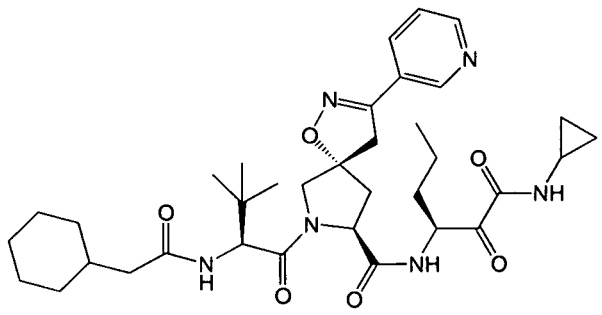
acid (12.5 g, 80 mmol) and $Pd(PPh_3)_4$ (920 mg, 0.8 mmol). The suspension was shaken overnight, drained, washed with DMF (10 times) and DCM (4 times). N-Fmoc-4-methyleneproline (3.0 g, 8.8 mmol), HBTU (3.3 g, 8.8 mml) and HOBt (1.1 g, 8.8 mmol) and DIEA (1.6 mL, 8.8 mmol) were dissolved in DMF (100 mL). The solution was added to the resin and shaken overnight. The resin was then drained, washed with DMF (10 times), DCM (4 times) and dried to afford resin **M4B**.

[0429] To the resin **M4B** (20g, 8 mmol) was added 20% piperdine in DMF (100 mL), shaken for 1 hour, and then washed with DMF (10 times), DCM (4 times). To the resin was added a mixture of Fmoc-tert-butylglycine (5.6 g, 16 mmol), HBTU (6.1 g, 16 mmol), HOBt (2.2 g, 16 mmol) and $(iPr)_2NEt$ (2.9 mL, 16 mmol) in DMF (100 mL). The suspension was shaken overnight, drained, washes with DMF (10 times), DCM (4 times) and dried to afford the resin **M4C**.

[0430] To the resin **M4C** (20 g, 8 mmol) was added 20% piperdine in DMF (100 mL), shaken for 1 hour, and then washed with DMF (10 times), DCM (4 times). To the resin was added a mixture of cyclohexylacetic acid (1.42 g, 10 mmol), HBTU (3.8 g, 10 mmol), HOBt (1.35 g, 10 mmol) and $(iPr)_2NEt$ (1.8 mL, 10 mmol) in DMF (100 mL). The suspension was shaken overnight, drained, washes with DMF (10 times), DCM (4 times) and dried to afford the resin **M4D**.



M4E **M4F**



Compound No. 303

[0431] A solution of 3-pyridinealdoxime (**M4E**) (122 mg, 1 mmol) in DMF (3 mL) was added NCS (134 mg, 1 mmol). The mixture was heated to 50-60 °C for 30 minutes. After cooling down to room temperature, the 3-pyridinechloroxime (**M4F**) solution was added to a resin **M4D** (300 mg, 0.12 mmol). To the mixture was added TEA (0.14 mL, 1 mmol) and the

reaction mixture was heated to 50-60 °C for 4 hours. The reaction mixture was drained and washed with DMF (6 times) and DCM (6 times). The resin was treated with 95% TFA for 5 hours. The mixture was drained, and washed with DCM. The filtrate was concentrated in vacuo, purified from column 50-100% EtOAc/Hex to afford 7 mg colorless solid as product Compound No. **303**. HPLC 5.7-6.4 minutes; MS 651.5 and LC-MS 3.9 minutes.

[0432] Listed below in Table 5 are additional compounds produced by Method 4.

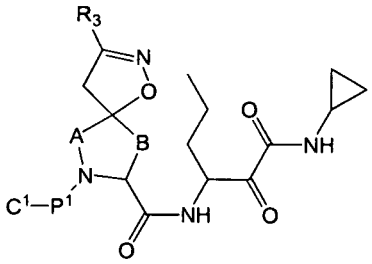
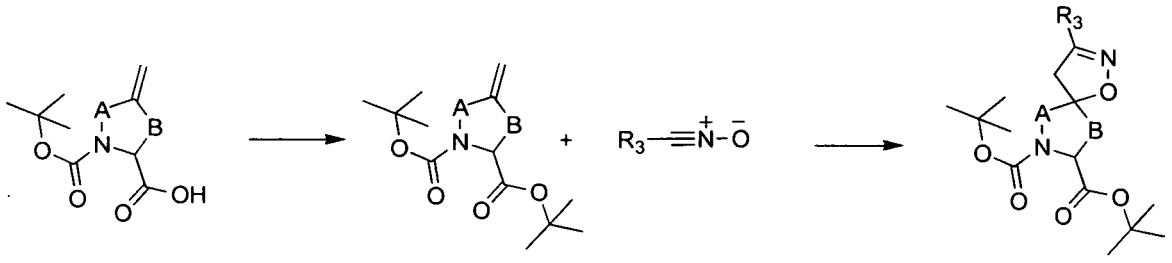


Table 5: Additional compounds of formula I produced by method 4.

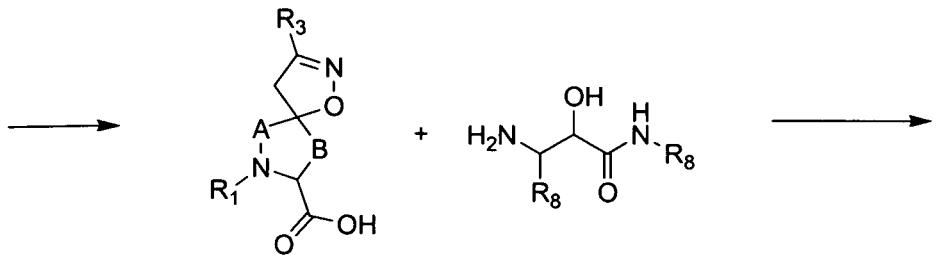
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
41	N-FMOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Bromoprydine-3-carbaldehyde
179	N-FMOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Bromo-2-Furaldehyde
230	N-FMOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2-methylbenzofuran-3-carbaldehyde
303	N-FMOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Pyridine-carboxaldehyde
495	N-FMOC-L-tert-butylglycine	2-Cyclohexylacetic acid	4-Chloro-1-methyl-1H-pyrazole-3-carbaldehyde
552	N-FMOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2,3-Dihydrobenzo[b]furan-5-carboxaldehyde

[0433] Certain other compounds of the invention may be prepared as illustrated in Methods 5A and 5B.

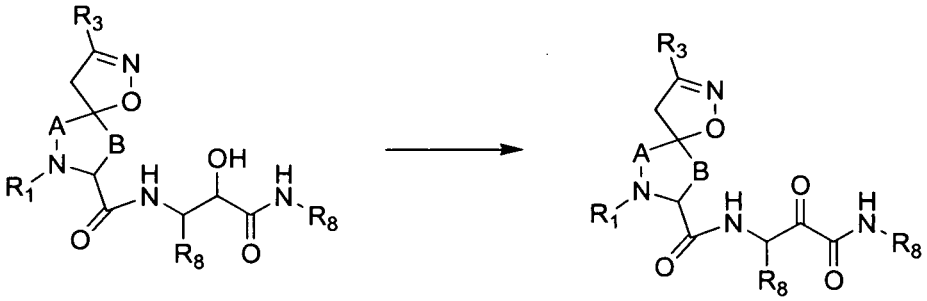
METHOD 5A:



A1 **E1** **1f** **B1**



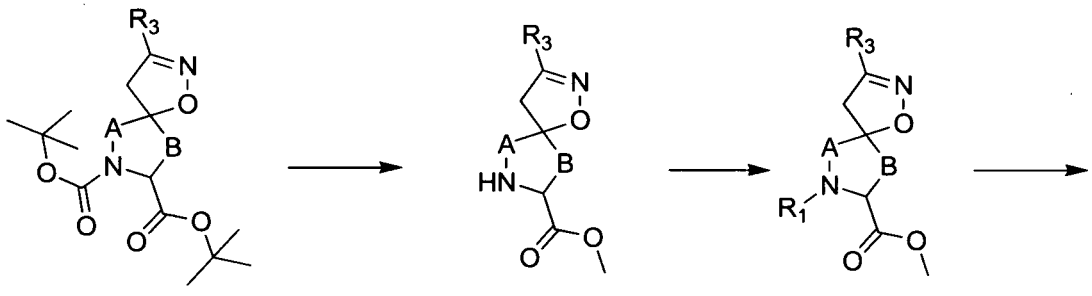
E2 E5



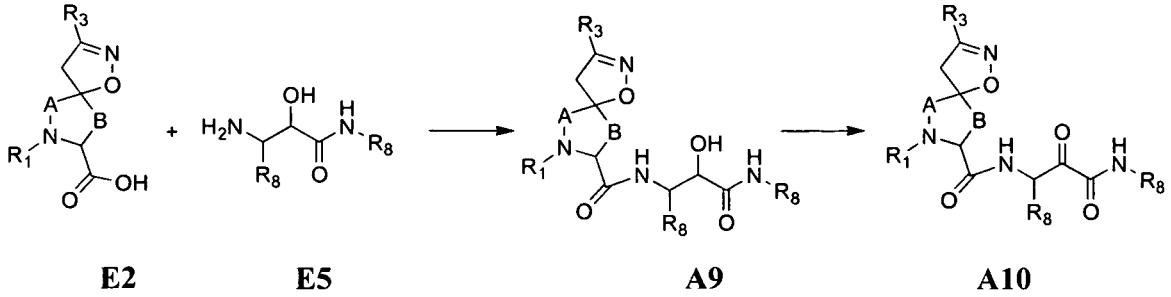
A9 A10

[0434] Referring to Method 5a, the exomethylene acid compound **A1** is protected to provide the di-t-butyl dicarboxylate **E1**. Reaction of **E1** with nitrile oxide of formula **1f** provides the intermediate **B1** which is transformed into amino acid derivative **E2**. Reaction of **E2** with an aminoalcohol **E5** provides **A9**. Compound **A9** is converted to **A10** as described in Method 1.

METHOD 5B:



B1 E6 E7

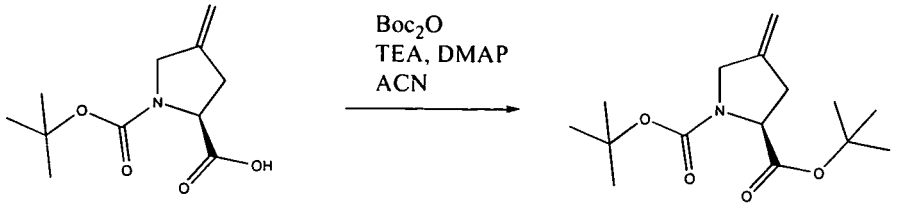


E2 E5 A9 A10

[0435] Referring to Method 5b, the intermediate compound **B1** is transformed into amino acid ester **E6**. Reaction of **E6** with an R_1 carboxylic acid in the presence of a coupling

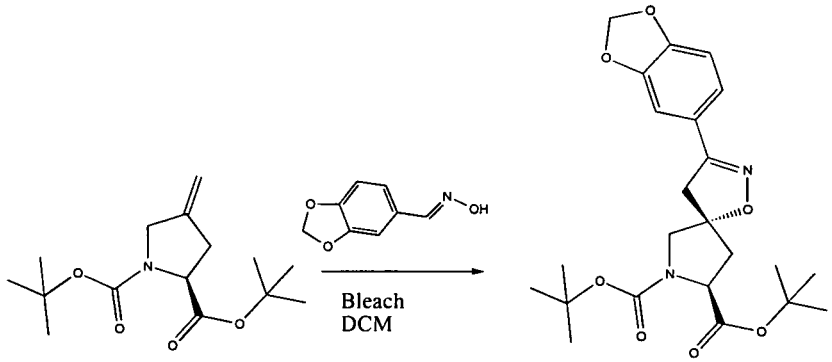
reagent provides **E7** wherein R_1 is $R_4C(O)$-. **E7** is deprotected to provide **E2** which is converted to **A10** as described in Method 1.

Example 10: Compound No. 422



10A **M2B**

[0436] Compound **10A** (5.0g, 1.0eq.) was stirred in 100 mL acetonitrile and to the solution were added ditertbutyldicarbonate (9.6 g, 2 eq.), dimethylaminopyridine (537 mg, 0.2 eq.), and triethylamine (6.13 mL, 2.0 eq.). The mixture was stirred overnight, concentrated, added ethyl acetate, washed with 1.0N HCl, dried over sodium sulfate, concentrated, and purified with silica gel chromatography (10-30% ethyl acetate/hexanes gradient) to give compound **M2B**. (M+H=284.0). ^{1}H-NMR ($CDCl_3$): 5.0 (m, 2H), 4.3-4.5 (m, 1H), 4.0-4.1 (m, 2H), 2.9-3.0 (m, 1H), 2.5-2.6 (d, 1H), 1.5(s, 3/9 of 18H), 1.4(s, 6/9 of 18H).

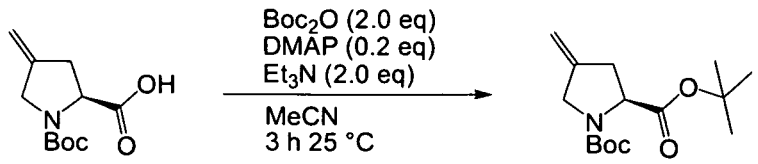


10B **10C**

[0437] Compound **10B** (10.0 g, 1.0 eq.) was stirred in 175 mL DCM with piperonaloxime (11.5 g, 2.0 eq.). The solution was cooled on an ice bath and to it added bleach (175 mL) slowly. The mixture was then warmed to room temperature, stirred for 2 hours, separated and its aqueous layer extracted with DCM twice. Organics were combined and dried over magnesium sulfate, filtered and concentrated. The residue was purified and separated diastereomers by silica gel chromatography (5-30% ethyl acetate/hexanes gradient) to yield 4.1 g of Compound **10C**. (M+H=446.9.) ^{1}H-NMR ($CDCl_3$): 7.25 (m, 1H), 7.0 (d, 1H), 6.8 (d, 1H), 6.0 (s, 2H), 4.6-4.4 (m, 1H), 4.0-3.8 (m, 1H), 3.7-3.6 (m, 1H) 3.4-3.3 (m, 1H), 3.3-3.2 (m, 1H), 2.8-2.7 (m, 1H), 2.3-2.2 (m, 1H), 1.5 (s, 9H), 1.4 (s, 9H).

[0438] Alternatively, compound **10B** was prepared by the following procedures:

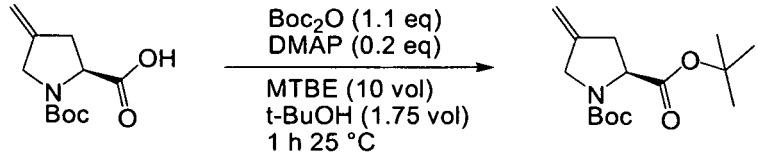
Preparation: (S)-di-tert-butyl 4-methylenepyrrolidine-1,2-dicarboxylate.



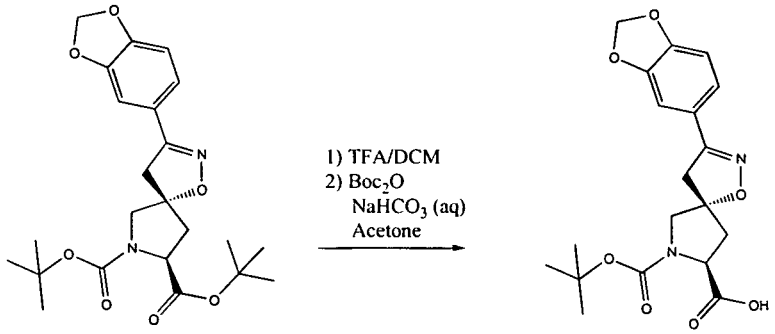
Procedure 1

[0439] Triethylamine (2 eq.) was added to a solution of (S)-1-(tert-butoxycarbonyl)-4-methylenepyrrolidine-2-carboxylic acid (1.0 eq.), di-tert-butyldicarbonate (2.0 eq.), and DMAP (0.2 eq.) in acetonitrile (10 vol) at ambient temperature. The reaction mixture was stirred for 16 h, then diluted with isopropyl acetate (25 vol). A wash with water (20 vol., twice) was followed by a filtration over Na_2SO_4 and solvent removal. The crude product was purified by filtration through a pad of silica gel (37 vol silica, first flush with heptane (80 vol), second flush with 10% ethyl acetate in heptane (30 vol)). Removal of solvent from the second flush gave compound **10B**.

Procedure 2



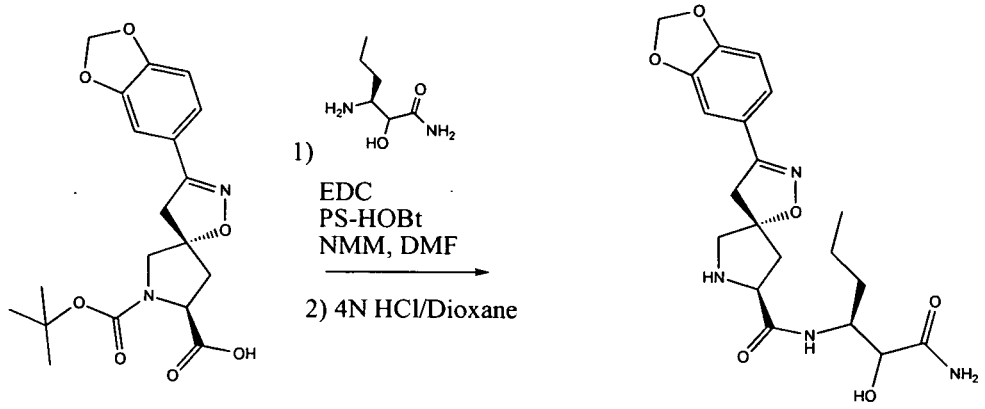
[0440] A solution of di-tert-butyl dicarbonate (1.1 eq.) in MTBE (2 vol.) was added to a mixture of (S)-1-(tert-butoxycarbonyl)-4-methylenepyrrolidine-2-carboxylic acid (1.0 eq.) and DMAP (0.2 eq.) in MTBE (8 vol) and t-butanol (1.75 vol.). The mixture was stirred for 1 hour, at which point gas evolution ceased. The mixture was washed with 1 N HCl (3 vol.), then saturated aqueous $NaHCO_3$ (3 vol.) and then brine (3 vol.). The solvent is then removed to afford compound **10B**.



10C **10D**

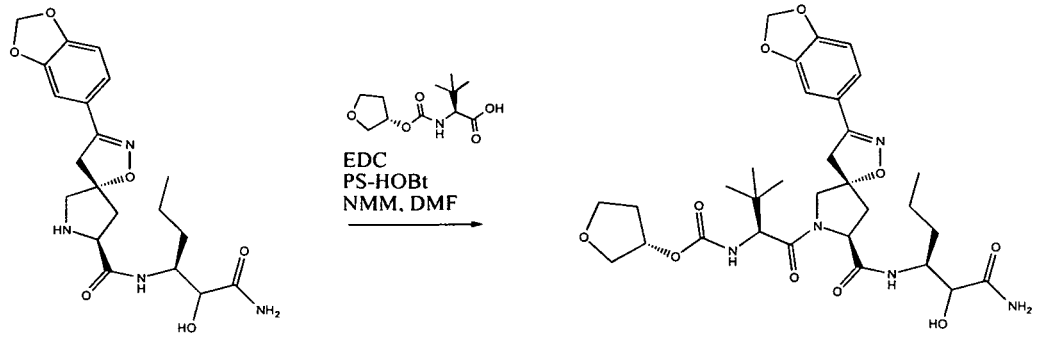
[0441] Compound **10C** (4.0 g, 1.0 eq.) was stirred in 1/1 TFA/DCM for 3 hours and the solution was concentrated. To the concentrate was added 100mL acetone, 100mL saturated sodium bicarbonate solution, and ditertbutyldicarbonate and the resulting solution was stirred

overnight and then acidified with 1.0 N HCl solution and extracted with ethyl acetate (thrice). The organics were washied with brine solution and dried over magnesium sulfate, filtered and concentrated to yield 4.0 g of Compound **10D** (M+H=391.1).



10D **10E**

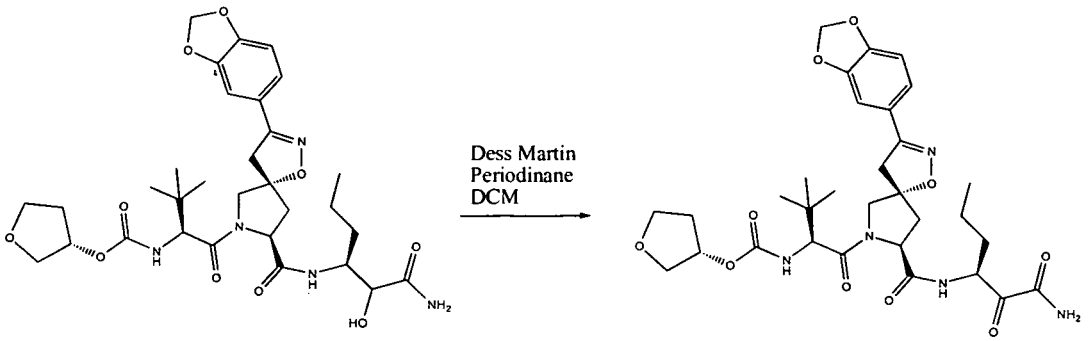
[0442] Compound **10D** (50 mg, 1.0 eq.) stirred in 0.5 mL DMF with EDC (37 mg, 1.5 eq.), PS-HOBt (137 mg, 1.5 eq.) and NMM (56 uL, 4.0 eq.), and to the solution was added 0.5mL DCM to assist in swelling of the resin. To the mixture was added 3-amino-2-hydroxyhexanamide (30 mg, 1.3 eq.) and the mixture was stirred overnight, filtered, diluted with ethyl acetate, washed with 1.0 N HCl, dried over sodium sulfate, filtered, and concentrated. The solution was purified by silica gel chromatography (100% DCM-5%MeOH/DCM gradient) to yield 21 mg of the crude product, which was then stirred in 4.0 N HCl/dioxane for 2 hours and concentrated to yield compound **10E** as an HCl salt (M+H=419.0).



10E **10F**

[0443] Compound **10E** (21mg, 1.0eq.) was stirred in DMF with NMM (13uL, 1.4eq.) and to the solution was added a solution of (S)-3,3-dimethyl-2-(((S)-tetrahydrofuran-3-yloxy)carbonylamino)butanoic acide (14 mg, 1.4 eq.), EDC (11 mg, 1.4 eq.), and PS-HOBt (40 mg, 1.4 eq.) in DMF, with enough DCM to swell the resin. The mixture was stirred overnight, filtered, washed with 1.0 N HCl, dried over sodium sulfate, filtered and then

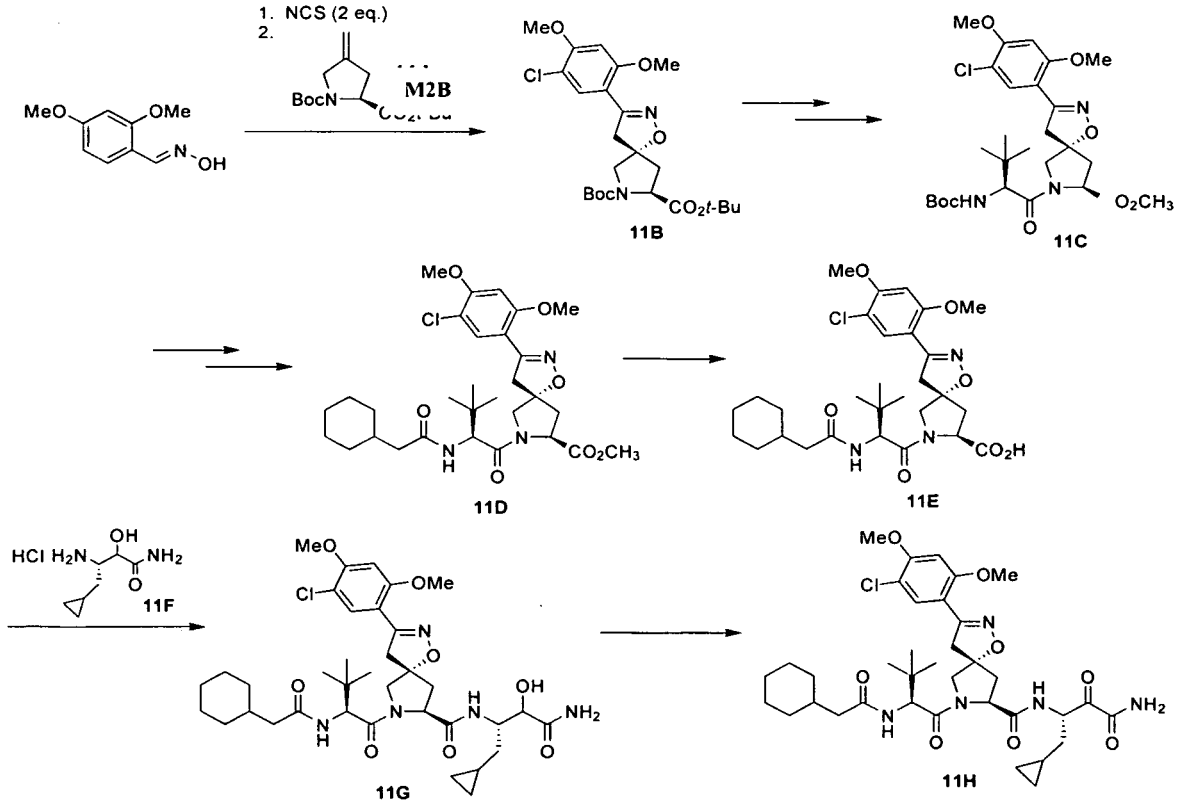
concentrated to give compound **10F**, which was used without further purification. (M+H=646.4)



10F **Compound No. 422**

[0444] Compound **10F** was stirred in DCM and to the solution was added Dess-Martin Periodinane (≈3.0eq.). The solution was stirred for 1 hour, added to it 1.0 N $Na_2S_2O_3$, and stirred. The mixture was purified by silica gel chromatography (10-90% ethyl acetate/hexanes gradient) to yield 9 mg of Compound No. **422** (M+H=644.3). ^{1}H-NMR ($CDCl_3$): 7.3 (m, 1H), 7.15(m, 1H), 6.95 (m, 1H), 6.8 (m, 1H), 6.75 (m, 1H), 6.0 (s, 2H), 5.5-5.4 (m, 2H), 5.4-5.3 (m, 2H), 4.8-4.7 (m, 1H), 4.3 (m, 1H), 4.2 (m, 1H), 4.0-3.8 (m, 3H), 3.7 (m, 1H), 3.4-3.2 (m, 2H), 2.6 (m, 1H,), 2.5 (m, 1H), 2.2-2.1 (m, 1H), 2.1-2.0 (m, 1H), 1.9 (m, 1H), 1.6 (m, 1H), 1.5-1.4 (m, 2H), 1.0-0.9 (m, 13H).

Example 11: Compound No. 562.



[0445] To 2,4-dimethoxybenzaldoxime (4.5 g, 24.8 mmol) in DMF (135 mL) was added dropwise over 2 h at room temperature a solution of N-chlorosuccinimide (6.6 g, 49.7 mmol) in DMF (135 mL). The reaction was stirred 14 hours and compound **M2B** (5.2 g, 18.4 mmol) was added followed by dropwise addition over 1 h of a solution of triethylamine in DMF (2.6 mL, 18.4 mmol, in 15 mL). After stirring for 3 h, the reaction mixture was washed with H_2O and dried over $MgSO_4$. The resulting residue was purified via silica gel chromatography to afford 5.8 g of compound **11B** as a tan solid. ES (+) MS: m/e 497 $(M + H)^+$.

[0446] To compound **11B** (5.5 g, 11.1 mmol) in CH_2Cl_2 (30 mL) was added trifluoroacetic acid (30 mL). The reaction mixture was stirred for 90 minutes at room temperature and concentrated under reduced pressure to provide a tan solid, which was dissolved in MeOH (60 mL) and heated to reflux. Concentrated sulfuric acid (≈5 mL) was added dropwise and the reaction was refluxed for 3 hours, after which the solvent was removed under reduced pressure. The resulting residue was dissolved in CH_2Cl_2 (75 mL) and carefully treated with a saturated $NaHCO_3$ solution until pH ≈ 9. The organic layer was dried over $MgSO_4$ and concentrated to provide the intermedaite amino ester. To N-Boc-tert-butylglycine (3.1 g, 13.6 mmol) in CH_2Cl_2 (60 mL) was added EDC (2.6 g, 13.6 mmol), HOBt (1.8 g, 13.6 mmol) and triethylamine (5.5 mL, 39.5 mmol). After stirring 5 minutes, the above amino ester was added and the reaction was stirred at room temperature 14 hours. The reaction mixture was washed with H_2O, 1 N HCl, and saturated $NaHCO_3$ solution. The organic layer was dried over $MgSO_4$ and concentrated in vacuo to provide 5.6 g of compound **11C** (over 3 steps) as a brown solid which was used without further purification. ES (+) MS: m/e 568 $(M + H)^+$.

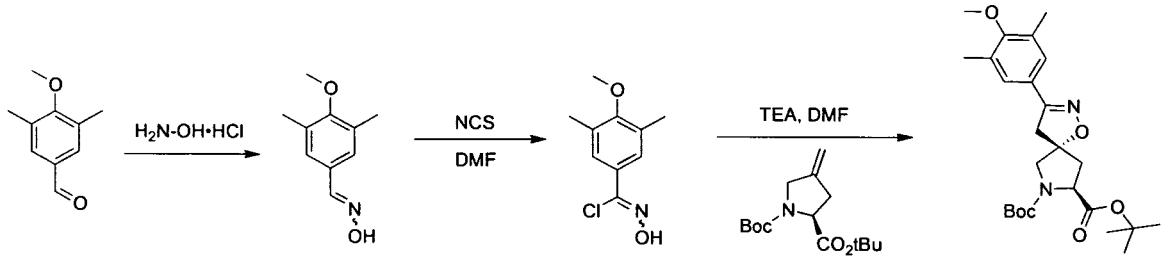
[0447] To compound **11C** (600 mg, 1.1 mmol) in CH_2Cl_2 (3 mL) was added trifluoroacetic acid (3 mL). The reaction was stirred for 1 hour and concentrated under reduced pressure to give the desired amine product as the TFA salt. To cyclohexylacetic acid (181 mg, 1.3 mmol) in CH_2Cl_2 (6 mL) was added EDC (243 mg, 1.3 mmol), HOBt (171 mg, 1.3 mmol) and triethylamine (516 μL, 3.7 mmol). After stirring for 5 minutes, the above amine was added and the reaction was stirred at room temperature 14 hours. The reaction mixture was washed with H_2O, 1 N HCl, and saturated $NaHCO_3$ solution. The organic layer was dried over $MgSO_4$ and concentrated in vacuo, and the resulting residue was purified via silica gel chromatography to provide 460 mg of compound **11D** (over 2 steps) as an off-white solid. ES (+) MS: m/e 592 $(M + H)^+$.

[0448] To compound **11D** (460 mg, 0.8 mmol) in a solution of THF/H_2O (5 mL, 3:1 v/v) was added LiOH monohydrate (82 mg, 1.9 mmol). The reaction mixture was stirred at room temperature 14 hours, acidified using 1 N HCl, and extracted with EtOAc. The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure to provide 405 mg of compound **11E**, which was used without further purification. ES (+) MS: m/e 578 $(M + H)^+$.

[0449] To compound **11E** (80 mg, 0.14 mmol) in CH_2Cl_2 (1 mL) was added EDC (38 mg, 0.2 mmol), HOBt (27 mg, 0.2 mmol) and triethylamine (68 μL, 0.5 mmol). After stirring for 5 minutes, compound **11F** was added and the reaction was stirred at room temperature 14 hours. The reaction mixture was washed with H_2O, 1 N HCl, and saturated $NaHCO_3$ solution. The organic layer was dried over $MgSO_4$ and concentrated in vacuo to provide 95 mg of compound **11G** as a brown solid which was used without further purification. ES (+) MS: m/e 718 $(M + H)^+$.

[0450] To compound **11G** (95 mg, 0.14 mmol) in CH_2Cl_2 (1 mL) was added Dess-Martin periodinane (71 mg, 0.17 mmol). After stirring for 30 minutes, the reaction was quenched with 1 N $Na_2S_2O_3$. The organic layer was purified via silica gel chromatography to give Compound No. **562**, i.e., compound **11H** shown above, as a white solid. ES (+) MS: m/e 716 $(M + H)^+$.

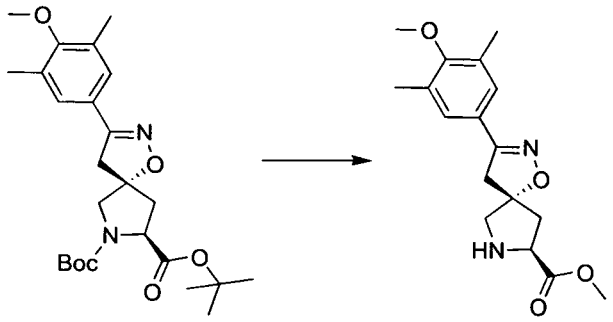
Example 12: Compound No. 362



12A

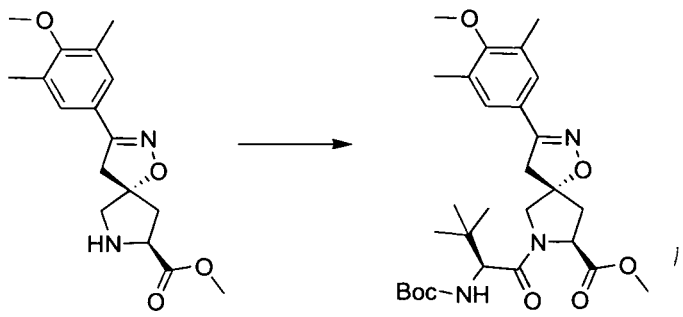
[0451] 4-Methoxy-3,5-dimethylbenzaldehyde (1.86 g, 11.3 mmol) was dissolved in ethanol (30 mL) and stirred with hydroxylamine hydrochloride (2.4 M aq. solution, 5.65 mL, 1.2 eq.) and Na_2CO_3 (1.2 M solution, 5.65 mL, 0.6 eq.) at room temperature for 2.5 hours. The mixture was then heated to 60 °C and additional hydroxylamine hydrochloride and Na_2CO_3 was added. The mixture was again stirred overnight at 60 °C, transferred to a separatory funnel, diluted with EtOAc. The organic layer was separated, washed with brine, dried over $MgSO_4$, filtered and concentrated. The product was purified by ISCO chromatography with EtOAc/hexanes eluent to yield 1.55 g (8.56 mmol) of 4-methoxy-3,5-dimethylbenzaldehyde oxime as a white solid. M+1 = 180.0.

[0452] To a solution 4-methoxy-3,5-dimethylbenzaldehyde oxime (1.34 g, 7.48 mmol) in DMF (10 mL) was added N-chlorosuccinimide (1.76 g, 13.2 mmol). This solution was stirred until starting material was consumed as indicated by HPLC. To the solution was then added a solution of (S)-di-tert-butyl 4-methylenepyrrolidine-1,2-dicarboxylate (2.1 g, 1.0 eq.) in DMF (5 mL). To the solution was added triethylamine (1.2 eq.) dropwise, and the reaction mixture was stirred for 2 hours. The reaction was then diluted with EtOAc and the organic phase was washed with water, brine, dried ($MgSO_4$), filtered and concentrated. The product was purified over silica gel on an ISCO Combiflash using EtOAc/hexanes as the eluent to yield 912 mg (1.98 mmol) of compound **12A**. M+1 = 461.4. ^{1}H-NMR (500 MHz, $CDCl_3$): 7.30 (s, 2H), 4.40-4.32 (m, 1H), 3.98-3.79 (m, 1H), 3.74 (s, 3H), 3.64-3.58 (m, 1H), 3.40-3.34 (m, 1H), 3.24-3.19 (m, 1H), 2.72 (dd, J = 8.7, 12.9 Hz, 1H), 2.29 (s, 6H), 2.11-2.07 (m, 1H), 1.54-1.45 (m, 18H).



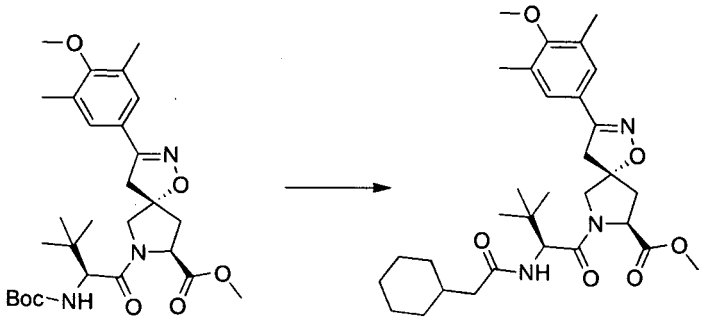
12A **12B**

[0453] Compound **12A** (910 mg, 1.98 mmol) was stirred in CH_2Cl_2/trifluoroacetic acid (1:1, 20 mL) until HPLC indicated complete deprotection of starting material. The intermediate amino acid was concentrated and then dissolved in methanol (30 mL) and heated to relux with concentrated H_2SO_4 until the starting material was consumed as indicated by HPLC. Concentrated material in vacuo, then dissolved in EtOAc and washed with $NaHCO_3$, brine, dried over $MgSO_4$ and concentrated to give compound **12B**. M+1 = 319.0



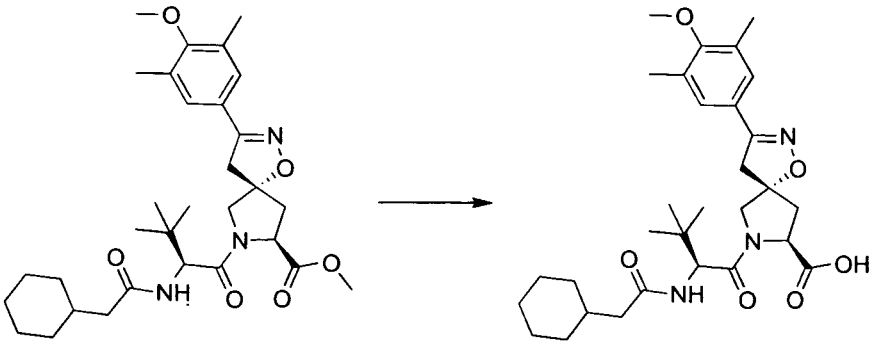
12B **12C**

[0454] Compound **12B** (727 mg, 2.28 mmol) was dissolved in DMF (3 mL) with Boc-t-butylglycine (686 mg, 3.0 mmol), EDC•HCl (659 mg, 3.43 mmol), HOBt (460 mg, 3.4 mmol), and DIEA (1.2 mL, 6.89 mmol) and stirred at room temperature overnight. The reaction was then transferred to a separatory funnel and diluted with EtOAc. The organic layer was washed with 1 N HCl (twice, 20 mL each), sat. aq. $NaHCO_3$ (25 mL), water (10 mL), brine (10 mL), dried over $MgSO_4$ and concentrated. The crude product **12C** was purified over silica gel on an ISCO Combiflash with EtOAc/Hexanes as eluent to yield 231 mg (0.435 mmol) of compound **12C** as a clear colorless oil. LCMS (M+1) = 532.45



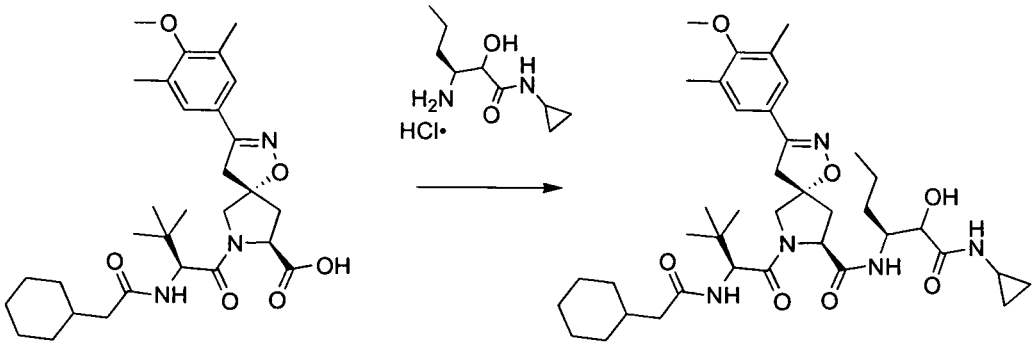
12C **12D**

[0455] Compound **12C** (231 mg, 0.435 mmol) was stirred in 4N HCl in dioxane (15 mL) for 90 minutes at which point TLC analysis indicated no starting material was present in the reaction mixture. The HCl and dioxane were evaporated to yield an off-white foam. A portion of this intermediate (0.35 mmol), EDC•HCl (96 mg, 0.50 mmol.), HOBt (72 mg, 0.53 mmol), and cyclohexaneacetic acid (78 mg, 0.55 mmol) were stirred in DMF (3.5 mL). To this was added DIEA (0.18 mL, 1.0 mmol) and the reaction was stirred overnight. The reaction was then diluted with EtOAc and transferred to a separatory funnel where the layers were separated and the organic phase was washed with 1.0 N HCl, saturated aq. $NaHCO_3$, brine, dried over $MgSO_4$ and concentrated. The product was purified over silica gel on an ISCO Combiflash with EtOAc/hexane as eluent to yield 219 mg (0.394 mmol) of compound **12D** as a clear oil. M+1 = 556.4



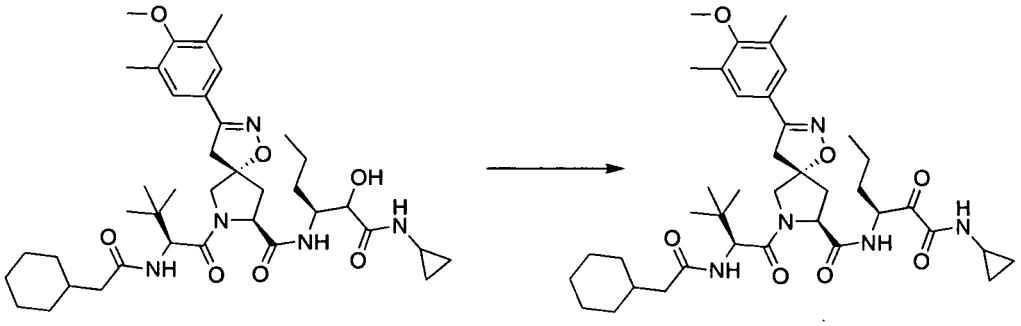
12D **12E**

[0456] Compound **12D** (219 mg, 0.394 mmol) in THF/H_2O/MeOH (4:1:1, 6 mL) was stirred with LiOH•H_2O (1.5 eq.) at room temperature overnight. The reaction was then acidified with 1.0 N HCl and extracted with CH_2Cl_2. The organic layer was washed with brine, dried over $MgSO_4$ and concentrated to yield 207 mg (0.382 mmol) of compound **12E**. M+1 = 548.4



12E **12F**

[0457] Compound **12E** (207 mg, 0.382 mmol) was stirred with HOBt (107 mg, 0.792 mmol), EDC•HCl (144 mg, 0.764 mmol), and hydroxyamine hydrochloride (168 mg, 0.75 mmol) in DMF (2.0 mL) at room temperature and treated with DIEA (0.400 mL, 2.3 mmol). The reaction was stirred overnight, diluted with EtOAc, washed with 1N HCl, saturated $NaHCO_3$, and the combined aqueous layers were back extracted with EtOAc. The organic layers were combined, dried over $MgSO_4$, concentrated and purified over silica gel on an ISCO combiflash with EtOAc/Hexanes as eluent to yield 227 mg (0.320 mmol) of compound **12F** as a white solid. (M+TFA) M–1 = 822.6.

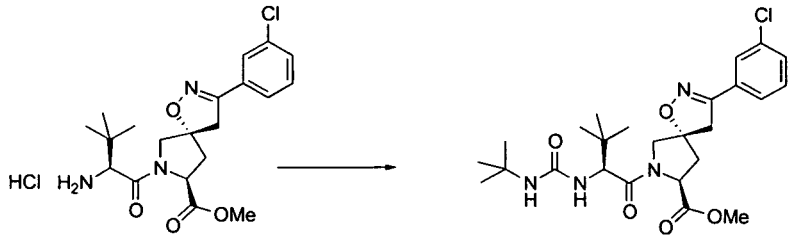


12F **Compound No. 362**

[0458] Compound **12F** (227 mg, 0.320 mmol) was dissolved at room temperature in CH_2Cl_2 (4 mL) and treated with Dess-Martin periodinane (142 mg, 1.0 eq.). After 15 minutes, TLC showed the reaction to be complete, and the reaction solution was quenched by the addition of water and stirred vigorously. Additional CH_2Cl_2 was added, the organic layer was separated and purified over silical gel on an ISCO combiflash with EtOAc/Hexanes as eluent to yield 159 mg (0.225 mmol) of Compound No. **362**. FIA MS (M+1) = 708.42. ^{1}H-

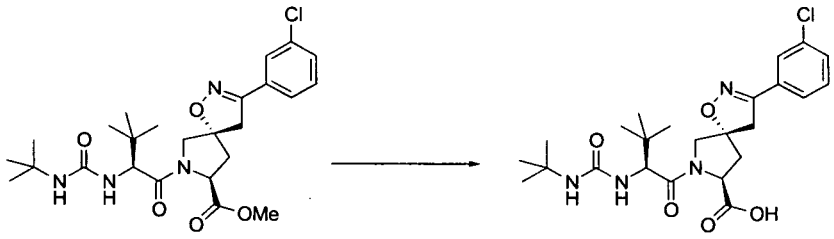
NMR (500 MHz, $CDCl_3$): 7.30 (s, 2H), 7.17 (d, 1H), 6.93 (d, 1H), 6.15 (d, 1H), 5.39-5.33 (m, 1H), 4.72 (t, 1H), 4.66 (d, 1H), 4.25 (d, 1H), 3.74 (s, 3H), 3.74-3.69 (m, 1H), 3.42 (d, 1H), 3.30 (d, 1H), 2.81-2.75 (m, 1H), 2.58-2.46 (m, 2H), 2.29 (s, 6H), 2.16-2.10 (m, 1H), 2.08-2.00 (m, 1H), 1.97-1.88 (m, 1H), 1.85-1.57 (m, 8 H), 1.51-1.35 (m, 2H), 1.33-1.22 (m, 2H), 1.20-1.07 (m, 1H), 1.02-0.96 (m, 10H), 0.92 (t, 3H), 0.88-0.80 (m, 2H), 0.66-0.56 (m, 2H).

Example 13: Compound No. 247



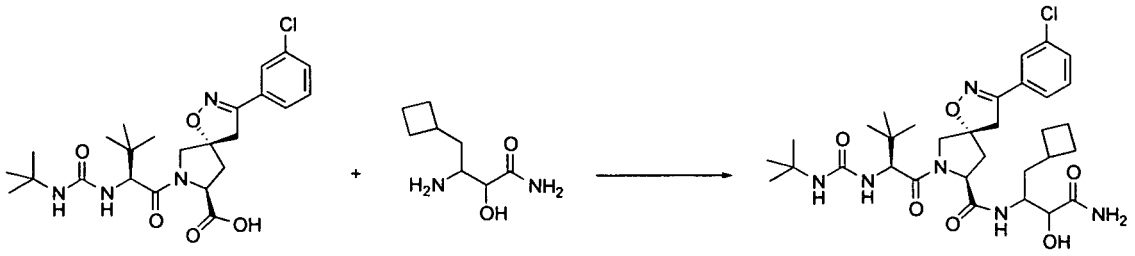
13A **13B**

[0459] To a solution of compound **13A** (222 mg, 0.5 mmol) was added TEA (0.14 mL) and t-butylisocyanate (0.6 mmol). The resulting solution was stirred overnight and then diluted with EtOAc (20 mL), washed with water (10 mL), dried over Na_2SO_4 and concentrated in vacuo. The crude product was purified chromatography on silica gel to afford compound **13B** as a white solid (190 mg). HPLC 8.48 min; LC-MS m/z 507.2 ES^+.



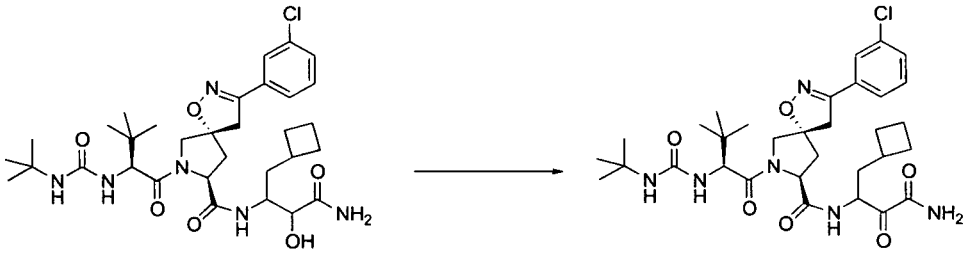
13B **13C**

[0460] Compound **13B** was dissolved in THF and the solution was treated with 1.0 N aqueous LiOH and water. The reaction mixture was stirred for 1 hour, and concentrated in vacuo. The residue was then diluted with water, washed with Et_2O and acidified with 1 N aqueous HCl. The resulting mixture was extracted twice with CH_2Cl_2 and the combined organics were dried over $MgSO_4$, filtered and concentrated in vacuo to give crude compound **13C** which was used without further purification for the next step. LC-MS m/z 493.22 ES^+, 491.21 ES^-.



13C D 13D

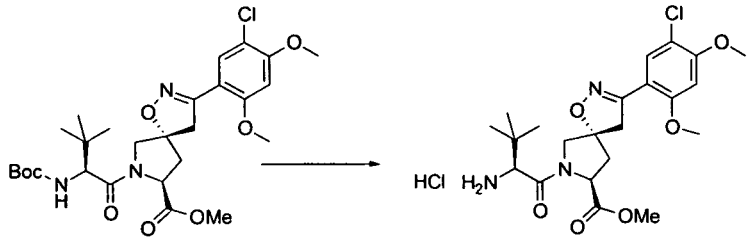
[0461] A solution of compound **13C** (20.6 mg) in CH_2Cl_2 (800 μL) was treated with 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (10 mg) and hydroxybenzotriazole (7 mg) for 1 hour. Diisopropylamine (16 μL) and 3-amino-4-cyclobutyl-2-hydroxybutanamide **D** (10.5 mg) were then added in one portion and the resulting reaction solution was stirred at room temperature for another 16 hours. The mixture was then washed with 1N aqueous HCl, 1:1 solution of 1N aqueous K_2CO_3: 1N aqueous $NaHCO_3$, and brine in succession. The organics were then dried ($MgSO_4$), concentrated in vacuo and purified by chromatography over silica (0% to 4% MeOH in CH_2Cl_2) to yield compound **13D** (11.6 mg). LC-MS m/z 647.25 ES^+.



13D Compound No. 247

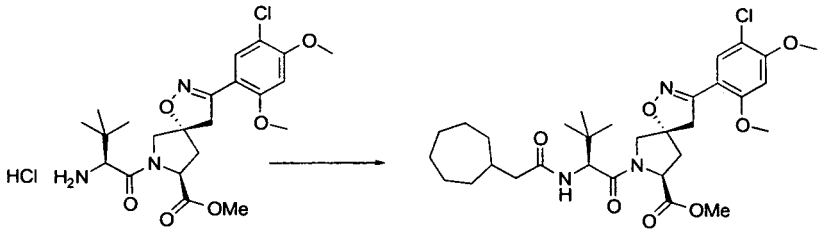
[0462] A solution of compound **13D** (11.6 mg) in CH_2Cl_2 (1 mL) was charged with Dess-Martin periodinane (8.4 mg) and the reaction mixture was stirred at room temperature for 2 hours. The resulting white mixture was then washed with 1.0 N aqueous $Na_2S_2O_3$, the phase were separated and the organics were the dried over $MgSO_4$, concentrated in vacuo and purified by chromatography over silica (30% to 65% EtOAc in hexanes) to yield 6.7 mg of Compound No. **247** as a white solid: ^{1}H-NMR (500 MHz, $CDCl_3$): 7.61 (s), 7.52 (d, J=6.1 Hz), 7.39 (d, J=7.8 Hz), 7.34 (t, J=7.8 Hz), 6.87 (s), 6.77 (s), 5.89 (s), 5.67 (s), 5.23-5.19 (m), 4.83-4.79 (m), 4.47 (s), 4.38 (d, J=11.0 Hz), 3.72 (dd, J=3.1, 11.2 Hz), 3.45 (m), 3.30 (d), 2.64 (m), 2.56 (m), 2.44-2.36 (m), 2.08-1.98 (m), 1.86-1.68 (m), 1.64-1.58 (m), 1.33-1.22 (m), 1.05-1.00 (m, H), 0.95-0.92 (m, H) ppm. LC-MS m/z 647.25 ES^+.

Example 14: Compound No. 57



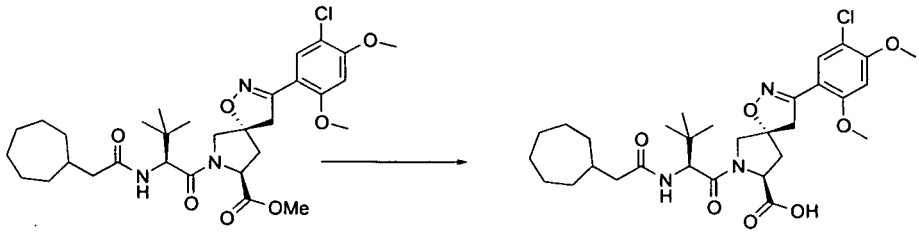
14A 14B

[0463] A solution of compound **14A** (512 mg) in dioxane was treated with 4 N HCl in dioxane. The reaction solution was stirred at room temperature for 45 minutes and concentrated in vacuo. The resulting residue was dissolved in a small amount of CH_2Cl_2 and crystallized from Et_2O/Hexanes to give compound **14B** as a white solid (362 mg). LC-MS m/z 468.24 ES^+.



14B 14C

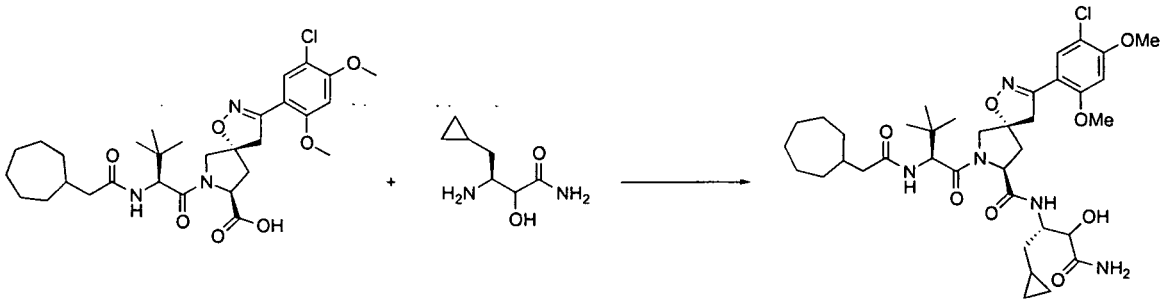
[0464] A solution of cycloheptane acetic acid (83 mg, Aldrich Chemical Co., Milwaukee, Wisc.) in CH_2Cl_2 (4 mL) was treated with 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (103 mg) and hydroxybenzotriazole (72 mg) for 1hour. Diisopropylamine (160 µL) and intermediate **14B** (179 mg) were then added in one portion and the resulting reaction solution was stirred at room temperature for another 2 hours. The mixture was then washed with 1 N aqueous HCl, 1:1 solution of 1 N aqueous K_2CO_3: 1 N aqueous $NaHCO_3$, and brine in succession. The organics were the dried ($MgSO_4$), concentrated in vacuo and purified by chromatography over silica (15% to 60% EtOAc in hexanes) to yield compound **14C** (188 mg). LC-MS m/z 606.25 ES^+.



14C 14D

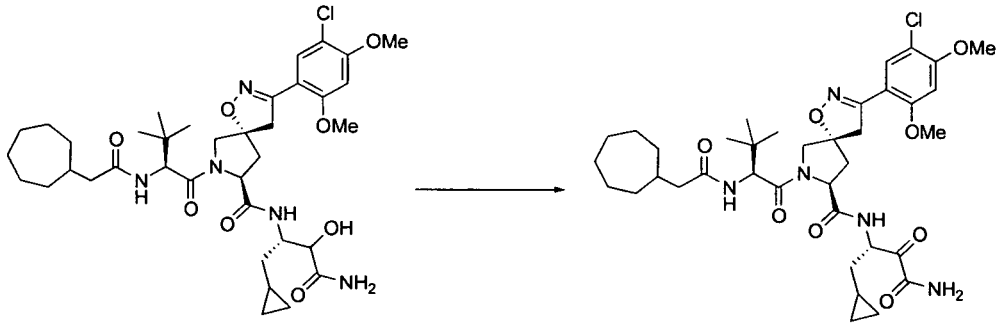
[0465] Compound **14C** (186 mg) was dissolved in THF (3 mL) and the solution was treated with 1 N aqueous LiOH (620 µL) and water (1 mL). The reaction mixture was stirred for 45 minutes at room temperature, and concentrated in vacuo. The residue was then diluted

with water, washed with Et_2O and acidified with 1 N aqueous HCl. The resulting mixture was extracted twice with EtOAc and the combined organics were dried over $MgSO_4$, filtered and concentrated in vacuo to give crude compound **14D** which was used without further purification for the next step. LC-MS m/z 592.25 ES^+, 590.35 ES^-.



14D **14F**

[0466] A solution of compound **14D** (89 mg) in CH_2Cl_2 (1 mL) / DMF (1 mL) was treated with 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (44 mg) and hydroxybenzotriazole (31 mg) for 1 hour. Diisopropylamine (70 μL) and (3S)-3-amino-4-cyclopropyl-2-hydroxybutanamide (35 mg) were then added in one portion and the resulting reaction solution was stirred at room temperature for another 16 hours. The mixture was then washed with 1 N HCl, 1:1 solution of 1N aqueous K_2CO_3: 1N aqueous $NaHCO_3$, and brine in succession. The organics were the dried over $MgSO_4$, concentrated in vacuo and purified by chromatography over silica (0% to 5% MeOH in CH_2Cl_2) to yield 96 mg of compound **14F**. LC-MS m/z 732.21 ES^+.

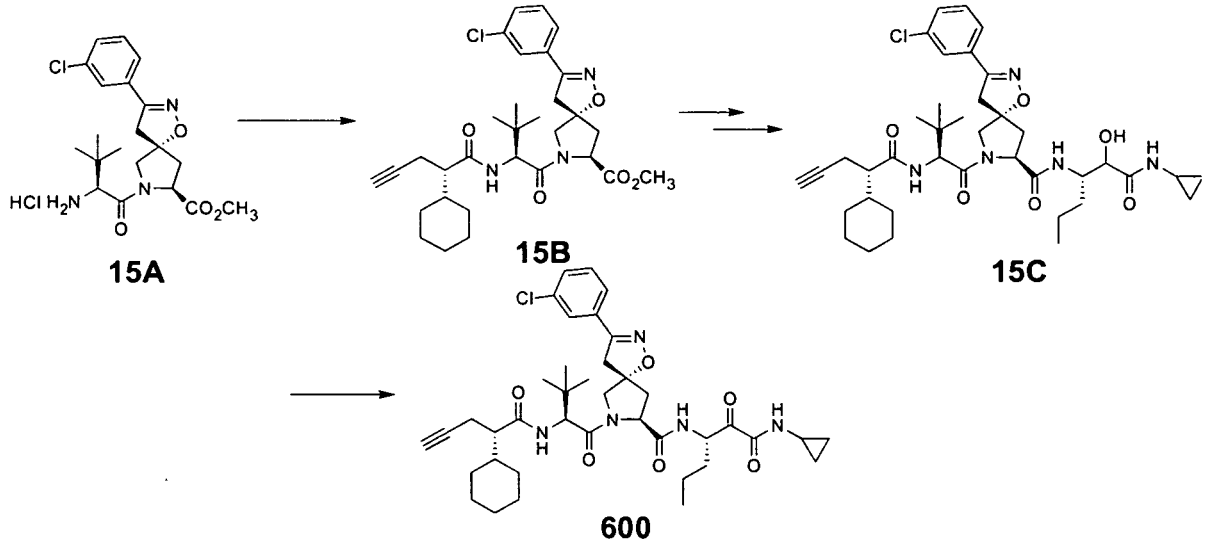


14F **Compound No. 57**

[0467] A solution of compound **14F** (96 mg) in CH_2Cl_2 (1.5 mL) was charged with Dess-Martin periodinane (83 mg) and the reaction mixture was stirred at room temperature for 2 hours. The resulting white mixture was then washed with 1 N aqueous $Na_2S_2O_3$, the phase were separated and the organics were the dried over $MgSO_4$, concentrated in vacuo and purified by chromatography over silica (10% to 95% EtOAc in hexanes) to yield Compound No. **57** (44 mg) as a white solid. ^{1}H-NMR (500 MHz, $CDCl_3$): 7.76 (s), 6.75 (br s), 6.48 (s), 6.07 (d), 5.40 (m), 4.67 (m), 4.22 (d), 3.95 (s), 3.87 (s), 3.75 (d), 3.43 (m), 2.51 (m), 2.10 (m),

1.30-1.87 (m), 1.12-1.28 (m), 0.97 (m), 0.79 (m), 0.15 (m), 0.03 (m) ppm. LC-MS m/z 730.35 ES^+, 728.35 ES^-.

Example 15: Compound No. 600



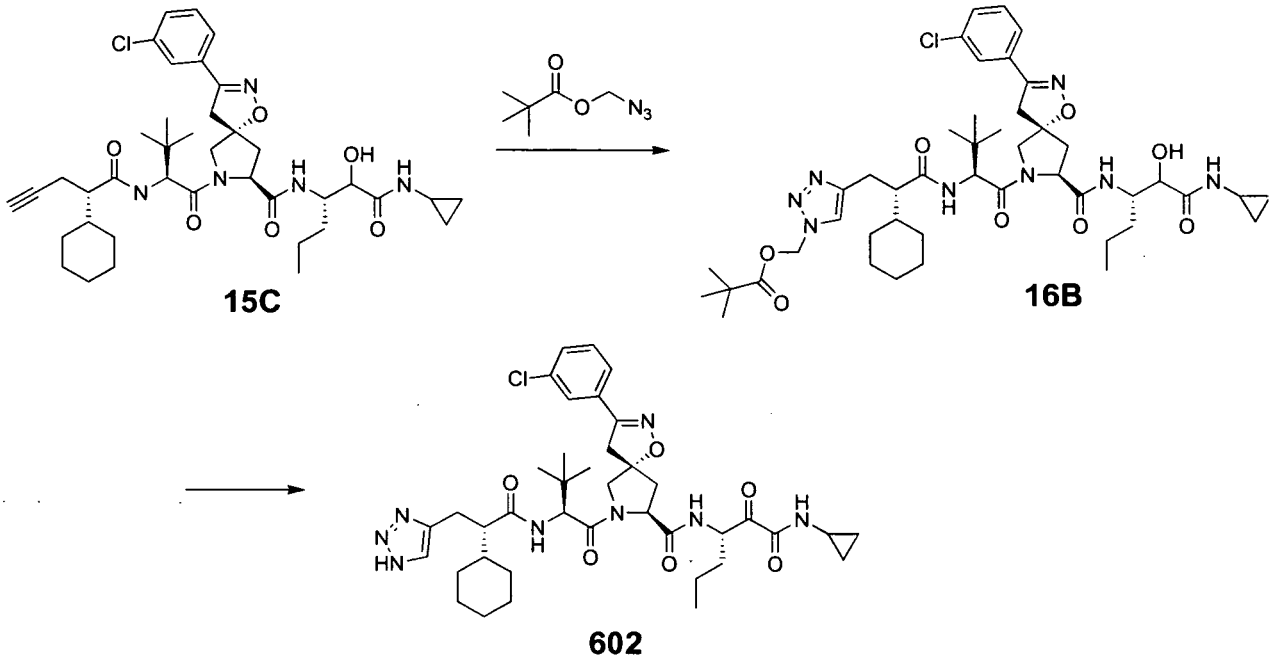
[0468] Compound 600 has the same structure as compound 266 in Table 1.

[0469] To a solution of (R)-2-cyclohexylbut-3-ynoic acid (430 mg, 2.4 mmol) in CH_2Cl_2 (10 mL) was added EDC (458 mg, 2.4 mmol), HOBt (324 mg, 2.4 mmol) and triethylamine (836 µL, 6.0 mmol). After stirring for 5 minutes, compound **15A** (800 mg, 2.0 mmol) was added and the reaction was stirred 16 hours. The mixture was washed with H_2O, 1 N HCl, and saturated $NaHCO_3$ solution. The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure to provide 1.23 g crude compound **15B**, which was purified by silica gel chromatography. ES (+) MS: m/e 570 $(M + H)^+$.

[0470] To a solution of compound **15B** (220 mg, 0.4 mmol) in THF / H_2O (2 mL, 3:1 v/v) was added LiOH monohydrate (115 mg, 3 mmol). The mixture was stirred for 2 hours, acidified using 1 N HCl (6 mL) and extracted with EtOAc (thrice, 10 mL). The combined organics were dried over $MgSO_4$ and concentrated to afford a colorless oil which was used without further purification. The oil was dissolved in CH_2Cl_2 (2 mL), then EDC (90 mg, 0.5 mmol), HOBt (63 mg, 0.5 mmol) and triethylamine (163 µL, 1.2 mmol) were added. After stirring for 5 minutes, (3S)-3-amino-N-cyclopropyl-2-hydroxyhexanamide (87 mg, 0.5 mmol) was added. The reaction was stirred 12 hours, washed with H_2O, 1 N HCl, and saturated $NaHCO_3$ solution. The organic layer was dried over $MgSO_4$ and concentrated under reduced pressure to provide 215 mg of compound **15C** as a colorless oil, which was used without further purification. ES (+) MS: m/e 724 $(M + H)^+$.

[0471] To a solution of compound **15C** (53 mg, 0.07 mmol) in CH_2Cl_2 (0.5 mL) was added Dess-Martin periodinane (41 mg, 0.1 mmol). The mixture was stirred for 30 minutes, quenched with 1 $Na_2S_2O_3$, and separated. The organic layer was purified by silica gel chromatography to provide 20 mg of Compound No. **600**. ^{1}H-NMR (500 MHz, $CDCl_3$): 7.53 (d, J = 1.6 Hz, 1H), 7.43 (d, J = 7.6 Hz, 1H), 7.31-7.25 (m, 2H), 6.83 (d, J = 3.3 Hz, 1H), 6.24-6.21 (m, 1H), 5.30-5.26 (m, 1H), 4.70-4.58 (m, 2H), 4.23-4.21 (m, 1H), 3.64 (dd, 1H), 3.36-3.20 (m, 2H), 2.70-2.68 (m, 1H), 2.57-2.35 (m), 2.04-1.82 (m), 1.72-1.30 (m, 10H), 1.18-0.75 (m), 0.55-0.40 (m).

Example 16: Compound No. 602



[0472] Compound 602 has the same structure as compound 212 in Table 1.

[0473] To a solution of compound **15C** prepared above (20 mg, 0.03 mmol) and azidomethyl pivalate (4 mg, 0.03 mmol, prepared according to Syn. Lett., 2005, 18, pp. 2847-2850) in tert-butanol / H_2O (120 μL, 1:1 v/v) was added an aqueous solution of sodium ascorbate (10 μL, 0.01 mmol, 1.0 M) followed by an aqueous solution of copper(II) sulfate pentahydrate (5 μL, 0.001 mmol, 0.3 M). The reaction mixture was stirred 12 hours at room temperature, diluted with H_2O, and extracted with EtOAc. The combined organics were washed with 5% ammonium hydroxide followed by brine, and were dried over $MgSO_4$ and concentrated under reduced pressure to provide 25 mg of crude compound **16B**, which was used without further purification. ES (+) MS: m/e 881 $(M + H)^+$.

[0474] To a solution of compound **16B** in MeOH (120 μL) was added aqueous NaOH (120 μL, 1 M). The reaction was stirred at room temperature for 2 hours, then treated with 1 M HCl (120 μL) followed by H_2O (120 μL). The mixture was extracted with CH_2Cl_2 (thrice, 200 μL each). The combined extracts were washed with brine and concentrated to a volume of approximately 100 μL. To this solution was added Dess-Martin periodinane (17 mg, 0.04 mmol) and the reaction was stirred 30 minutes. The mixture was quenched with 1 M $Na_2S_2O_3$ (150 μL), and the organic layer was separated and purified via silica gel chromatography to afford afford 3 mg of Compound No. **602**. ES (+) MS: m/e 765 $(M + H)^+$.

[0475] Listed below in Table 6 are additional compounds of Formula I prepared by Methods 5a and 5b.

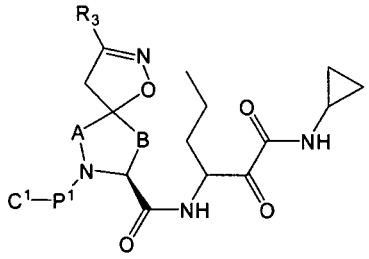


Table 6: Additional compounds of formula I produced by methods 5a and 5b.

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
5	N-BOC-L-tert-butylglycine	2-(4-hydroxy-4-methylcyclohexyl)acetic acid	3-Chlorobenzaldoxime
10	(S)-4-(benzylamino)-2-isopropyl-4-oxobutanoic acid	N/A	3-chlorobenzaldoxime
13	N-BOC-L-tert-butylglycine	2-Norbornaneacetic acid	3-chlorobenzaldoxime
19	N-BOC-L-tert-butylglycine	2-(bicyclo[4.1.0]heptan-1-yl)acetic acid	3-chlorobenzaldoxime
21	(S)-4-(cyclohexylamino)-2-isopropyl-4-oxobutanoic acid	N/A	3-chlorobenzaldoxime
23	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
26	N-BOC-L-tert-butylglycine	N-Benzoyl-L-Proline	3-Chlorobenzaldoxime
27	N-BOC-L-tert-butylglycine	Cyclobutaneacetic acid	3-Chlorobenzaldoxime
43	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
48	N-BOC-L-tert-butylglycine	2-Norbornaneacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
50	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
59	N-BOC-L-tert-butylglycine	2-cycloheptylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
63	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
67	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chloro-5-fluoro-4-ethoxybenzaldoxime
86	N-BOC-L-tert-butylglycine	Isopropyl isocyanate	Piperonal oxime
90	N-BOC-L-tert-butylglycine	N/A	3-Chlorobenzaldoxime
104	N-BOC-L-tert-butylglycine	Tert-butylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
105	N-BOC-L-tert-butylglycine	N/A	3-Chlorobenzaldoxime
117	N-BOC-L-tert-butylglycine	4-methyltetrahydro-2H-pyran-4-carboxylic acid	3-Chlorobenzaldoxime
121	N-BOC-L-tert-butylglycine	2-(2,2-dimethyltetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
126	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-5-fluoro-4-ethoxybenzaldoxime
129	N-BOC-L-tert-butylglycine	2-cycloheptylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
131	N-BOC-L-tert-butylglycine	N/A	2,4-Dimethoxy-5-chlorobenzaldoxime
136	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-5-fluoro-4-ethoxybenzaldoxime
145	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran4-yl)acetic acid	Piperonal oxime
153	N-BOC-L-tert-butylglycine	2-((2R,5R)-2,5-dimethyltetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
168	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	Thiophene-3-carboxaldehyde
172	N-Phenyl-L-tert-butylglycine	N/A	3-chlorobenzaldoxime
178	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chloro-5-fluoro-4-ethoxybenzaldoxime
184	N-BOC-L-tert-butylglycine	4-methyltetrahydro-2H-pyran-4-carboxylic acid	3-Chlorobenzaldoxime
188	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
192	N-BOC-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	2,4-Dimethoxy-5-chlorobenzaldoxime
195	N-BOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-chloro-4-methoxy-5-methylbenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
211	2-(tert-butoxycarbonylamino)-2-(1-methoxycyclopropyl)acetic acid	2-cyclohexylacetic acid	3-chlorobenzaldoxime
212	N-BOC-L-tert-butylglycine	(S)-2-cyclohexyl-3-(1H-1,2,3-triazol-4-yl)propanoic acid	3-chlorobenzaldoxime
214	N-(3-methoxyphenyl)-L-tert-butylglycine	N/A	3-chlorobenzaldoxime
217	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
219	N-BOC-L-tert-butylglycine	2-cycloheptylacetic acid	3-Chlorobenzaldoxime
225	N-BOC-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	3-Chlorobenzaldoxime
231	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
233	N-BOC-L-tert-butylglycine	2-(1-hydroxycyclohexyl)acetic acid	3-Chlorobenzaldoxime
247	N-BOC-L-tert-butylglycine	tert-Butyl isocyanate	3-Chlorobenzaldoxime
256	N-BOC-L-tert-butylglycine	2-cyclohexylacetic acid	5-Ethyl-2-furaldoxime
263	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-5-fluoro-4-ethoxybenzaldoxime
264	N-BOC-L-tert-butylglycine	N/A	2,4-Dimethoxy-5-chlorobenzaldoxime
266	N-BOC-L-tert-butylglycine	(S)-2-cyclohexylpent-4-ynoic acid	3-chlorobenzaldoxime
268	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
273	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
280	(S)-2-isopropyl-4-(isopropylamino)-4-oxobutanoic acid	N/A	3-chlorobenzaldoxime
282	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	2,4-Dimethoxy-5-Chlorobenzaldoxime
284	N-BOC-L-tert-butylglycine	(S)-2-cyclohexylpropanoic acid	3-chlorobenzaldoxime
286	N-BOC-L-tert-butylglycine	2-(4-methyltetrahydro-2H-pyran-4-yl)acetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
290	N-CBZ-L-tert-butylglycine	N/A	Piperonal oxime
294	N-BOC-L-tert-butylglycine	2-cyclohexylacetic acid	3-chlorobenzaldoxime
295	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	Piperonal oxime

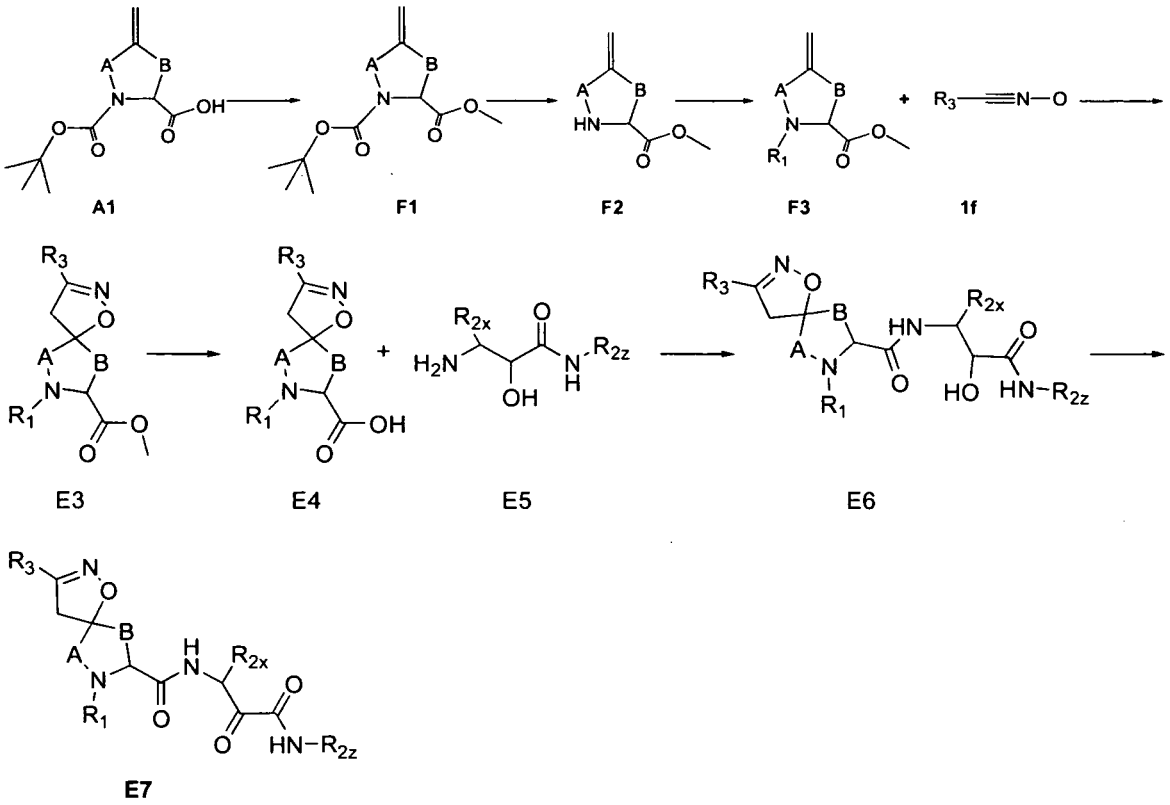
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
297	N-BOC-L-tert-butylglycine	Tert-butylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
307	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
310	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3,5-Dimethyl-4-methoxybenzalehyde
326	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	2,4-Dimethoxy-5-Chlorobenzaldoxime
335	N-CBZ-L-tert-butylglycine	N/A	2,4-Dimethoxybenzaldoxime
337	N-BOC-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	3-Chlorobenzaldoxime
344	N-BOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	3-Chlorobenzaldoxime
346	N-BOC-L-tert-butylglycine	2-Norbornaneacetic acid	3-Chlorobenzaldoxime
351	N/A	Tert-butylacetic acid	3-chlorobenzaldoxime
356	N-BOC-L-tert-butylglycine	2-(4-methyltetrahydro-2H-pyran-4-yl)acetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
362	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3,5-Dimethyl-4-methoxybenzalehyde
369	N-BOC-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	3-Chlorobenzaldoxime
375	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	Piperonal oxime
382	N-BOC-L-tert-butylglycine	isopropylisocyanate	Piperonal oxime
388	N-BOC-L-tert-butylglycine	Cyclohexylacetic acid	3-Chlorobenzaldoxime
411	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chloro-5-fluoro-4-ethoxybenzaldoxime
415	N-CBZ -L-tert-butylglycine	N/A	Piperonal oxime
418	N-BOC-L-tert-butylglycine	2-((2S,5R)-2,5-dimethyltetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
419	N-BOC-L-tert-butylglycine	2-(2,2-dimethyltetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
440	N-BOC-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	2,4-Dimethoxy-5-chlorobenzaldoxime
442	N-BOC-L-tert-butylglycine	2-((2S,5R)-2,5-dimethyltetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
445	N-BOC-L-tert-butylglycine	2-(1,4-dioxaspiro[4.5]decan-8-yl)acetic acid	3-Chlorobenzaldoxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
446	N-BOC-L-tert-butylglycine	2-Norbornaneacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
453	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
468	N-BOC-L-tert-butylglycine	2-((2R,5R)-2,5-dimethyltetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
473	N-BOC-L-tert-butylglycine	Tert-butylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
485	N-BOC-L-tert-butylglycine	trans-2-phenyl-1-cyclopropanecarboxylic acid	3-Chlorobenzaldoxime
502	N-BOC-L-tert-butylglycine	N-FMOC-L-cyclohexylglycine followed by 2-pyrazine carboxylic acid	3-Chlorobenzaldoxime
510	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	2,4-Dimethoxy-5-Chlorobenzaldoxime
516	N-((S)-tetrahydrofuran-3-yloxy)carbonyl)-L-tert-butylglycine	N/A	2,4-Dimethoxybenzaldoxime
522	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chlorobenzaldoxime
529	N-BOC-L-tert-butylglycine	2-(4-methyltetrahydro-2H-pyran-4-yl)acetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
541	N-BOC-L-tert-butylglycine	2-(4-methyltetrahydro-2H-pyran-4-yl)acetic acid	3-chlorobenzaldoxime
542	N-BOC-L-tert-butylglycine	tert-Butyl isocyanate	3-chlorobenzaldoxime
549	N-BOC-L-tert-butylglycine	(S)-2-cyclohexyl-4-oxo-4-(pyrrolidin-1-yl)butanoic acid	3-chlorobenzaldoxime
554	N-BOC-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	5-Ethyl-2-furaldoxime
562	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
569	N-BOC-L-tert-butylglycine	(S)-2-cyclohexyl-4-(methylamino)-4-oxobutanoic acid	3-chlorobenzaldoxime
575	N-BOC-L-tert-butylglycine	2-(4-hydroxy-4-methylcyclohexyl)acetic acid	3-chlorobenzaldoxime
577	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2,4-Dimethoxy-5-chlorobenzaldoxime
581	N-BOC-L-tert-butylglycine	N/A	2,4-Dimethoxy-5-chlorobenzaldoxime
589	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-8-quinolinecarbaldoxime
590	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2-Methoxy-3-methylbenzaldoxime

[0476] Certain other compounds of Formula I may be prepared by Method 6 as

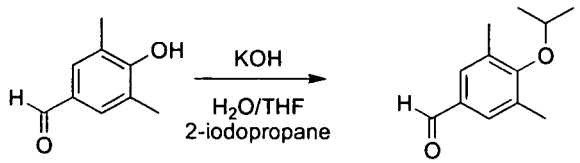
illustrated below.

METHOD 6:



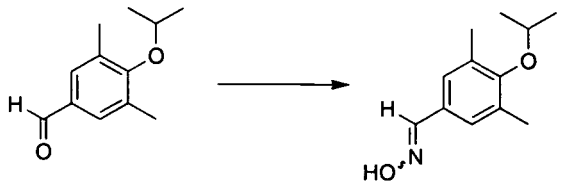
[0477] Referring to Method 6, the intermediate **A1** is converted to the Boc-methyl ester **F1**. Removal of the Boc group from F1 provides the amine-ester **F2** which is reacted with an R_1 carboxylic acid in the presence of a coupling reagent to provide **F3** wherein R_1 is $R_4C(O)$-. F3 reacts with a nitrile oxide **1f** to provide the spiroisoxazoline acid **E4** after hydrolysis of the corresponding methyl ester **E3**. Conversion of **E4** to **E7** is achieved as described in Method 5a.

Example 17: Compound No. 267

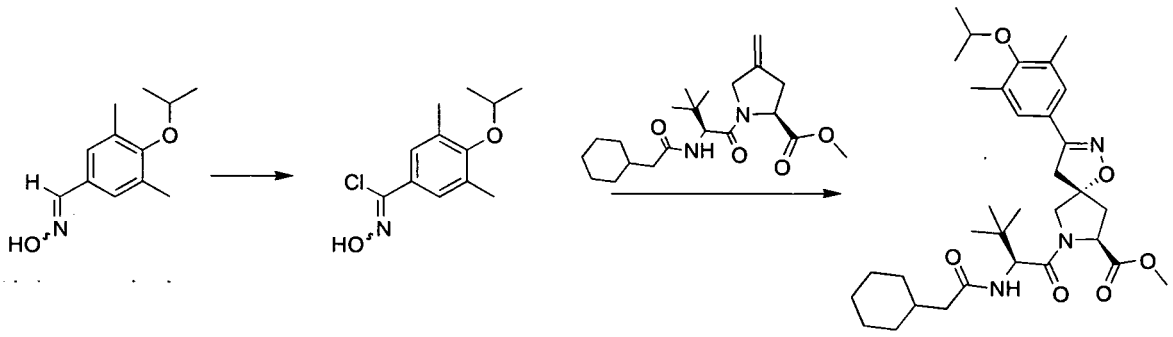


[0478] 4-Hydroxy-3,5-dimethylbenzaldehyde (2.5 g, 16.6 mmol) in THF (100 mL) was treated with KOH (1.5 eq. of 1 N aq. solution, 25 mL) and 2-iodopropane (2.0 eq.) and heated at reflux for 5 days. The reaction was then cooled, transferred to a separatory funnel, diluted with MTBE, washed with H_2O, 1 N NaOH (twice), 0.5 N HCl (aq.), brine, dried over $MgSO_4$ and concentrated. The product was purified over silica gel on an ISCO combiflash to yield 1.99 g (10.34 mmol) 4-isopropoxy-3,5-dimethylbenzaldehyde as a colorless liquid. H^1 NMR

(300 MHz, CDCl3) 9.89 (s, 1H), 7.55 (s, 2H), 4.41-4.26 (m, 1 H), 2.32 (s, 6H), 1.32 (d, J = 6 Hz, 6H).

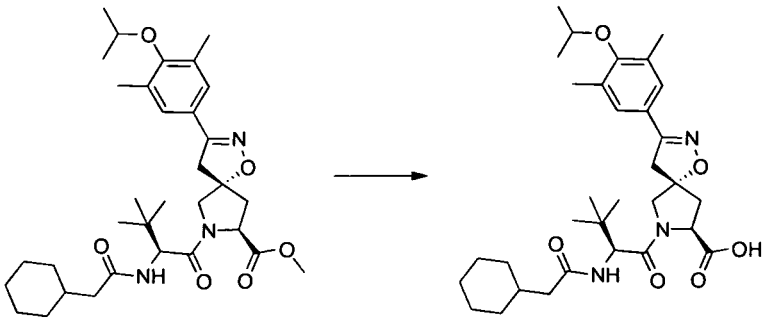


[0479] 4-(Isopropoxy)-3,5-dimethylbenzaldehyde (1.98 g, 10.3 mmol) in EtOH (60 mL) was heated to 60 °C with hydroxylamine hydrochloride (2.4 M aq. solution, 5.2 mL, 1.2 eq.) and Na_2CO_3 (1.2 M solution, 5.2 mL, 0.6 eq.) at room temperature for 2 hours. The reaction was transferred to a separatory funnel, diluted with EtOAc; the organic layer was separated, washed with brine, dried ($MgSO_4$), filtered and concentrated to yield 710 mg (3.24 mmol) of 4-(isopropoxy)-3,5-dimethylbenzaldehyde oxime as a light yellow oil. ^{1}H-NMR (500 MHz, $CDCl_3$): 8.10 (s, 1H), 7.23 (s, 2H), 4.29-4.18 (m, 1H), 2.29 (s, 6H), 1.29 (d, 6H).



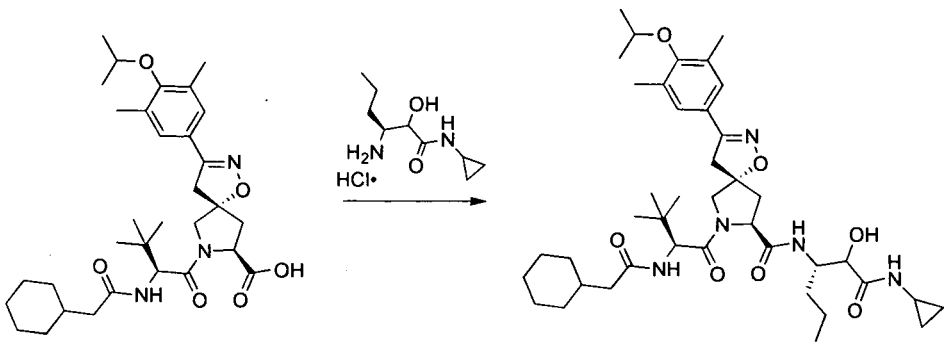
17A

[0480] 4-(Isopropoxy)-3,5-dimethylbenzaldehyde oxime (166 mg, 0.801 mmol) in DMF (3 mL) at room temperature was stirred overnight with NCS (130 mg, 0.974 mmol). To this reaction was added the methyl ester (257 mg, 0.679 mmol) in DMF (1.5 mL) and triethylamine (1.2 eq.). This was stirred overnight at room temperature. The reaction was then diluted with EtOAc/Hexanes (4:1) and washed with 1N HCl (aq.). The layers were separated and the aqueous layer was back extracted with EtOAc/Hexanes (4:1). The organic layers were combined, washed with brine, dried (MgSO4), and concentrated. The compound was purified over silica gel on an ISCO Combiflash with EtOAc/Hexanes as eluent to yield 173 mg (0.296 mmol) of compound **17A** as a white solid. LCMS (M + 1) = 584.3



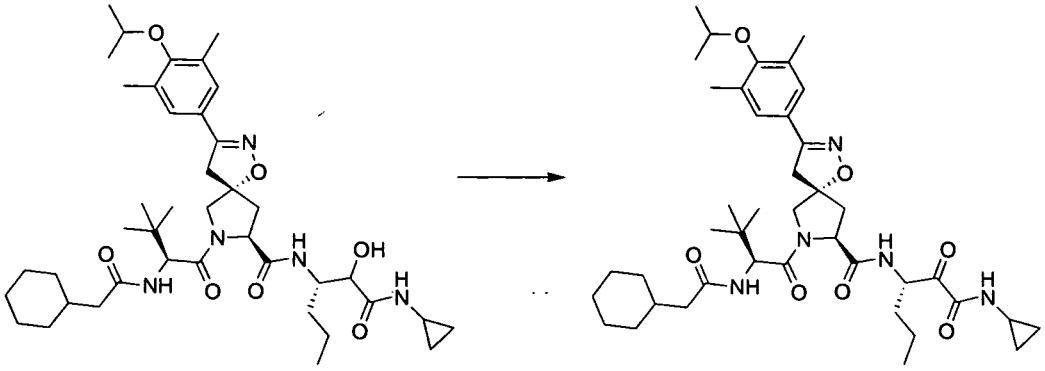
17A 17B

[0481] The compound **17A** (173 mg, 0.30 mmol) was stirred with $LiOH \cdot H_2O$ (1.1 eq.) in THF/MeOH/H_2O (4:1:1, 3 mL) at RT overnight. The reaction was diluted with EtOAc, acidified with 1N HCl (aq) and the layers were separated. The aqueous layer was back extracted with EtOAc, the organic layers combined, washed with brine, dried ($MgSO_4$) and concentrated to yield 171 mg (0.30 mmol) of compound **17B** as a white solid. FIA MS (M+1) = 570.3.



17B 17C

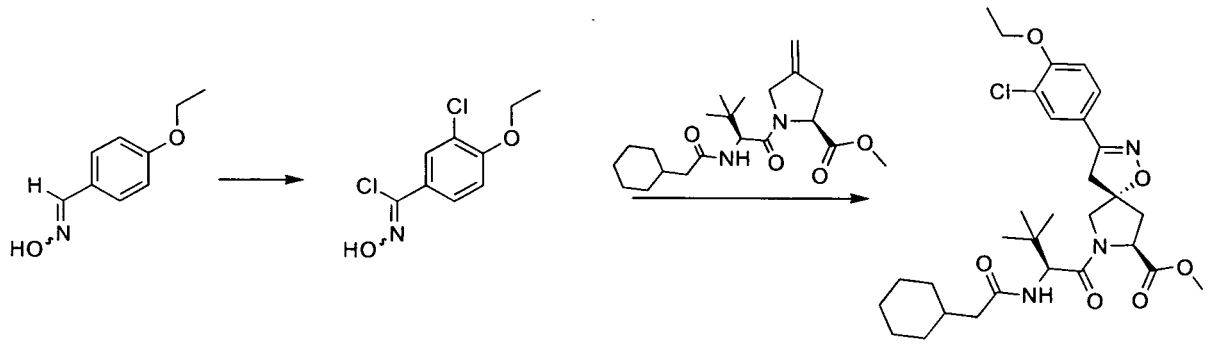
[0482] Carboxylic acid **17B** (83 mg, 0.146 mmol), EDC•HCl (37 mg, 1.3 eq.), HOBt (26 mg, 1.3 eq.), (3S)-3-amino-N-cyclopropyl-2-hydroxyhexanamide hydrochloride (64 mg, 2.0 eq.), and DIEA (0.100 mL, 4.0 eq.) were stirred in DMF (0.9 mL) at room temperature overnight. The reaction mixture was then diluted with EtOAc and washed with 1 N HCl (aq) (twice). The aqueous layer was separated and back extracted with EtOAc. The organic layers were combined, washed with brine, dried ($MgSO_4$), and concentrated. The product was purified over silica gel on an ISCO combiflash to yield 85 mg (0.115 mmol) of compound **17C**. LCMS (M + 1) = 738.3



17C **Compound No. 267**

[0483] Compound **17C** (85 mg, 0.115 mmol) in CH_2Cl_2 (1.0 mL) was treated with Dess-Martin periodinane (54 mg, 1.1 eq.) for 30 minutes. The reaction was quenched with equal volumes (≈1 mL) of saturated aqueous $NaHCO_3$ and 1 N $Na_2S_2O_3$ (aq). The organic layer was separated and purified directly over silica gel on an ISCO combiflash to yield 77 mg (0.105 mmol) of Compound No. **267**. FIA MS (M + 1) = 736.2. ^{1}H-NMR (300 MHz, $CDCl_3$): 7.33-7.26 (m, 2H), 7.12 (d, 1H), 6.91 (d, 1H), 6.12 (d, 1H), 5.45-5.32 (m, 1H), 4.78-4.63 (m, 2H), 4.29-4.17 (m, 2H), 3.71 (d, 1H), 3.43 (d, 1H), 3.30 (d, 1H), 2.86-2.74 (m, 1H), 2.63-2.42 (m, 2H), 2.29 (s, 6H), 2.19-1.85 (m, 3H), 1.84-0.82 (m, 34H), 0.65-0.58 (m, 2H).

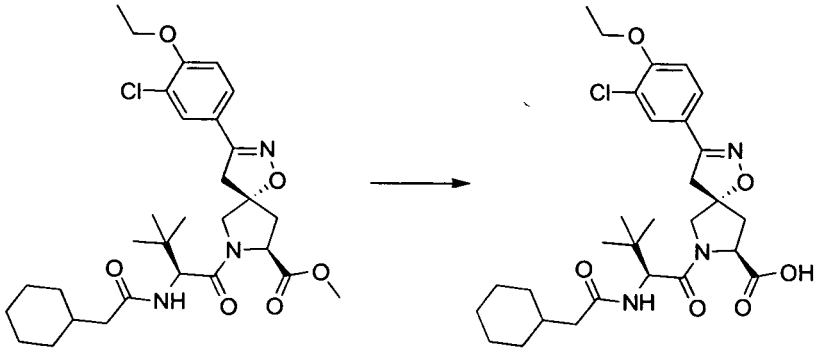
Example 18: Compound No. 556



18A

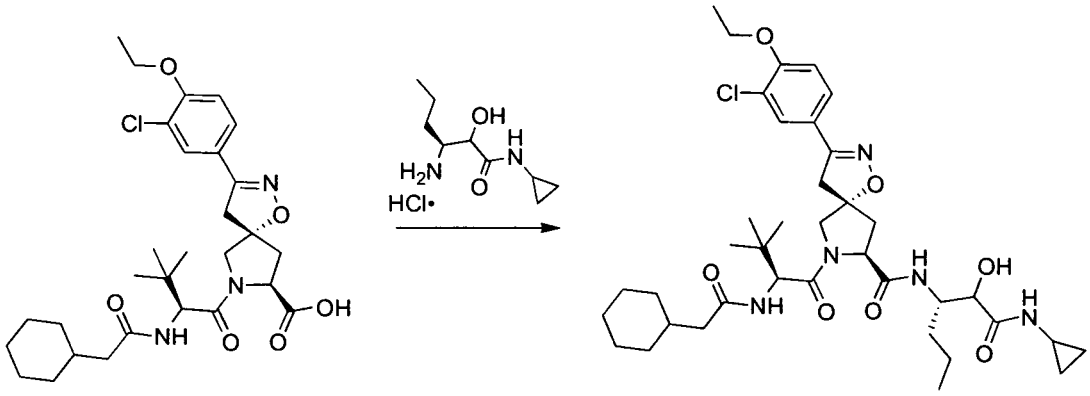
[0484] 4-Ethoxybenzaldehyde oxime (204 mg, 1.24 mmol), was dissolved in DMF (to 0.2 M) and treated with NCS (1 eq.). The reaction was stirred until starting material was consumed. One half of the reaction volume was removed and treated with additional NCS (1.5 eq.) and stirred overnight. To this solution was then added the methyl ester (200 mg, 0.85 eq.) in DMF (0.3 mL) and triethylamine (0.10 mL, 1.1 eq.). The reaction was stirred overnight at room temperature, then diluted with EtOAc, washed with 1 N HCl (aq.), and washed with brine. The aqueous layer was back extracted with EtOAc and the combined organic layers were washed with brine, dried ($MgSO_4$), and concentrated to a dark oil. The

product was purified over silica gel on an ISCO combiflash to yield 97 mg (0.168 mmol) of compound **18A**. LCMS (M+1) = 576.3



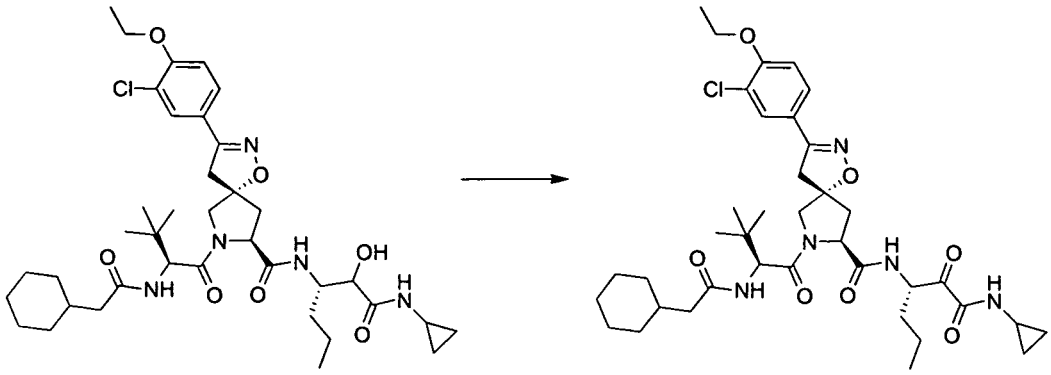
18A **18B**

[0485] Compound **18A** (97 mg, 0.168 mmol) was dissolved in THF/MeOH/H2O (8:1:1, 5 mL) and treated with LiOH•H2O (1.1 eq.) at room temperature overnight. The reaction was concentrated, diluted in EtOAc and methanol and washed with 1N HCl (aq). The aqueous layer was separated and extracted with EtOAc. The combined organic layers were washed with brine, dried over $MgSO_4$ and concentrated to yield 76 mg (0.135 mmol) of compound **18B**. FIA MS (M-1) = 560.4



18B **18C**

[0486] Compound **18B** (35 mg, 0.062 mmol), EDC•HCl (15 mg, 1.3 eq.), HOBt (12 mg, 1.3 eq.), an amino alcohol hydrochloride (55 mg, 2.0 eq.), and DIEA (0.044 mL, 4.0 eq.) were stirred in DMF (0.7 mL) at room temperature overnight. The reaction was then diluted with EtOAc and washed with 1 N HCl (aq) (twice). The aqueous layer was separated and back extracted with EtOAc. The organic layers were combined, washed with brine, dried ($MgSO_4$), anc concentrated. The product was purified over silica gel on an ISCO combiflash to yield 28 mg (0.038 mmol) of compound **18C**. LCMS (M+1) = 730.2



18C **Compound No. 556**

[0487] Compound **18C** (28 mg, 0.038 mmol) in CH_2Cl_2 (0.7 mL) was treated with Dess-Martin periodinane (18 mg, 1.1eq.) for 30 minutes. The reaction was quenched with equal volumes (≈1 mL) of saturated aqueous $NaHCO_3$ and 1N $Na_2S_2O_3$ (aq.). The organic layer was separated and purified directly over silica gel on an ISCO Optix 10x to yield 24 mg (0.033 mmol) of Compound No. **556**. FIA MS (M+1) = 728.2. ^{1}H-NMR (300 MHz, $CDCl_3$): 7.65 (d, 1H), 7.48 (dd, 1H), 7.11 (d, 1H), 6.95-6.88 (m, 2H), 6.08 (d, 1H), 5.40-5.31 (m, 2H), 4.78-4.63 (m, 2H), 4.26 (d, 1H), 4.20-4.11 (m, 2H), 3.71 (d, 1H), 3.42 (d, 1H), 3.27 (d, 1H), 2.84-2.73 (m, 1H), 2.63-2.46 (m, 2H), 2.20-1.86 (m, 3H), 1.62-0.85 (m, 30H), 0.66-0.58 (m, 2H)

[0488] Listed below in Table 7 are additional compounds of Formula I prepared by Method 6.

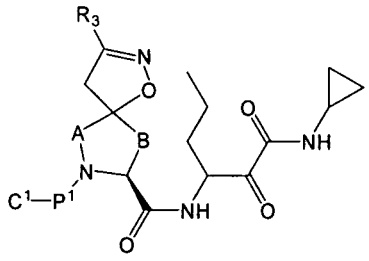


Table 7: Additional compounds of formula I prepared by method 6.

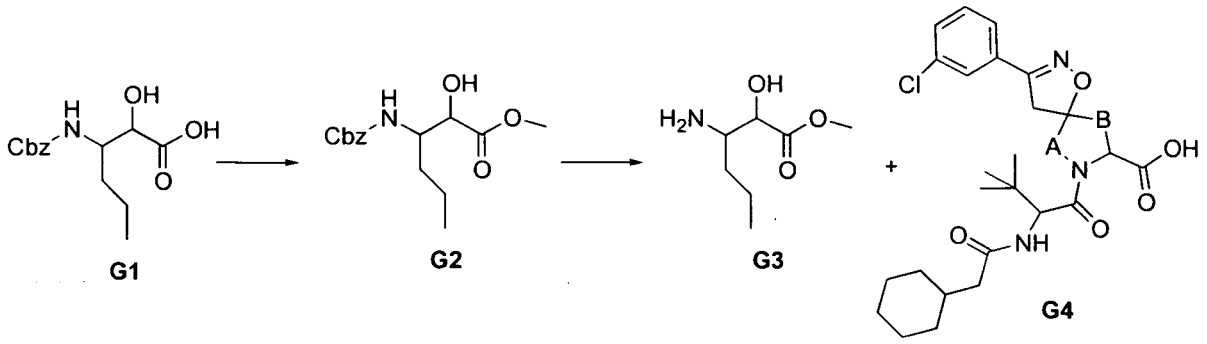
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
18	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	7-Chloro-2,3-dihydrobenzo[b]furan-5-carboxaldoxime
19	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-4-methylbenzaldoxime
28	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2-Cyanobenzaldoxime
31	N-BOC-L-tert-butylglycine	2-cyclohexylacetic acid	8-Quinoline-carbaldoxime
38	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2,5-Dichloro-3-methoxybenzaldoxime

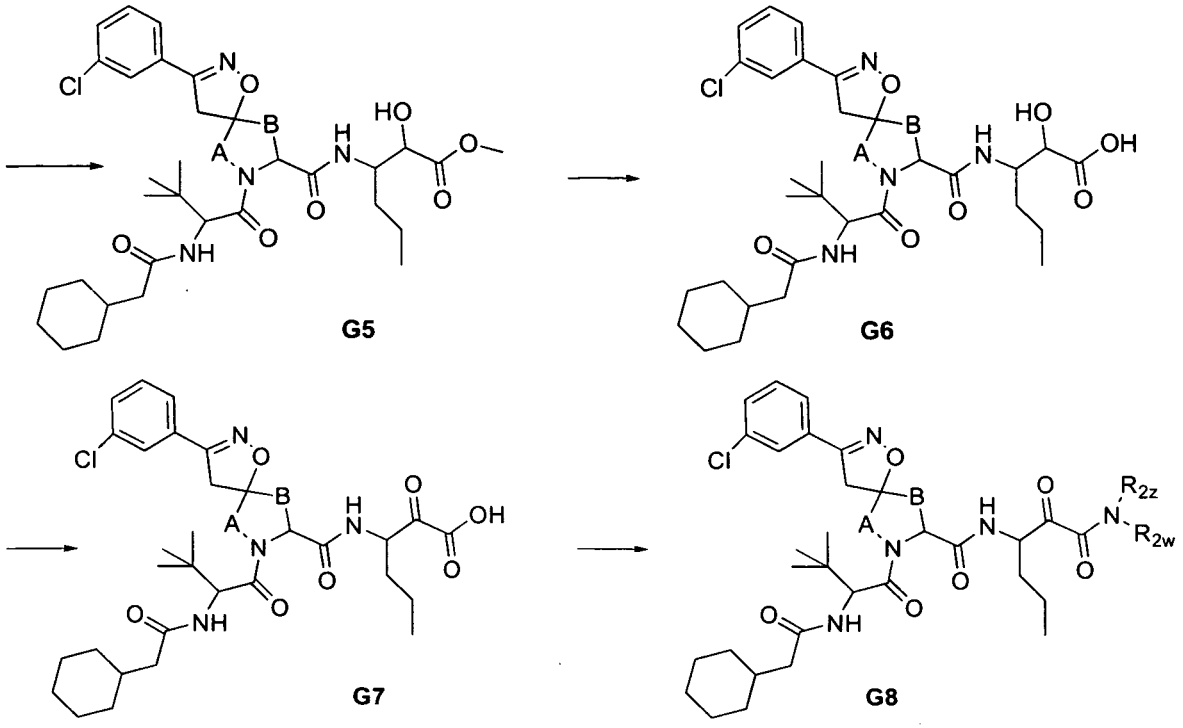
Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
42	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	8-Quinolinecarboxaldoxime
62	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-chloro-3-Thiophenecarboxaldoxime
68	N-BOC-L-tert-butylglycine	2-cyclohexylacetic acid	8-Quinoline-carbaldoxime
74	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	8-chloro-2,2-dimethylchromane-6-carbaldoxime
89	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-nitrobenzaldoxime
97	N-BOC-L-tert-butylglycine	2-cyclohexylacetic aicd	3-Chloro-4-isopropoxybenzaldoxime
111	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-4-methoxy-5-methylbenzaldoxime
114	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-4-methoxy-5-methylbenzaldoxime
132	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-nicotinaldoxime
134	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-2,3-dihydrobenzo[b]furan-7-carboxaldoxime
158	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-6-methoxybenzaldoxime
165	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-2-methoxynicotinaldoxime
168	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Thiophenecarboxaldoxime
169	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-2-fluorobenzaldoxime
170	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-2,2-dimethyl-2,3-dihydrobenzo[b]furan-7-carboxaldoxime
250	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-5-methoxybenzaldoxime
267	N-BOC-L-tert-butylglycine	2-cyclohexylacetic aicd	4-Isopropoxy-3,5-dimethylbenzaldoxime
292	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-nicotinaldoxime
305	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	6-Fluoro-1,3-benzodioxene-8-carbaldoxime
312	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-6-methoxynicotinaldoxime
315	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-4-Methyl-3,4-dihydro-2H-1,4-benzoxazine-7-carbaldoxime
321	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-2,3-dimethoxy-benzaldoxime
366	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-4-methoxy-2-methylbenzaldoxime
370	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-piperonal oxime

Compound No.	Starting Material for P^1	Starting Material for C^1	Starting Material for R_3
396	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-5-methylbenzaldoxime
406	N-BOC-L-tert-butylglycine	2-cyclohexylacetic aicd	4-Cyclopropylmethoxy-3,5-dimethylbenzaldoxime
430	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	8-Chloro-1-methyl-1,2,3,4-tetrahydro-quinoline-6-carbaldoxime
469	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2-Methoxy-nicotinaldoxime
478	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	5-Chloro-2-thiophenecarboxaldoxime
494	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-4,5-dimethoxybenzaldoxime
499	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	7-Chloro-2,3-dihydrobenzo[b]furan-5-carboxaldoxime
500	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	4-Methoxy-3-methylbenzaldoxime
513	N-BOC-L-tert-butylglycine	2-cyclohexylacetic aicd	4-Ethoxy-3,5-dimethylbenzaldoxime
556	N-BOC-L-tert-butylglycine	2-cyclohexylacetic aicd	3-Chloro-4-ethoxybenzaldoxime
591	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2-Pyridinecarboxaldoxime
592	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	2-Pyridinecarboxaldoxime
593	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	4-Chloro-2-pyridinecarboxaldoxime
594	N-BOC-L-tert-butylglycine	2-Cyclohexylacetic acid	3-Chloro-6-fluorobenzaldoxime

[0489] Certain other compounds of the invention may be prepared by Method 7 as illustrated below.

METHOD 7:

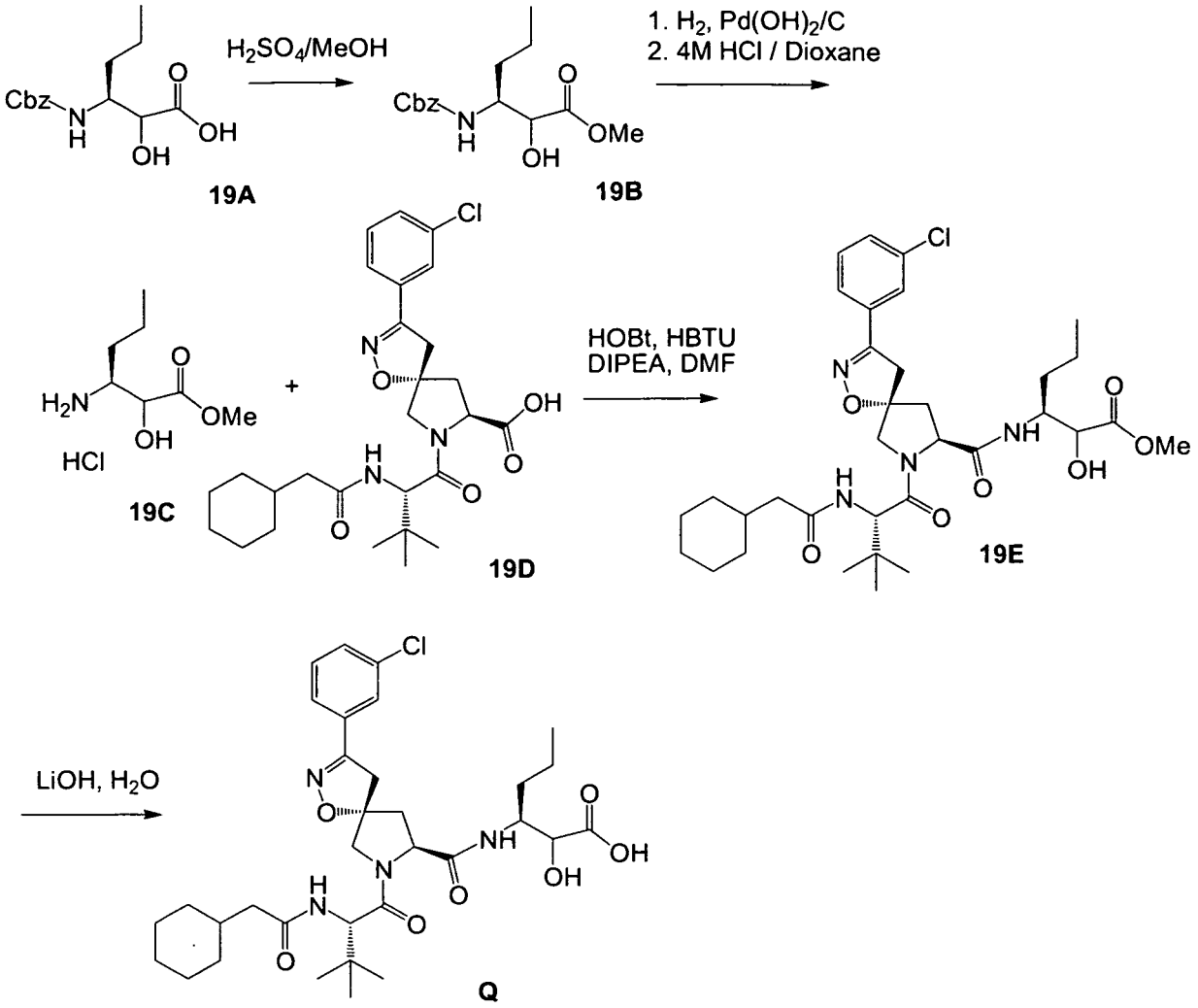




[0490] Referring to Method 7, the Cbz hydroxy acid **G1** is converted to the methyl ester **G2** and deprotected to provide the amino-ester **G3**. Reaction of **G3** with the spiroisoxazoline acid **G4** in the presence of a coupling reagent provides the intermediate **G5**. Hydrolysis of the methyl ester of **G5** provides the hydroxy acid **G6** which is oxidized with, for example, Dess-Martin periodinane to provide the ketoacid **G7**. Reaction of **G7** with an amine $R_{13}R_{10}NH$ in the presence of a coupling reagent provides the final product **G8**.

Example 19: Compound No. 275

Step 1: Preparation of Compound Q.



[0491] 1.00 g of acid **19A** was dissolved in 14 mL of methanol and heated to reflux. Two drops of concentrated H_2SO_4 was added and the reaction refluxed overnight. The mixture was cooled to room temperature, and neutralized with 50 mL of $NaHCO_3$ (sat. aq.). The reaction mixture was extracted three times with 50 mL of ethyl acetate. The combined organic extracts were dried over magnesium sulfate and evaporated to yield 1.01 g of compound **19B** as a white powder. Major diastereomer ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 7.40-7.31 (m, 5H), 5.12 (s, 2H), 4.99 (d, 1H, J=8.7 Hz), 4.35 (s, 1H), 4.15-4.02 (m, 1H), 3.81 (s, 3H), 3.05 (br s, 1H), 1.67-1.17 (m, 4H), 0.91 (t, 3H, J=6.8 Hz). Minor diastereomer ^{1}H-NMR (300 MHz, CDCl3) δ: 7.40-7.31 (m, 5H), 5.07 (s, 2H), 4.90 (d, 1H, J=9.8 Hz), 4.19 (s, 1H), 4.15-4.02 (m, 1H), 3.76 (s, 3H), 3.03 (br s, 1H), 1.67-1.17 (m, 4H), 0.96 (t, 3H, J=7.1 Hz).

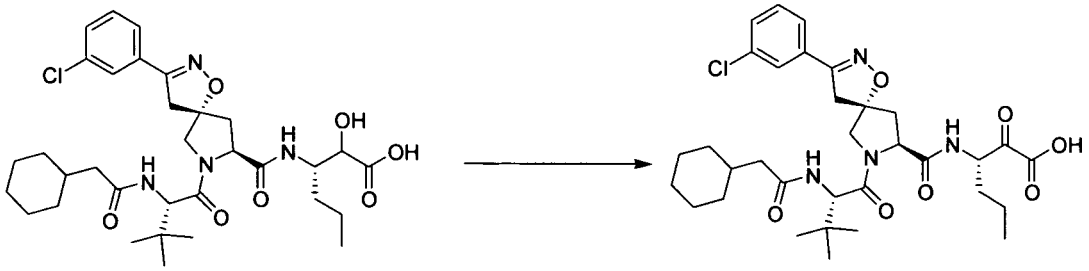
[0492] 1.00 g of CBz-protected methyl ester **19B** was dissolved in 11 mL of methanol. 150 mg of $Pd(OH)_2$ (20 wt% on carbon) was added, and the mixture flushed with 1 atm of hydrogen gas and stirred at room temperature for 3 hours. The methanolic solution was filtered through a Celite® plug and the filter pad rinsed with additional methanol. Upon

evaporation, a light yellow oil was collected and redissolved in 5 mL of DCM and treated with 1.5 mL of 4 M HCl solution in dioxane. Upon stirring for 1 minute, the reaction was evaporated. 0.65 g of compound **19C** was collected as a white powder, and characterized by LCMS (M+1 = 162.0).

[0493] 0.80 g of the spiroisoxazoline acid of compound **19D** was stirred with 0.33 g of HOBt, 0.81 g of HBTU, and 15 mL of DMF. To the stirring solution was added 807 μL of DIPEA, and stirred for 10 minutes. 0.33 g of the hydrochloride salt **19C** was added. The reaction was stirred at room temperature for 3 hours. To the reaction mixture was added 200 mL of EtOAc, and the mixture washed twice with 100 mL of $NaHCO_3$ (sat. aq.), then 100 mL of brine. The organic phase was dried over $MgSO_4$ and evaporated. The crude reaction mixture was purified by elution through silica gel column (40 g column, gradient elution, 40-55% EtOAc:Hexanes) to give 1.02 g of compound **19E** as a white powder, which was identified by LCMS (M+1 = 661.3).

[0494] 1.04 g of methyl ester **19E** was stirred in 6 mL of THF and to this solution was added 3 mL of 1 M LiOH(aq). The reaction was stirred at room temperature for 2 hours where it was determined by HPLC to be complete. The reaction was treated with 6 mL of 1 M HCl, and extracted three times with 15 mL of ethyl acetate. The combined extracts were evaporated to give 1.00 g of compound **Q** as a beige solid which was carried on to the next step.

Step 2: Preparation of Compound R

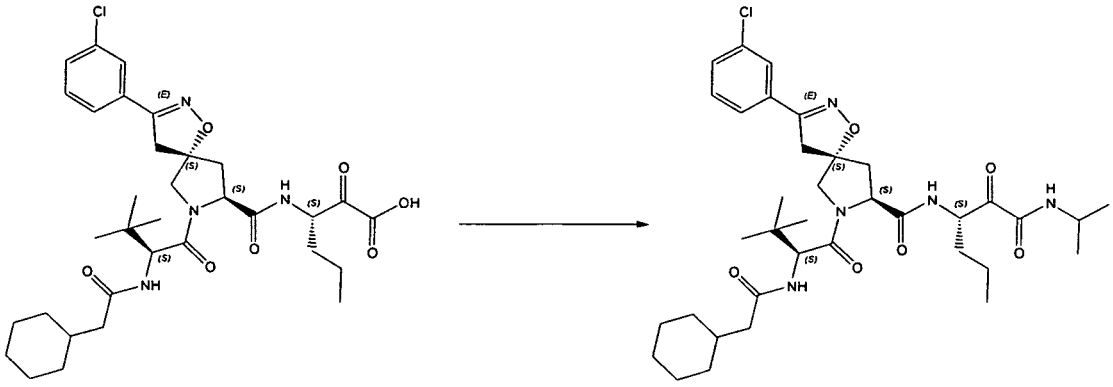


Q **R**

[0495] To a solution of compound **Q** (0.300 g, 0.46 mmol) in CH_2Cl_2 (15 mL) was added 5.58 mL of a 0.16 M solution of Dess Martin periodinane in CH_2Cl_2 dropwise. After it was stirred for 4 hours at room temperature, 10 mL of 1M $Na_2S_2O_3$ solution was added and the reaction mixture was stirred for 30 minutes at ambient temperature. The organic layer was separated, washed with water, dried over Na_2SO_4, filtered and concentrated. The crude mixture was redissolved in CH_2Cl_2 and precipitated with Hexanes and filtered to give 230 mg

of compound **R**. LC/MS: m/z 645.7 $(M+H)^+$ at 1.99 minutes (10-99% CH_3CN (0.035% TFA)/H_2O (0.05% TFA))

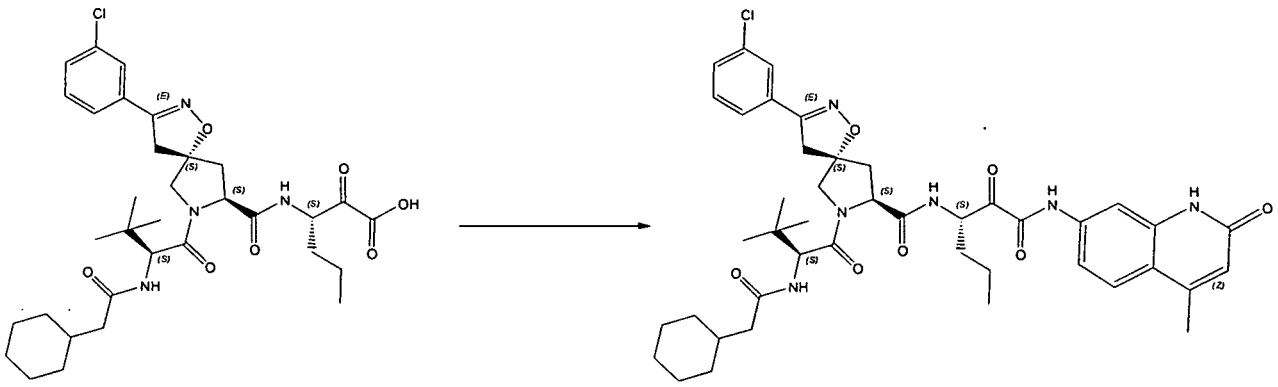
Step 3: Preparation of Compound No. 275



R **Compound No. 275**

[0496] To a suspension of compound **R** (20 mg, 0.0.031 mmol) in anhydrous acetonitrile was added pyridine (10 μL, 0.124 mmol), 2-chloro-1-methyl-pyridinium iodide (15.3 mg, 0.06 mmol), HOBt (6.8 mg, 0.05 mmol), followed by the addition of a 50μL solution of isopropylamine (3.7 mg, 0.062 mmol) in anhydrous acetonitrile. The reaction was allowed to stir at room temperature and complete after two hours. The reaction mixture was quenched with 1mL of saturated aqueous sodium bicarbonate solution, the layers were separated and aqueous layer was extracted three times with CH_2Cl_2. The combined organics were dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was dissolved in 1.5 mL CH_2Cl_2 and purified by normal phase HPLC (10-99% EtOAc/Hexanes) to yield Compound No. **275**. LC/MS: m/z 686.7 $(M+H)^+$ at 2.01 minutes (10-99% CH_3CN (0.035% TFA)/H_2O (0.05% TFA))

Example 20: Compound No. 181.

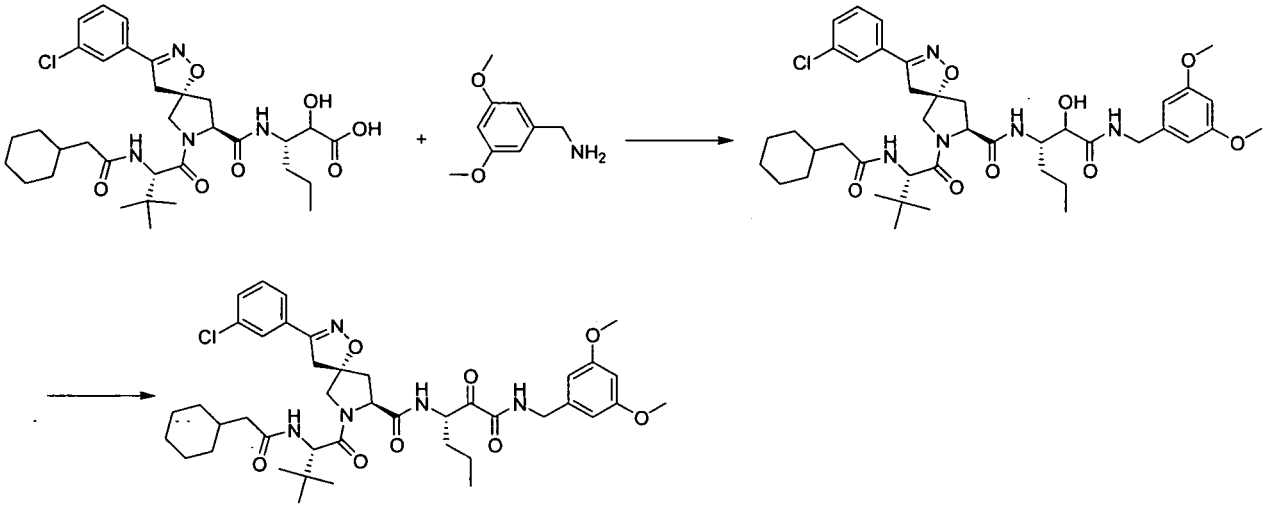


R **Compound No. 181**

[0497] To a suspension of **R** (20 mg, 0.031 mmol) in anhydrous 1,4-dioxane was added pyridine (7.6μL, 0.093 mmol), then pentafluorophenyl trifluoroacetate (8.8μL, 0.05 mmol)

and allowed to stir for 1.5 hours at room temperature, upon which 7-amino-4-methyl-1H-quinolin-2-one (14 mg, 0.08 mmol) was added. The reaction was allowed to stir at room temperature and complete after one hour. The reaction mixture was quenched with 1mL of saturated aqueous sodium bicarbonate solution, the layers were separated and aqueous layer was extracted three times with CH_2Cl_2. The combined organics were dried over Na2SO4, filtered and concentrated under reduced pressure. The residue was dissolved in 1.5 mL CH_2Cl_2 and purified by normal phase HPLC (10-99% EtOAc/Hexanes) to yield Compound No. **181**. LC/MS: m/z 801.7 $(M+H)^+$ at 2.06 minutes (10-99% CH_3CN (0.035% TFA)/H_2O (0.05% TFA)).

Example 21: Compound No. 605



Compound No. 605

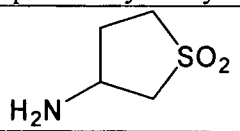
[0498] A mixture of (3S)-3-((5S,8S)-3-(3-chlorophenyl)-7-((S)-2-(2-cyclohexylacetamido)-3,3-dimethylbutanoyl)-1-oxa-2,7-diazaspiro[4.4]non-2-enecarboxamido)-2-hydroxyhexanoic acid (0.02 g, 0.03 mmol), (3,5-dimethoxyphenyl)methanamine (5.68 mg, 0.033 mmol), HOBt (6.8 mg, 0.05 mmol), DIPEA (22 µL, 0.124 mmol) and CH_2Cl_2 (70 µL) was stirred at room temperature for 10 minutes. To the mixture was then added a solution of Mukaiyama's reagent (2-chloro-1-[4-(1H,1H, 2H, 2H-perfluoro-9-methyldecyl) benzyl]pyridinium hexafluorophosphate) in 200 µL of acetonitrile and the reaction was stirred at room temperature. After 5 hours, 1.34 mL of 0.3 M Dess-Martin Periodinane in CH_2Cl_2 was added and the mixture stirred. After 2 hours, the oxidant was quenched with 1.0 mL of saturated $NaHCO_3$, 1 mL of 1N $Na_2S_2O_3$ and stirred vigorously. The organic layer was separated, dried over Na_2SO_4, filtered and concentrated. The residue was dissolved in 1.5 mL CH_2Cl_2 and purified by normal phase HPLC (10%-99% Ethyl acetate/ Hexanes) to yield Compound No. **605,** (5S,8S)-3-(3-chlorophenyl)-7-((S)-2-

(2-cyclohexylacetamido)-3,3-dimethylbutanoyl)-N-((S)-1-(3,5-dimethoxybenzylamino)-1,2-dioxohexan-3-yl)-1-oxa-2,7-diazaspiro[4.4]non-2-ene-8-carboxamide. LC/MS: m/z 794.7 $(M+H)^+$ at 4.11 minutes (10%-99% CH_3CN (0.035% TFA)/H_2O (0.05% TFA)).

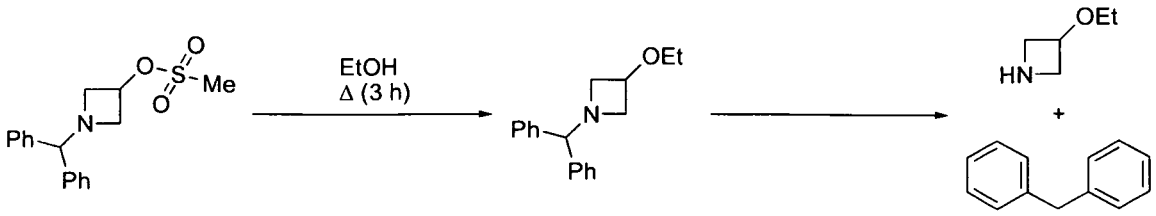
[0499] Listed below in Table 8 are reagents used to prepare additional compounds of Formula I by Method 7.

Table 8: Reagents used to prepare additional compounds of formula I by method 7.

Compound No.	$R_{2z}R_{2w}NH$
2	tert-butylamine
6	2-aminoindane
17	benzo[d]thiazol-2-amine
49	3-((tetrahydrofuran-3-yl)methoxy)azetidine
58	(R)-(+)-1-(3-methoxyphenyl)ethylamine
69	6-Methoxytryptamine
73	4-1H-pyrazol-1-yl-benzylamine
77	benzylamine
79	azetidine
84	2,5-dimethoxyaniline
91	(4-(4-methoxyphenyl)tetrahydro-2H-pyran-4-yl)methanamine
96	3-cyano-4-methylaniline
99	cyclohexylamine
113	N,N-Diethylamine
120	Phenyl-2-pyridinemethylamine
127	3',5'-dimethoxybenzylamine
133	3-Ethoxyazetidine
138	1-(3-(2-aminopropyl)-1H-indol-5-yl)ethanone
140	Ethylamine
141	2,3-dihydro-1,4-benzodioxin-2-ylmethylamine
143	Isobutylamine
148	N-(3-aminophenyl)methanesulfonamide
176	(2-Phenyl-1,3-thiazol-4-yl)metylamine
181	7-amino-4-methylquinolin-2(1H)-one
182	N-Methylethylamine
186	(3R)-(+)-3-acetamidopyrrolidine
206	beta-alanine-4-methoxy-betanaphthylamide
221	N-ethyl-3,4-methylenedioxyamphetamine
238	(R)-3-((tetrahydrofuran-2-yl)methoxy)azetidine
253	Dimethylamine
255	(S)-(-)-1-(3-methoxyphenyl)ethylamine
265	cyclopropylmethylamine
275	Isopropylamine
277	(S)-(+)-tetrahydrofurfurylamine
293	3-aminoisoxazole
296	(S)-alpha-methylbenzylamine
298	3-Pyrazol-1-yl-benzylamine
300	1-(Ethyl)propylamine
302	5-Methoxytryptamine
347	(R)-(-)-2-(methoxymethyl)pyrrolidine
350	N-Methyl-N-propylamine

Compound No.	$R_{2z}R_{2w}NH$
355	3-Aminobenzamide
368	3-(tetrahydrofuran-3-yloxy)azetidine
372	Cyclopentylamine
399	1-Aminocyclopropane-1-carboxylic acid methyl ester
401	Cyclobutylamine
404	2-Methoxyethylamine
408	3-(Aminoethyl)pyridine
410	Morpholine
426	3-Hydroxy-3-methylazetidine
429	1-Phenylcyclopropylamine
433	[3-(4-chlorophenyl0-5-isoxazolyl}methanamine
441	Furfurylamine
447	2-(3-Pyridyl)ethylamine
452	(R)-2-Butylamine
458	3-(2-aminoethyl)indolin-2-one
461	4-(Aminomethyl)pyridine
479	2-Fluoroethylamine
488	2-methoxyphenoxyethylamine
493	Methylamine
496	Pyrrolidine
507	(S)-2-Amino-1,1-dihenyl-1-propanol
508	(S)-(+)-2-(methoxymethyl)pyrrolidine
521	3,3-difluoro-azetidine
537	Propylamine
540	2-(3-methoxyphenyl)ethylamine
546	(R)-alpha-methylbenzylamine
565	
567	2-aminomethyl benzimidazole
568	Pipecoline
573	3,4-Difluoroaniline
588	3-cyanoaniline

Preparation of Non-Commercial Azetidines Listed in Table 8

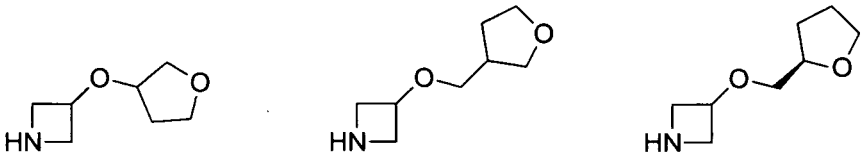


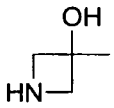
[0500] N-Benzyhydryl-3-methanesulfonylazetidine (104 mg) was combined with ethanol (1.0 mL) and heated in a sealed vial at 95 °C overnight. The reaction was monitored by TLC (30% EtOAc:Hexane). Workup was conducted by adding 1 mL of saturated potassium carbonate solution, and extracting twice with 0.5 mL of ethyl acetate. The combined organic extracts were purified on silica (4g column, gradient elution, 0-30%

EtOAc:hexane). Yielded 49 mg of N-benzhydryl-3-ethoxyazetidine as a clear colorless oil. LCMS (M+1 = 268.2).

[0501] N-Benzhydryl-3-ethoxyazetidine (49 mg) was dissolved in 1 mL of methanol. 22 mg of 10% Pd/C (Degussa-type) was added, and the reaction was carried out under a hydrogen atmosphere. The reaction was stirred at room temperature for 64 h. The mixture was filtered through the Celite®, washed thoroughly with methanol, and evaporated to give a yellow oil (30 mg). The oil consists of a mixture of diphenylmethane and the free azetidine. The crude oil mixture was carried onto subsequent transformations and used in excess.

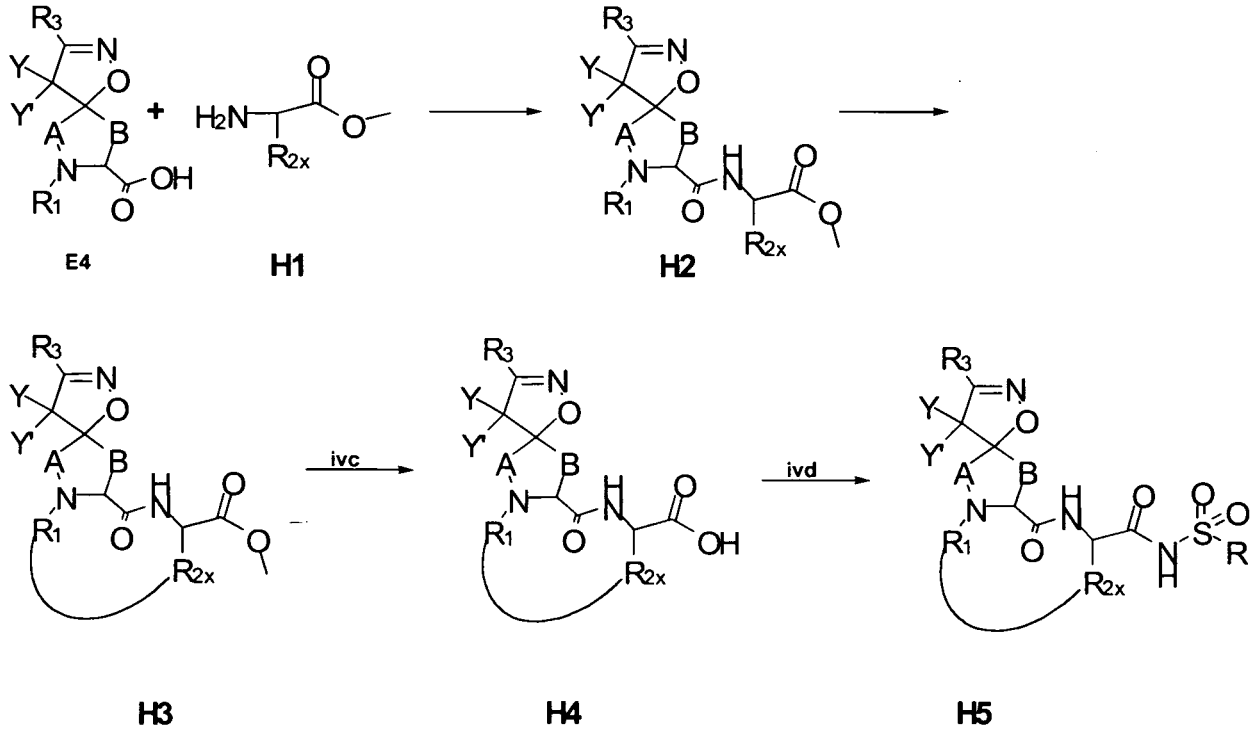
[0502] The following azetidines were prepared in a similar fashion as above, by using the corresponding alcohols.



[0503] The azetidine  was prepared in the method described by Frigola, J. et al. in J. Med. Chem., 36 (1993), 801-810.

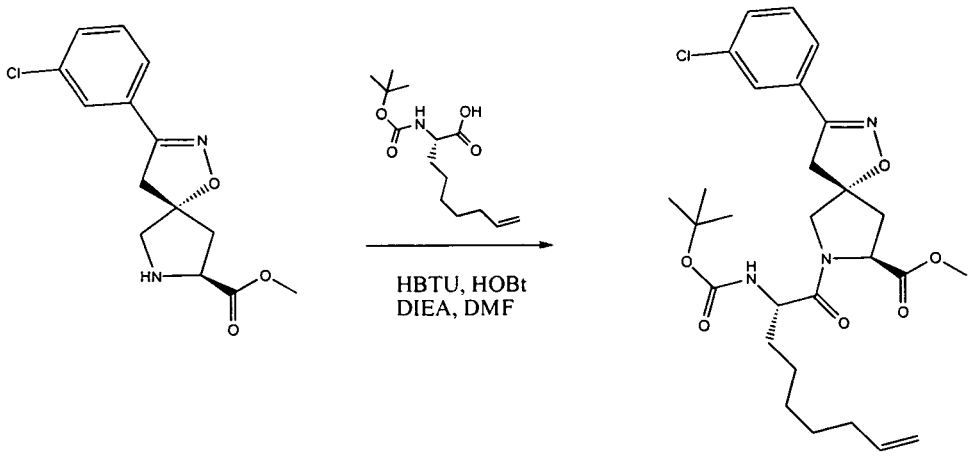
[0504] Certain other compounds of Formula I may be prepared by Method 8 as illustrated below.

METHOD 8:



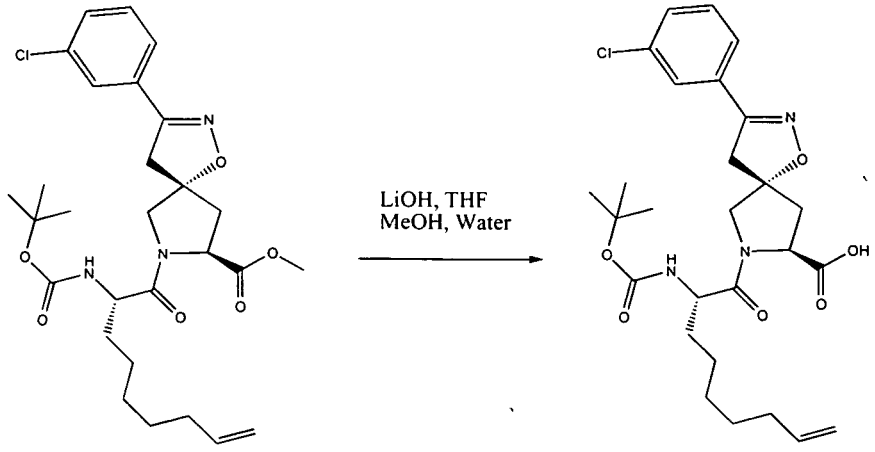
[0505] Referring to Method 8, the spiroisoxazoline acid **E4** reacts with the amino ester **H1** in the presence of a coupling reagent to provide the intermediate **H2**. Macrocyclization of **H2** results in compound **H3**. Hydrolysis of the ester **H2** provides acid **H4**. Reaction of acid **H4** with a sulfonamide or sulfamide in the presence of a coupling reagent provides the product **H5**.

Example 22: Compound No. 409.



22A **22B**

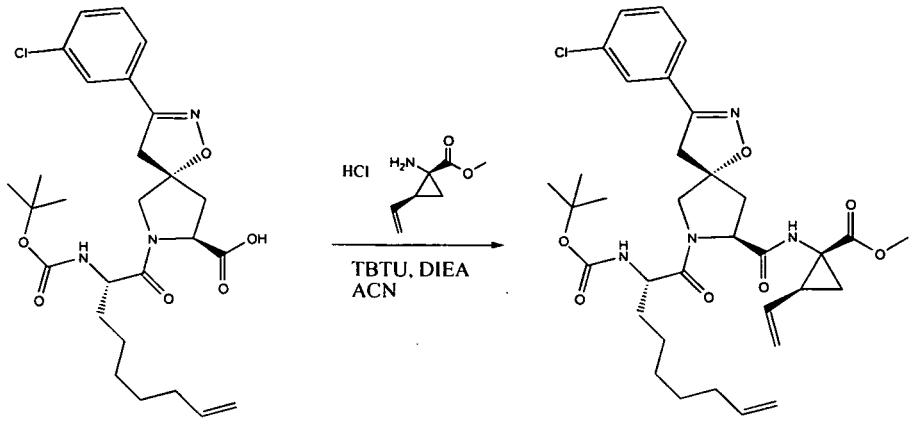
[0506] (S)-2-(tert-butoxycarbonylamino)non-8-enoic acid, purchased from RSp Amino Acid located in Massachusetts, (179 mg, 1.0 eq.) was stirred in DMF with HBTU (376 mg, 1.5 eq.), HOBt (94 mg, 1.05 eq.), and DIEA (345 uL, 3.0 eq.) for 15 minutes. Added compound **22A** (194 mg, 1.0 eq.) and stirred overnight. To the solution was added ethyl acetate. The solution was washed with 1 N HCl (thrice) followed by brine, dried over sodium sulfate, filtered, concentrated and purified by silica chromatography (10-30% ethyl acetate/hexanes gradient) to yield compound **22B** (253 mg). (M+H=548.2).



22B **22C**

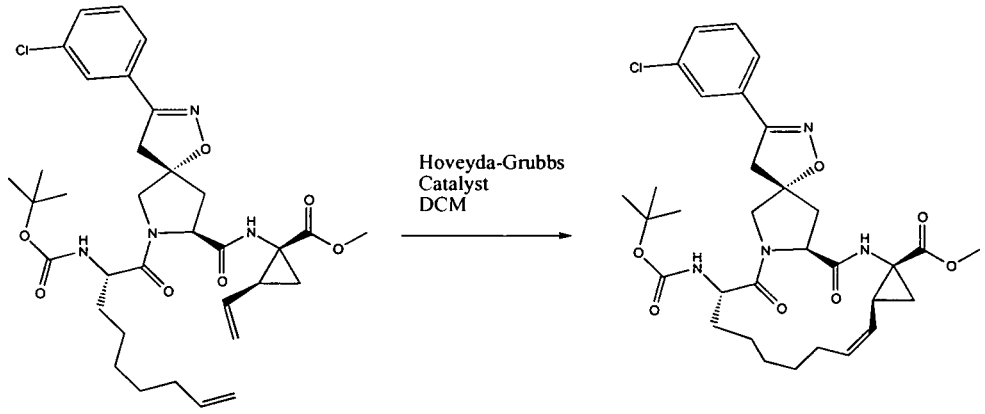
[0507] Compound **22B** (253 mg, 1.0 eq.) was stirred in THF (1 mL) and methanol (0.5 mL). To the solution was added lithium hydroxide (97 mg, 5.0 eq.) in water (0.5 mL) and

stirred for 2 more hours. The mixture was diluted with ethyl acetate, washed with 1 N HCl, then brine, and the solution was dried over $MgSO_4$, filtered and concentrated to yield compound **22C** (235mg) as a pure white solid (M+H=534.2).



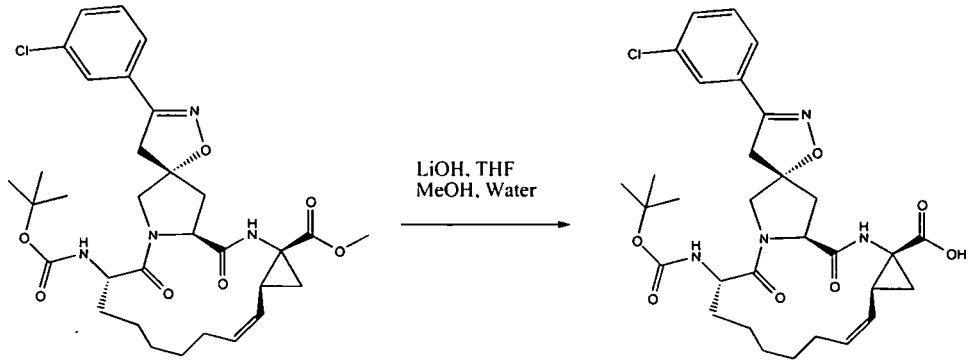
22C **22D**

[0508] Compound **22C** (247 mg, 1.0 eq.) stirred in 1 mL acetonitrile. To the solution was added TBTU (297 mg, 2.0 eq.), DIEA (241 uL, 3.0 eq.), then (1R,2S)-methyl-1-amino-2-vinylcyclopropanecarboxylate (86 mg, 1.2 eq.) and stirred overnight. The solution was diluted with ethyl acetate and washed with 1 N HCl then brine, dried over sodium sulfate, filtered, concentrated and purified by silica chromatography (10-70% ethyl acetate/hexanes gradient) to yield compound **22D** (230 mg). (M+H=657.2).



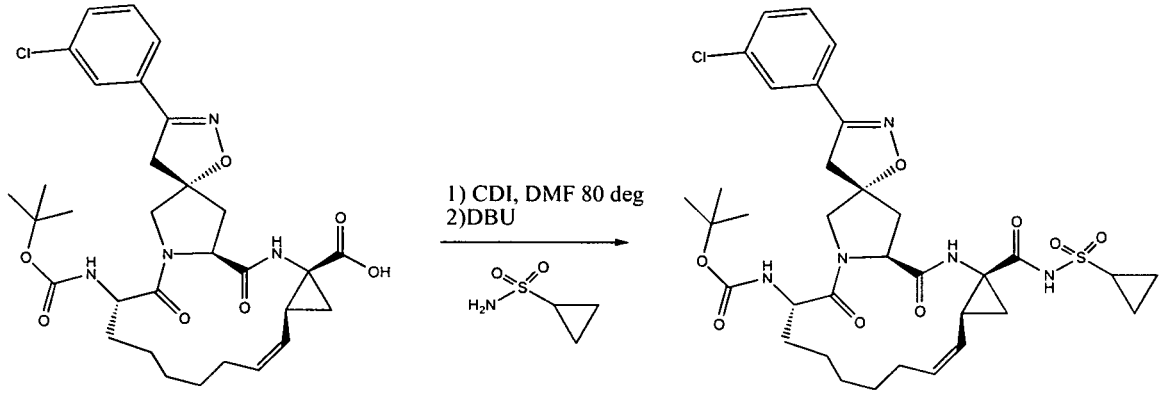
22D **22E**

[0509] Compound **22D** (230mg, 1.0eq.) was stirred in 70mL CH_2Cl_2 with Hoveyda-Grubbs catalyst (22 mg, 0.1 eq.) at reflux for 1 hour, and the solution cooled to room temperature and purified by silica chromatography (10-70% ethyl acetate/hexanes) to yield compound **22E** (172 mg)



22E **Compound 137**

[0510] Compound **22E** (172 mg, 1.0 eq.) was stirred in THF (1 mL) and methanol (0.5 mL). To the solution was added LiOH (46 mg, 4.0 eq.) in 0.5mL water and solution stirred for 2 more hours. To the solution again was added ethyl acetate and washed with 1N HCl and brine, dried over magnesium sulfate, filtered, and concentrated to yield compound **22F** (155mg) as a pure white solid (M+H=617.1).



22F **Compound No. 409**

[0511] Compound **22F** (155 mg, 1.0 eq.) stirred in 1 mL DMF with carbonyldiimidazole (49 mg, 1.2 eq.) at 80 °C for 15 minutes. To the solution was added cyclopropanesulfonamide (49 mg, 1.6 eq.) followed by DBU (36 uL, 1.0eq.) and stirred for another 10 minutes at 80 °C. Then to the solution was added ethyl acetate and solution washed with 1 N HCl and brine, dried over $MgSO_4$, filtered, and concentrated. The product was purified by silica chromatography (100% DCM to 5% methanol/DCM gradient) to give Compound No. **409** (64 mg, 35%). (M+H=718.1.)

[0512] Listed below in Table 9 are additional compounds of Formula I prepared by Method 8.

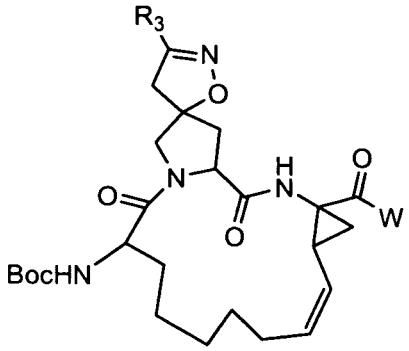
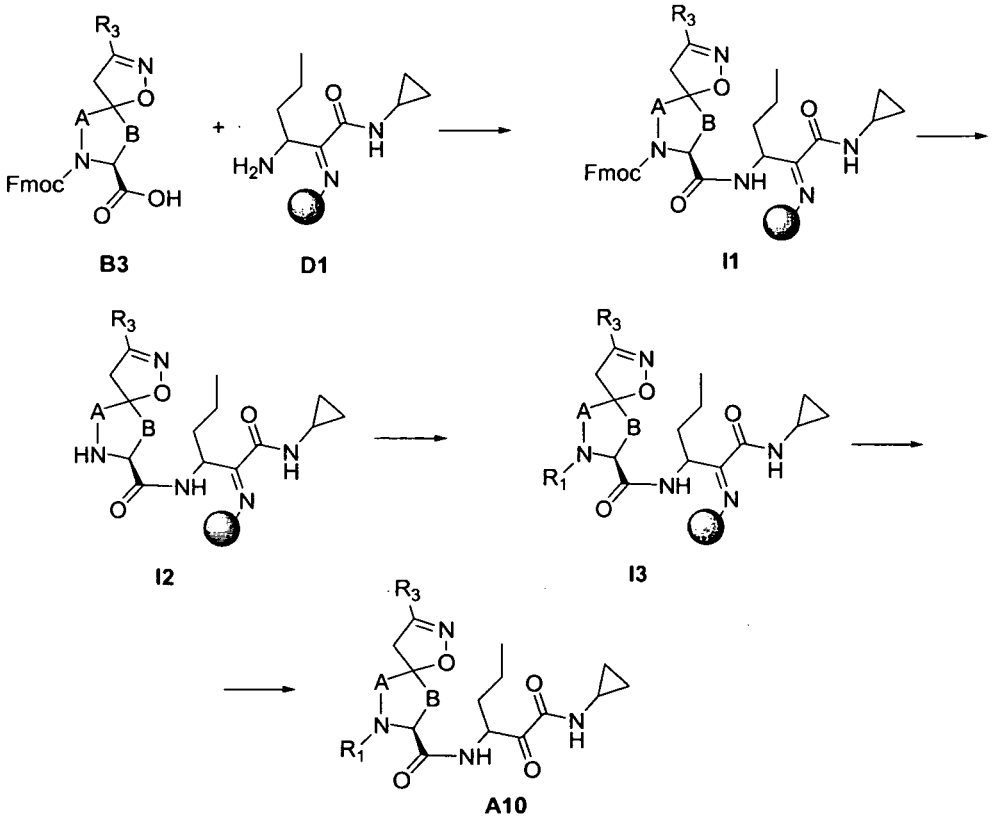


Table 9: Additional compounds of formula I prepared by method 8.

Compound No.	Starting Material for W	Starting Material for R3
1	OH	7-Chloro-2,3-dihydrobenzo[b]furan-5-carboxaldoxime
137	OH	3-Chlorobenzaldoxime
163	Cyclopropane sulfonamide	Phenylglyoxylohydroxamyl chloride
232	Cyclopropane sulfonamide	3-Chlorobenzaldoxime
320	OH	Phenylglyoxylohydroxamyl chloride
386	Cyclopropane sulfonamide	7-Chloro-2,3-dihydrobenzo[b]furan-5-carboxaldoxime
409	Cyclopropane sulfonamide	3-Chlorobenzaldoxime
470	OH	3-Chlorobenzaldoxime

[0513] Certain other compounds of Formua I may be prepared in Method 9 as illustrated below.

METHOD 9:



[0514] Referring to Method 9, the protected spiroisoxazoline **B3** (prepared by Method 2) reacts with the resin bound imino-amine **D1** to provide the intermediate **I1**. Deprotection of **I1** provides the amine **I2** which reacts with an R_1 carboxylic acid in the presence of a coupling reagent to provide **I3** wherein R_1 is $R_4C(O)$-. Hydrolysis of **I3** provides the final compound **A10**.

[0515] A person skilled in the art can use the examples and methods described herein, along with known synthetic methodologies, to synthesize compounds of Formula I according to Method 9 illustrated above.

[0516] Listed below in Table 10 are additional compounds of Formula I prepared by Method 9.

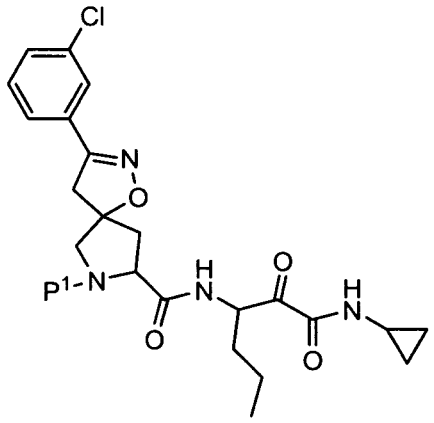


Table 10: Additional compounds of formula I prepared by method 9.

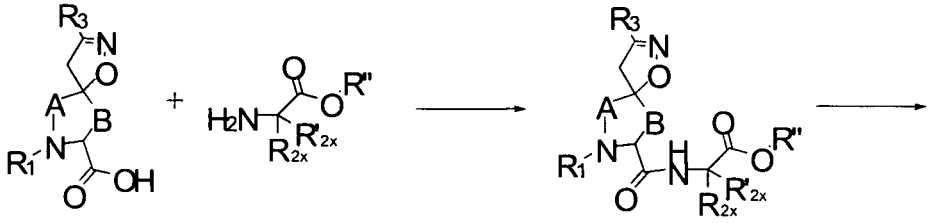
Compound No.	P^1
46	5-Bromoindole-2-carboxylic acid
54	Acetyl-D-ethionine
60	2-(R)-[[(4-Methylphenyl)Sulfonyl]amino]-2-phenylacetic acid
65	2-oxo-1-phenylpyrrolidine-3-carboxylic acid
88	Acetyl-D-Methyltyrosine-OH
98	N-Acetyl-L-leucine
100	2-[[(4-Fluorophenyl)Sulfonyl]amino]-3-methylbutanoic acid
157	5,6-dimethoxyindole-2-carboxylic acid
218	Pyr-Val-OH

Compound No.	P^1
227	1-carbamoylcyclopropanecarboxylic acid
246	5-(2,4-dimethylphenylamino)-5-oxopentanoic acid
248	4-Chloro-2-(6-methoxypyridin-3-ylamino)benzoic acid
309	3-[[(4-acetamidophenyl)sulfonyl]amino]-3-propanoic acid
328	3-(3,4-dihydroisoquinolin-2(1H)-ylsulfonyl)benzoic acid
332	(S)-2-acetamido-3-(4-isopropoxyphenyl)propanoic acid
376	3-(2-oxobenzo[d]oxazol-3(2H)-yl)propanoic acid
380	4-trifluoromethoxyphenylacetic acid
395	2-[[(4-Methoxyphenyl)Sulfonyl]amino]-3-methylbutanoic acid
397	2-((S)-2-oxo-4-phenyloxazolidin-3-yl)acetic acid
412	Acetyl-D-tyrosine-OH
416	2-(R)-[[(4-Chlorophenyl)Sulfonyl]amino]-3-methylpentanoic acid
420	3-(2-diethylamino)-2-oxoethyl)1H-indole-2-carboxylic acidi
421	trans-2-Phenyl-1-cyclopropanecarboxylic acid
466	2-[[(4-Fluorophenyl)Sulfonyl]amino]-2-phenylacetic acid
476	2-(S)-[[(4-Methylphenyl)Sulfonyl]amino]-2-phenylacetic acid
483	3-(N-Phenylphenylsulfonamido)propanoic acid
489	2-(R)-[[(4-Methoxyphenyl)Sulfonyl]amino]-3-methylbutanoic acid
501	2-[(PhenylSulfonyl)amino]-2-phenylacetic acid
534	2-(R)-[[(4-Methoxyphenyl)Sulfonyl]amino]-3-methylpentanoic acid
574	2-(1-oxoisoindolin-2-yl)propanoic acid
586	6-(2,5-dimethoxyphenyl)-2-oxo-1,2,3,6-tetrahydropyrimidine-4-carboxylic acid

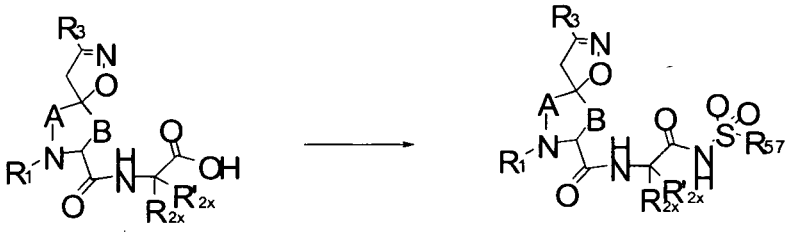
Compound No.	P[1]
587	2-(R)-[[(4-Methoxyphenyl)Sulfonyl]amino]-4-methylpentanoic acid

[0517] Certain other compounds of Formua I may be prepared in Method 10 as illustrated below.

METHOD 10:



B3 **M10A** **M10B**



M10C **M10D**

[0518] Refering to Method 10, the protected spiroisoxazoline **B3** (e.g., R_1 is Fmoc) reacts with **M10A** (R'', e.g., can be methyl or immobilized on PS-Wang resin) to provide intermediate **M10B**. Hydrolysis of **M10B** yields the carboxylic acid **M10C**, which is subsequently coupled with the appropriate sulfonamide to afford the final compound **M10D**. **M10C** can also be a final compound of formula I.

[0519] Similarly, a person skilled in the art can use the examples and methods described herein, along with known synthetic methodologies, to synthesize compounds of Formula I according to Method 10 illustrated above.

[0520] Listed below in Table 11 are additional compounds of Formula I prepared by Method 10.

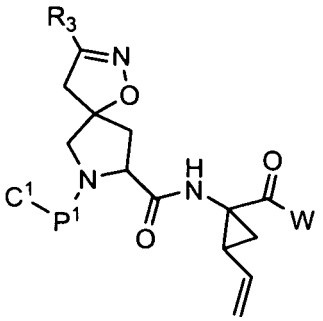
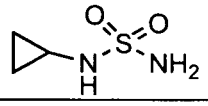


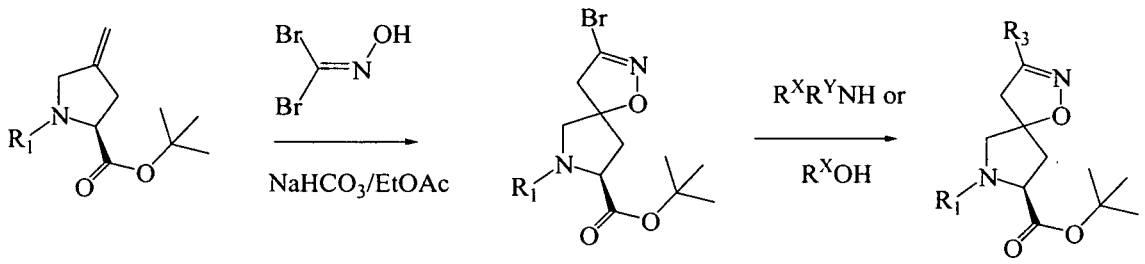
Table 11: Additional compounds of formula I prepared by method 10.

Compound No.	W Starting Material	P1 Starting Material	C1 Starting Material	R3 Starting Material
35	Cyclopropane sulfonamide	N-Boc-L-tert-butylglycine	NA	3-Chlorobenzaldoxime
45	Cyclopropane sulfonamide	N-((S)-tetrahydrofuran-3-yloxy) carbonyl)-L-tert-butylglycine	NA	3-Chlorobenzaldoxime
57	Cyclopropane sulfonamide	N-Boc-L-tert-butylglycine	NA	7-Chloro-2,3-dihydrobenzo[b]furan-5-carboxaldoxime
115		N-Boc-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chlorobenzaldoxime
130	Cyclopropanesulfonamide	N-Alloc-L-tert-butylglycine	NA	3-Chlorobenzaldoxime
144	OH	N-Boc-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chlorobenzaldoxime
162	Cyclopropane sulfonamide	N-Boc-L-tert-butylglycine	2-cyclohexylacetic acid	3-Chlorobenzaldoxime
190	Cyclopropane sulfonamide	N-Boc-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	3-Chlorobenzaldoxime
269	OH	N-((S)-tetrahydrofuran-3-yloxy) carbonyl)-L-tert-butylglycine	NA	3-Chlorobenzaldoxime
272	OH	N-Boc-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	Nitropropane
359	Cyclopropane sulfonamide	N-Boc-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime

Compound No.	W Starting Material	P1 Starting Material	C1 Starting Material	R3 Starting Material
384	OH	N-Boc-L-tert-butylglycine	2-(tetrahydro-2H-pyran-4-yl)acetic acid	3-Chlorobenzaldoxime
438	Cyclopropane sulfonamide	N-Boc-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	Nitropropane
439	OH	N-Boc-L-tert-butylglycine	NA	3-Chlorobenzaldoxime
443	OH	N-Boc-L-tert-butylglycine	cyclopentyl 2,5-dioxopyrrolidin-1-yl carbonate	3-Chlorobenzaldoxime
457	OH	N-Boc-L-tert-butylglycine	tert-Butylisocyanate	3-Chlorobenzaldoxime
460	OH	N-Alloc-L-tert-butylglycine	NA	3-Chlorobenzaldoxime

[0521] Additional compounds of the invention may be prepared as illustrated in the scheme Method 11.

METHOD 11:

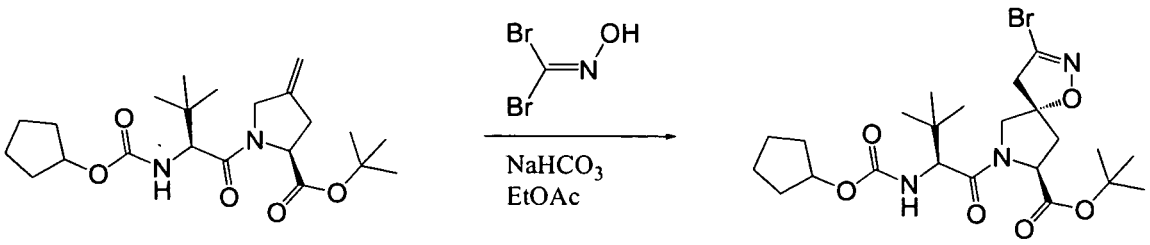


M11A **M11B** **M11C**

[0522] Referring to Method 11, reaction of the methylene compound M11A with 1,1-dibromoformaldoxime in the presence of a mild base such as, for example, sodium bicarbonate provides the bromospiroisoxazoline M11B. Reaction of M11B with an amine R^XR^YNH or an arylalcohol R^XOH provides the intermediate M11C wherein R_3 is an optionally substituted amino or optionally substituted aryloxy. Deprotection of M11C to provide the corresponding acid followed by reaction with an appropriate amino compound according to procedures previously described provides componds of the invention.

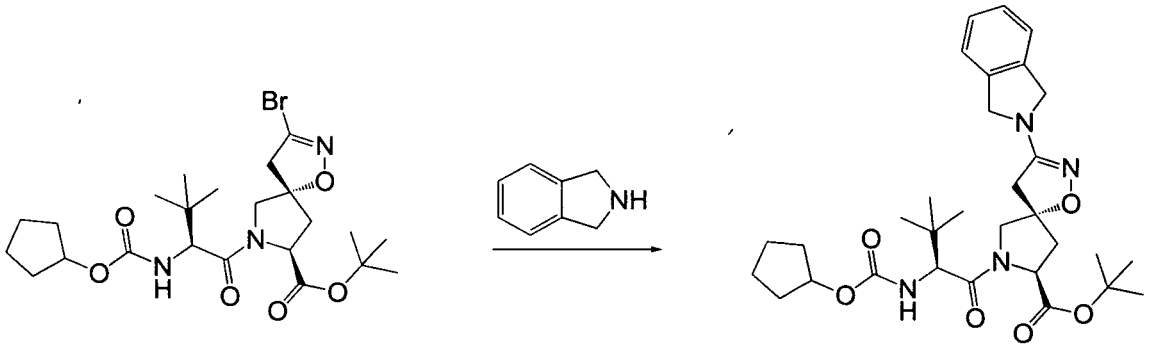
Example 23: Preparation of Intermediates for Compounds Wherein R_3 is Amino or Aryloxy.

Step1:



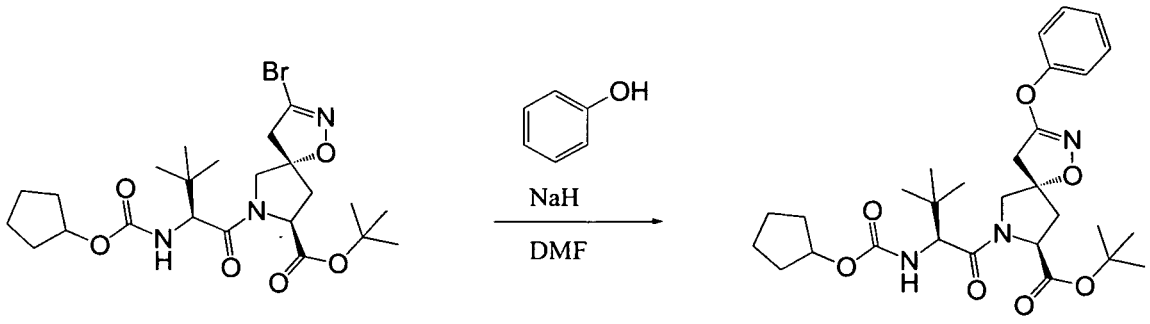
M11D **M11E**

[0523] Compound **M11D** (6.96g, 1.0eq) was stirred in 85mL ethyl acetate. Sodium bicarbonate (6.30g 4.4eq) was added followed by dibromoformaldoxime (4.11g, 1.2 eq) and the mixture stirred overnight at room temperature. Water (85mL) was added and the mixture stirred until clear. The phases were separated, the aqueous phase extracted with ethyl acetate and the combined organic phases were dried over sodium sulfate, filtered, and concentrated. Chromatography on silica column with ethyl acetate/hexanes gradient yielded 5.55grams of Compound **M11E** and 0.93 grams of its diastereomer (6:1 ratio).



M11F **M11G**

[0524] Compound **M11F** (100mg, 1.0eq) was stirred in 270uL of isoindoline at 95° C overnight. The mixture was diluted with ethyl acetate and washed with 1N HCl. The organic phase was dried over sodium sulfate, filtered and concentrated. Chromatography on silica with ethyl acetate/hexanes yielded 83mg of Compound **M11G**.

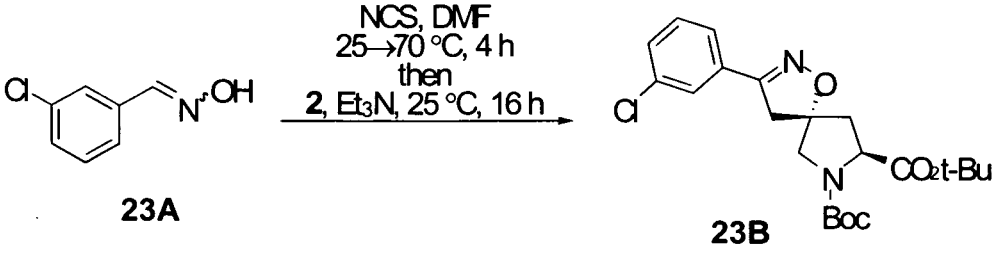


M11F **M11H**

[0525] Phenol (20mg, 1.1eq) stirred in 350uL DMF. Sodium hydride (8.3mg, 1.1eq, 60%) was added and the mixture stirred for 5 minutes. Compound **M11F** was added and the mixture stirred at 90° C overnight. The mixture was partitioned between 1N HCl and ethyl acetate. and the organic phase dried over sodium sulfate, filtered and concentrated to give 70mg Compound **M11H**.

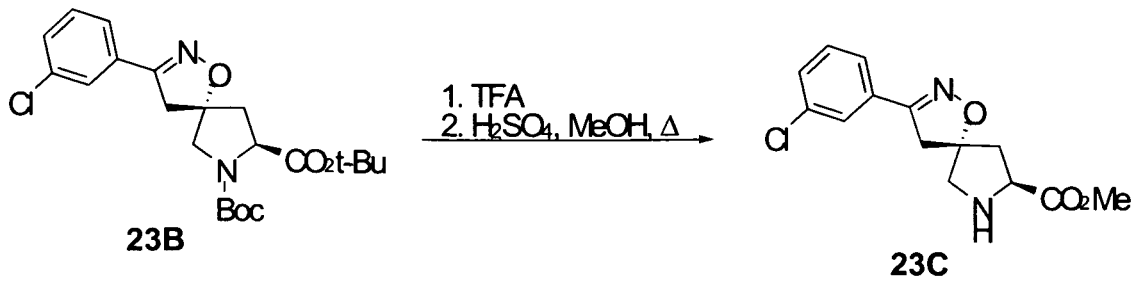
ADDITIONAL EXAMPLES

Example 23: Compound No. 610



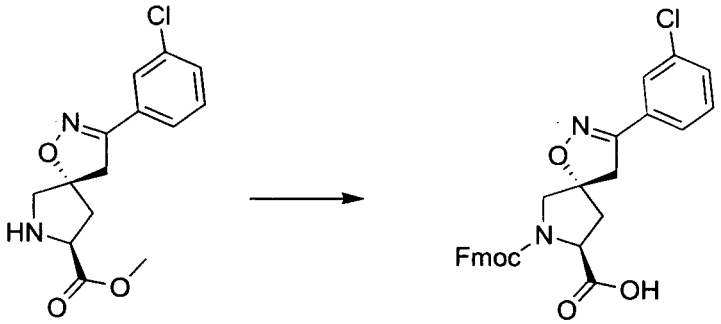
23A 23B

[0526] Oxime **23A** (6.29 g, 40.4 mmol) was dissolved in DMF (63 mL) and N-chlorosuccinimide (5.39 g, 40.4 mmol) was added portionwise to the stirring solution. Stirring continued for 3 hours at room temperature when conversion was determined to be 56% (by HPLC). The reaction was pushed to completion by gentle heating at 70 °C for 45 minutes. 4-Methyleneproline derivative (8.81 g, 31.1 mmol) was added and rinsed into the solution using DMF (5 mL). Triethylamine (5.7 mL) was carefully added dropwise over 30 minutes. The reaction was then stirred at room temperature for 16 hours overnight. An aliquot was analyzed by HPLC and it was determined to contain a 4:1 ratio of cycloaddition diasteromers. Ethyl acetate (200 mL) was added and the organic phase was washed with water (thrice, 200 mL each) and brine (200 mL). The organic phase was then dried over magnesium sulfate and evaporated. The crude oil was divided into two portions and each was purified using an ISCO combiflash equipped with a 330 g silica column (10-20% EtOAc: pet. ether, 72 minutes). The desired product was the major isomer which eluted from the column ahead of the minor isomer and 9.42 g of **23B** was obtained as an orange oil. The minor isomer was also isolated, subjected to a recrystallization from EtOAc:hexane, and obtained as an off-white crystalline powder (1.53 g).



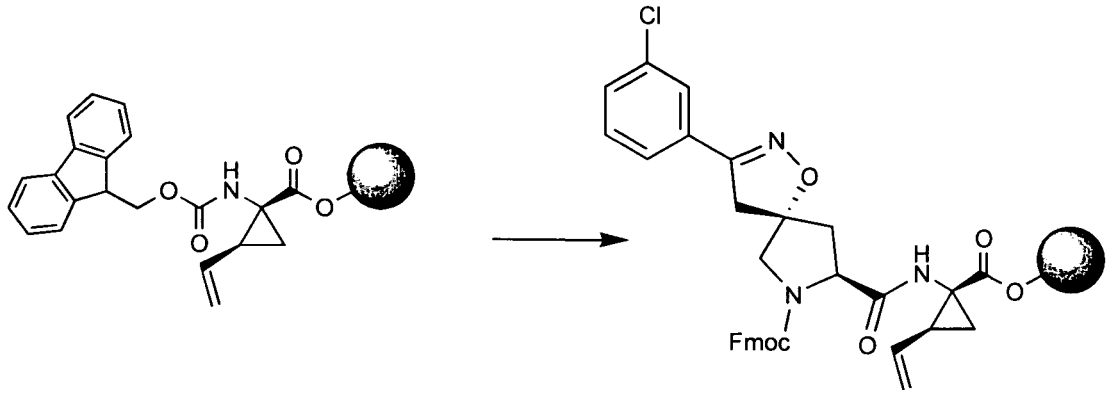
23B 23C

[0527] Compound **23B** (9.42 g) was stirred in trifluoroacetic acid (12 mL) for 2 hours. The solvent was evaporated and replaced with methanol (50 mL). The solution was heated to reflux and H_2SO_4 (3.0 mL) was added dropwise. The reaction was refluxed for a total of 6 hours when by HPLC, conversion to the methylester was determined to be greater than 95%. The reaction was cooled and evaporated to remove the excess methanol. The resulting oil was redissolved in CH_2Cl_2 (200 mL) and neutralized with saturated sodium bicarbonate (200 mL). The organic phase was collected, and the aqueous phase was extracted with CH_2Cl_2 (twice, 100 mL each). The organic extracts were combined, evaporated over magnesium sulfate, and evaporated to give 5.09 g of compound **23C** as an oil that was immediately carried onto the next step.



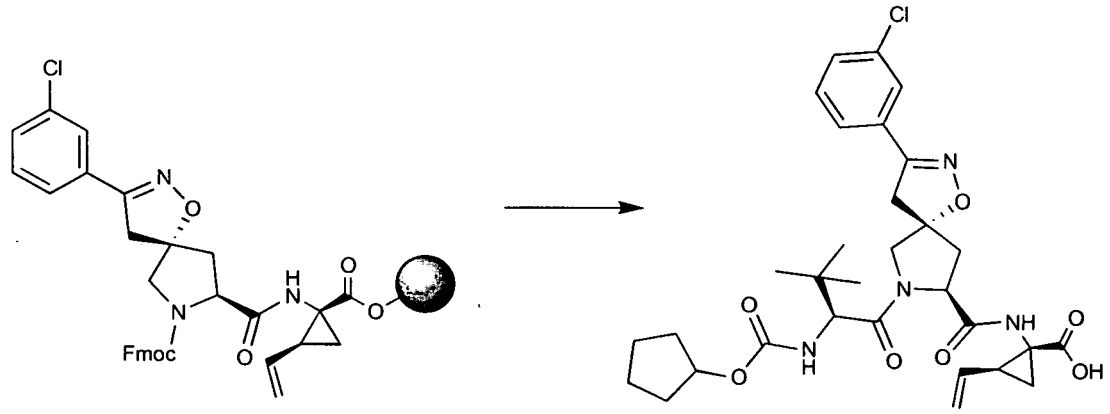
23C 23D

[0528] The amino ester **23C** (1.25 g, 4.24 mmol) was treated with $LiOH \cdot H_2O$ (186 mg, 4.4 mmol) in THF/H_2O (3:1, 10 mL) for 45 minutes. The solvents were removed in vacuo to obtain a solid. This solid was slurried in acetone (20 mL) and saturated $NaHCO_3$ (aq) (20 mL) at room temperature. Fmoc-Cl (1.12 g, 4.33 mmol) was added and the reation was monitored by HPLC. After 20 minutes, the contents of the reaction flask were transferred to a separatory funnel with CH_2Cl_2 and acidified with 2 N HCl (aq.). The aqueous phase was extracted with CH_2Cl_2 (twice, 100 mL each). The resulting emulsion was filtered, and the organic layers were combined, dried over $MgSO_4$, and concentrated to give compound **23D**.



XX4 23F

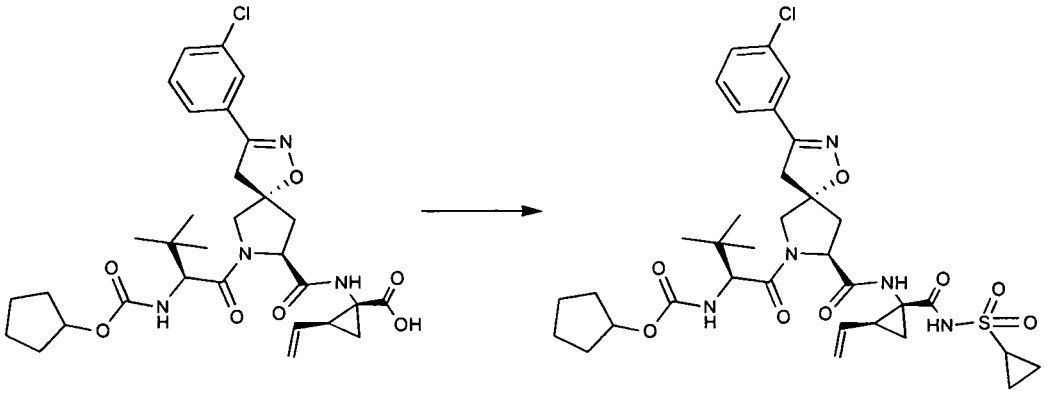
[0529] Compound **XX4** was shaken in a solution of 20% piperidine in DMF (20 mL) for 60 minutes. The resin was washed with DMF (thrice), CH_2Cl_2 (thrice) and repeated. The resulting resin was then shaken with compound **23D** (437 mg, 0.87 mmol), HATU (392 mg, 1.03 mmol), and DIEA (0.300 mL, 1.72 mmol) in DMF (10 mL) overnight. The result compound bound resin **23F** was then washed with DMF (thrice), CH_2Cl_2 (thrice) and repeated. (M+1) = 612.26.



23F **Compound 443**

[0530] The combound bound resin **23F** was shaken in 20% piperidine in DMF (8 mL) for 2 hours. The resin was then washed with DMF (thrice), CH_2Cl_2 (thrice) and repeated. (M+1) = 390.1. This resin was then shaken overnight in DMF with (S)-2-(cyclopentyloxycarbonylamino) -3,3-dimethylbutanoic acid (3 eq.), HOBT (3 eq.), HBTU (3 eq.), and DIEA (6 eq.). The resin was washed with DMF (thrice) and CH_2Cl_2 (thrice) and repeated, then shaken for 100 minutes in TFA (5 mL). The resulting resin was filtered and the filtrate concentrated and purified by reverse phase chromatography to yield 9.4 mg of compound **Compound 443** as a white solid. (M+1) = 615.6, ^{1}H-NMR (500 MHz, DMSO-d_6): 8.63 (s, 1H), 7.67 (s, 1H), 7.63 (d, J=6.7 Hz, 1H), 7.55-7.49 (m, 2H), 6.90 (d, J=8.4 Hz, 1H), 5.77-5.69 (m, 1H), 5.20-5.17 (m, 1H), 5.06 (d, J=10.5 Hz, 1H), 4.93 (brs, 1H), 4.35 (t,

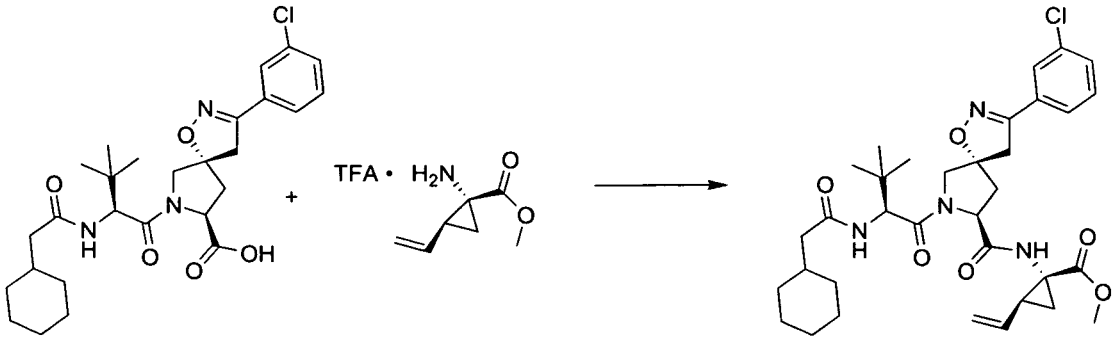
J=7.7 Hz, 1H), 4.11 (d, J=8.8 Hz, 1H), 4.06 (d, J=10.9 Hz, 1H), 3.80 (d, J=11.6 Hz, 1H), 3.62-3.50 (m, 2H), 2.63-2.31 (m, 2H), 2.18-2.13 (m, 1H), 2.07-2.01 (m, 1H), 1.87-1.51 (m, 9H), 1.29-1.28 (m, 1H), 0.95-0.91 (brs, 9H).



23G **Compound No. 190**

[0531] Compound **23G** (6.6 mg, 0.011mmol) was stirred in DMF (0.5 mL) with CDI (2.8 mg, 0.017 mmol) for 1 hour at 80 °C. Cyclopropyl sulfonamide (3.8 mg, 0.031 mmol) and DBU (0.01 mL) were added, the heat was removed and the reaction was stirred overnight at room temperature. The reaction was purified by reverse phase chromatography to yield 2.8 mg of Compound No. **190** (0.0039 mmol). (M+1) = 718.1. ^{1}H-NMR (500 MHz, methanol-d_4): 9.26 (s, 0.4H), 9.02 (s, 0.6H), 7.72 (d, J=1.7 Hz, 1H), 7.61 (dd, J=1.3, 7.3 Hz, 1H), 7.47-7.41 (m, 2H), 5.81 - 5.73 (m, 1H), 5.33-5.30 (m, 1H), 5.14-5.10 (m, 1H), 5.03 (brs, 1H), 4.45-4.41 (m, 1H), 4.31-4.25 (m, 2H), 3.94 (d, J=11.0 Hz, 1H), 3.62-3.53 (m, 2H), 2.99-2.92 (m, 1H), 2.55-2.49 (m, 1H), 2.29-2.23 (m, 2H), 1.89-1.53 (m, 10H), 1.44-1.40 (m, 1H), 1.32-1.24 (m, 1H), 1.19-1.02 (m, 2H), 0.90 (s, 9H).

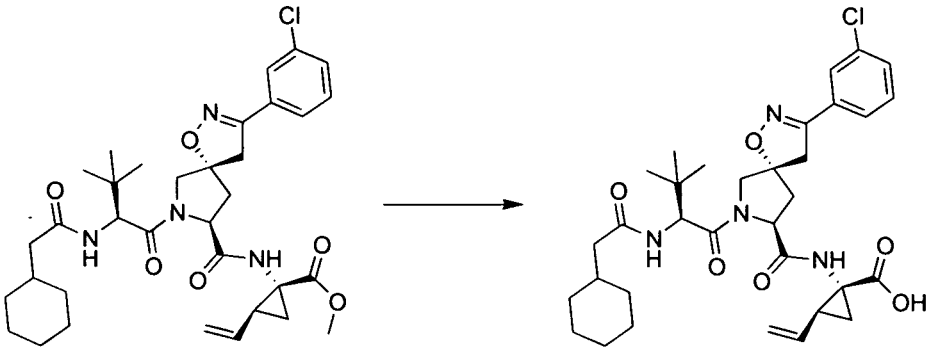
Example 24: Compound No. 618



24A **24B** **24C**

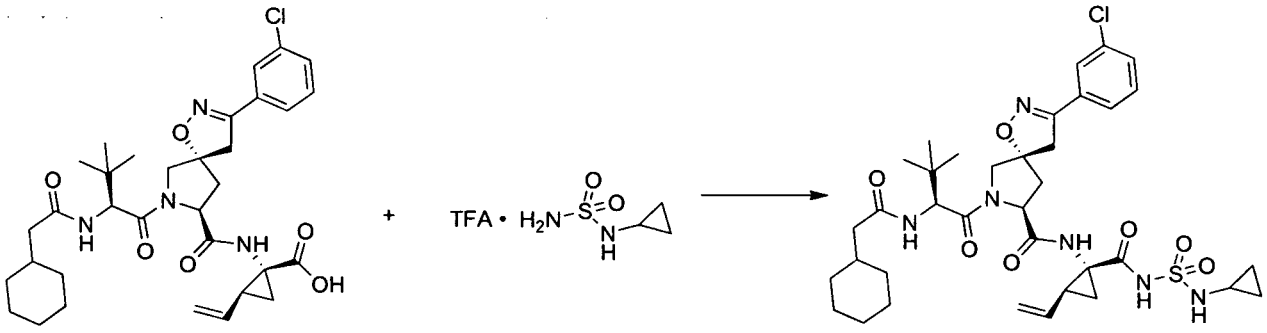
[0532] Carboxylic acid **24A** (69 mg, 0.13 mmol), HATU (50 mg, 0.13 mmol), compound **24B** (0.13 mmol), and DIEA (0.045 mL, 0.26 mmol) were stirred in acetonitrile (1.5 mL) for 2 hours. The reaction was then diluted in EtOAc, washed with saturated

$NaHCO_3$ (aq), brine, dried (MgSO4), and concentrated. Purification on silica gel yielded 76 mg (0.12 mmol) of compound **24C**. LCMS (M+1) = 614.4.



24C **Compound 144**

[0533] The methyl ester **24C** (76 mg, 0.12 mmol) dissolved in THF/H_2O (5:1, 2 mL) and stirred overnight with LiOH•H_2O (1.5 eq.). Acidified reaction with 1N HCl (aq) and concentrated. Residue was dissolved in CH_2Cl_2/MeOH (93:7) and eluted through a plug of silica gel to yield 75 mg (0.1 1mmol) of Compound No. **144** . LCMS (M+1 = 627.4). ^{1}H-NMR (500 MHz, Methanol-d_4): 7.84 (d, J = 9.1 Hz, 0.5H), 7.71 (s, 1H), 7.60 (d, J=7.2 Hz, 1H), 7.45-7.40 (m, 2H), 5.90-5.83 (m, 1H), 5.23 (d, J=1.4 Hz, 1H), 5.07 (d, J=10.3 Hz, 1H), 4.60 (m, 1H), 4.52-4.49 (m, 1H), 4.27 (m, 1H), 3.90 (m, 1H), 3.59-3.48 (m, 2H), 2.58 (dd, J=8.0, 12.6 Hz, 1H), 2.37-2.32 (m, 1H), 2.21-2.12 (m, 4H), 1.76-1.61 (m, 6H), 1.45-1.42 (m, 1H), 1.32-1.14 (m, 4H), 1.05-0.95 (m, 9H), 0.91 (m, 3H).



24D **24E** **Compound No. 115**

[0534] The carboxylic acid **22D** (18.5 mg, 0.029 mmol) stirred with CDI (6.0 mg) in DMF (1.5 mL) at 80 °C for 10 minutes. The reaction was cooled to room temperature and compound **24E** in DMF (0.15 mL) with DBU (4 eq.) was added and the reaction was heated in an 80 °C bath for 20 minutes. The reaction was purified directly by reverse phase chromatography to yield 7.6 mg of Compound No. **115**. LCMS (M+1=745.2), ^{1}H-NMR (500 MHz, methanol-d_4): 9.30 (s, 0.5H), 8.02 (m, 0.5H), 7.71 (m, 1H), 7.60 (dt, J=7.2, 1.3 Hz, 1H), 7.46-7.41 (m, 2H), 5.86-5.79 (m, 1H), 5.35-5.28 (m, 1H), 5.12-5.10 (m, 1H), 4.65 (m, 1H), 4.42 (dd, J=6.9, 10.6 Hz, 1H), 4.28 (d, J=11.3 Hz, 1H), 3.95 (d, J=11.4 Hz, 1H), 3.62-

3.47 (m, 2H), 2.51-2.47 (m, 1H), 2.35-2.31 (m, 1H), 2.25-2.12 (m, 4H), 1.89 (dd, J=5.4, 8.1 Hz, 1H), 1.80-1.64 (m, 6H), 1.45-1.39 (m, 1H), 1.33-1.15 (m, 3H), 1.04-0.97 (m, 11H), 0.73-0.57 (m, 4H).

[0535] Listed below in Table 12 are some physical data of exemplary compounds of Formula I.

[0536] LC/MS data were acquired using the following:

Mass spectrometers: PESciex API-150-EX or Waters/Micromass ZQ or Waters/Micromass Quattro II, or Waters/Micromass ZMD; Pumps: Shimadzu LC-8A or Agilent 1100; Autosamplers: Gilson 215 or Gilson 819.

[0537] The following methods were used: 3.0 mL/min flow rate, 10-99% CH_3CN (0.035% TFA) / H2O (0.05% TFA) gradient, Phenomenex Luna 5m C18 column (50 x 4.60 mm); 1.5 mL/min flow rate, 10-90% CH_3CN (0.2% Formic acid) / H2O (0.2% Formic Acid) in 3 minutes, YMC-Pack Pro-C18 column (50 x 4.6 mm); 1.0 mL/min flow rate, 10-90% CH3CN (0.2% Formic acid) / H_2O (0.2% Formic Acid) in 5 minutes, YMC-Pro-C18 column (50 x 2.0 mm); 1.5 mL/min flow rate, 10-90% CH_3CN (0.1% TFA)) / H2O (0.1TFA) in 3 minutes,, YMC-Pack Pro-C18 column (50 x 4.60 mm).

Table 12: Physical data for exemplary compounds of Formula I.

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
1			754.4	756.1	
2	700.2	3.99			
3	698.8	3.84			
4			763	765	
5	686.3	2.8			
6	760.9	2.4			
7	710.2	3.35			
8				707.9	
9			638	640	
10			724	726.1	
11			662.4	664.3	
12	602	2.94			
13	668.2	3.6			
14				580.9	
15			725.1	726.1	
16				691.8	
17	791.8	2.29			
18	752.4	3.74			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
19	696.3	3.7			H-NMR (500 MHz, $CDCl_3$)7.61 (d, J = 1.7 Hz, H), 7.51(d, J = 7.7 Hz, H), 7.38(d, J = 8.2 Hz, H), 7.33(t, J = 7.8 Hz, H), 7.26(s, CDCl3), 7.16 (t, J = 6.5 Hz, H),6.91 (d, J = 3.3 Hz, H), 6.55 - 6.50(m, H), 5.35 (dd, J = 4.1, 8.5 Hz, H),4.77 - 4.74 (m, H), 4.66(d, J = 9.4 Hz, H), 4.29(d, J = 11.1 Hz, H), 3.71(d, J = 11.2 Hz, H), 3.47 (d, 1H),3.29 (d, 1H), 2.78 (td, J = 7.3, 3.7 Hz, H),2.63 - 2.59 (m, H), 2.52 - 2.49 (m, H),2.07-2.30 (m, 2H), 1.98 - 1.92 (m, H),1.77 - 1.61 (m, H), 1.47 - 1.40 (m, H),1.35 - 1.25 (m, H), 1.21(dd, J = 6.6, 12.5 Hz, H),1.01 (s, 9H), 0.98 - 0.83 (m, H),0.61 - 0.51 (m, H),0.39 (t, J = 4.8 Hz, H) ppm
20	672	3.24			
21			697.9	699.8	
22	713	3.9			$CDCl_3$; 7.77 (d, 2H),7.58 (m, 4H),7.41 (t, 2H),7.32 (3H)
23	710	4.5			
24			630.3	632.2	
25			684.6	686.5	
26	761.4	3.23			
27	672.4	3.7			
28	675.3	3.44			
29	698.361	3.8			
30	767.6	3.15			
31	701.3	2.74			
32			730.3	732.1	
33	685	3.5			
34				559	
35	706.1	3.66			
36			675.5	677.3	
37	712	3.19			
38	748.1	3.86			
39	728	3.82			
40			655.3	657.2	
41	729	3.4			
42	701.3	2.75			
43			654.1	656.1	
44	658	3			
45	720.2	3.29			
46	670	3.78			
47			734.2	736.1	
48	728.2	3.47			
49	784.2	3.71			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
50	696.6	3.76			
51			548.1	550	
52				704	
53	696	2.04			
54	634	3.24			
55			642.3	644.2	
56			600.6	602.4	
57				755	
58	778.9	2.38			
59	730.2	3.58			
60	734.4	3.67			
61	591.8	2.6			
62	690.2	3.64			
63	698	3.83			
64				595	
65	634.4	1.8			
66			734.4	736.2	
67			720.4	722.2	
68	675.2	2.6			
69	817.7	3.97			
70			712.4	714.3	
71	722.1	3.59			
72	670	3.53			
73	800.7	3.97			
74			682	684	
75	713.1	2.8			
76	643	3.09			
77	734.2	3.92			
78					
79	684.2	3.7			
80			553.1	555	
81	687	2.1			
82			611	612.6	
83			772.1	774	
84	780.9	4.39			
85	650	3.26			
86			676.5	678.3	
87	712	3.2			
88	666	3.31			
89	695.2	3.53			
90			758	759.9	
91	848.7	2.3			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
92	686	3.05			
93	670.4	3.56			
94	660	2.69			
95			583.9	585.7	
96	759.3	4.02			
97			670.1	672.1	
98	602.6	1.63			
99	726.7	2.4			
100	704	3.64			
101			666.1	668.3	
102	710.2	3.6			
103	657	3.2			
104	690	3.24			
105			682.1	684.2	
106			690.1	691.9	
107	659	3.22			
108	700	3.32			
109			706.4	708.4	
110	754	3.6			
111			652.1	654	
112				684	
113	700.7	2.3			
114			712.4	714.3	
115	745.2	3.73			
116			716.3	718	
117				653	
118	726.4	3.6			
119				691.8	
120	811.5	3.95			
121	714	3.27			
122	711	3.42			
123	631	3.09			
124	686.2	3.4			
125	732.2	3.7			
126			680.55	682.4	
127	794.7	4.11			
128	774	3.28			
129	758.2	3.78			
130	690.2	3.48			
131	720	3.53			
132	711.4	3.5			
133	728.6	3.8			
134	726.2	3.7			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
135	773.1	3.5			
136			666	668.1	
137			708.2	710.1	
138	843.9	2.2			
139	708	3.35			
140	672.7	2.13			
141	792.7	4.13			
142			676.2	677.9	
143	700.7	2.32			
144	627.4	3.43			
145				707.8	
146	635	2.47			
147			690	691.9	
148	813.9	2.15			
149				606.8	
150			713.5	715.2	
151			706	707.8	
152	688.4	3.2			
153	714.3	3.2			
154			700.1	701.9	
155	709.6	3.59			
156	658	3.35			
157	650	3.32			
158	714.3	3.72			
159			688.3	690.3	
160			613.5	615.4	
161	769.1	3.5			
162	730.2	3.7			
163				708	
164			630.1	632.1	
165	715.2	3.73			
166	710	3.46			
167	709.4	2.2			
168	656.3	3.46			
169	702.2	3.71			
170	754.2	3.96			
171				613.1	
172				717.1	
173	667	3.17			
174			642.3	644.3	
175	701.2	2.76			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
176	817.7	4.1			
177			787	789	
178			576.1	578.1	
179	718.2	3.55			
180			770.1	772.1	
181	801.7	2.06			
182	686.7	2.19			
183			714	715.9	
184	672.2	2.96			
185	724.2	3.6			
186	755.3	3.41			
187	708	3.53			
188	698	3.76			
189	722	3.5			
190	718.1	3.68			
191			710.4	712.4	
192			683.3	685.3	
193	646	3.44	702	703.7	
194			672.1	674	
195	700.2	3.59			
196			672.1	674	
197	722	3.39			
198			723.8	725.7	
199			766.2	768.2	
200			746.2	748.1	
201			567.1	568.8	
202			638	640	
203	730.3	3.7			
204	734.2	3.85			
205	637	3.35			
206	871.9	3.96			
207	576	2.87			
208			718.2	719.95	
209			716.3	718.1	
210	675	3.5			
211	698.2	3.47			
212			692.6	694.5	
213	748.2	3.6			
214			769.3	770.9	
215			733.3	735.3	

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
216	767.1	3.1			
217	670	3.6			
218	657	3			
219	670.2	3.65			
220			504.1	506	
221	792.9	4.1			
222			593	594.9	
223	678	3.42			
224			707.9	710.1	
225				795	
226	698	3.89			
227	558.5	2.91			
228				670	
229	649.7	2.55			
230	704.3	3.7			
231	684	3.73			
232	718.1	3.82			
233	700.34	3.3			1H-NMR (500MHz, $CDCl_3$): 7.61 (t, J = 1.6 Hz, 1H), 7.53 (dd, J = 1.2, 7.6 Hz, 1H), 7.39 (dd, J = 1.7, 6.9 Hz, 1H), 7.34 (t, J = 7.8 Hz, 1H), 7.26 (s, CDCl3), 7.16 (d, J = 7.3 Hz, 1H), 6.91 (d, J = 3.4 Hz, 1H), 6.62 (d, J = 9.2 Hz, 1H), 5.35 (d, J = 4.1 Hz, 1H), 4.76 (t, J = 7.8 Hz, 1H), 4.64 (d, J = 9.3 Hz, 1H), 4.29 (d, J = 10.8 Hz, 1H), 3.71 (d, J = 11.2 Hz, 1H), 3.29-3.49 (dd, J = 2H), 2.78 (m, 1H), 2.63 - 2.59 (m, 1H), 2.53 - 2.51 (m, 1H), 2.37 (d, J = 2.2 Hz, H), 1.90-1.96 (m, 1H), 1.68 - 1.60 (m, H), 1.47 - 1.40 (m, H), 1.21-1.32 (m), 0.99 (s, 9H), 0.95 - 0.83 (m, H), 0.60 (dd, J = 3.4, 9.5 Hz, H) ppm
234			722	724	
235	626.1	3.34			
236			716.1	718.2	
237	696.2	2.04			
238	784.2	3.77			
239	738	3.8			
240			668.2	670.2	
241	713.4	2.94			
242			667.4	669.4	
243	605.9	2.77			
244				653.2	
245	682	3.58			
246	664.7	3.39			
247	645.3	3.3			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
248	707.4	3.7			
249	665	3.49			
250	714.2	3.8			
251			613.4	615.2	
252	682	3.7			
253	672.3	3.72			
254	728.4	3.6			
255	778.9	2.37			
256	668.5	3.6			
257			626	628	
258			566.3	568.1	
259	783	3.24			
260	662	3.16			
261	821	3.21			
262	612	3.4			
263			608.9	611	
264	692	3.31			
265	698.3	3.87			
266			704.3	706.2	
267			575.6	577.4	
268	752	3.87			
269	617.6	2.71			
270				720	
271	704	3.61			
272			696	698.1	
273	644	3.46			
274			714.1	716.2	
275	686.7	2.21			
276			716.1	717.8	
277	728.3	3.75			
278	782	3.9			
279			670.3	672.2	
280			638.2	640	
281			684.5	686.4	
282	772.2	3.19			
283	645	2.02			
284	698	3.8			
285			695.7	697.6	
286	760	3.13			
287	708	3.4			
288	695	3.46			
289	786.2	3.7			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
290			695.7	697.6	
291	787	3.02			
292	685.4	3.36			
293	711.1	3.7			
294	684	3.76			
295			654.5	656.5	
296	748.7	2.37			
297	718	3.48			
298	800.6	2.3			
299			706.6	708.3	
300	714.7	2.36			
301			692.1	694.1	
302	817.9	3.96			
303	651.5	2.9			
304	727	2.87			
305	726.2	3.52			
306			757.5	759.5	
307	744	3.57			
308	702.5	3.4			
309	715.3	3.59			
310			683	685	
311	767.2	3.4			
312			789.1	791	
313	640	3.1			
314			718	720	
315	755.3	3.67			
316			813	815	
317	747.2	3.35			
318			644.1	645.9	
319	658	3.57			
320			775.3	777.1	
321	744.3	3.75			
322				666	
323				746.3	
324	716.9	3.5			
325				676.5	
326			654.1	655.9	
327			686.5	688.4	
328	746.7	3.65			
329			613.4	615.2	

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
330				678	
331			726.5	728.3	
332	694.5	1.82			
333	707	3.43			
334	705.8	3.66			
335	720	3.5			
336	774.2	3.8			
337			680.6	682.6	
338	736.2	3.6			
339	757	3.24			
340			682	684	
341	697.1	2.86			
342	626	2.3			
343				559	
344				684.2	
345	696	3.98			
346	710.2	3.52			
347	742.7	2.28			
348			531.6	533.3	
349	730.4	3.5			
350	700.3	3.94			
351			634.5	636.3	
352			675.6	677.3	
353				691.9	
354			700.6	702.5	
355	762.2	3.91			
356	760	3.11			
357	695	2.24			
358			686.1	687.9	
359	732.4	3.12			
360	698	3.87			
361	698	3.83			
362			652	654	
363			722	724.1	
364	701.9	3.21			
365				676.1	
366	728.1	3.6			
367	636	2.8			
368	770.2	3.64			
369			721.1	723.2	
370	728.3	3.62			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
371	695	3.7			
372	712.3	3.98			
373	723.4	2.3			
374	688.4	3.2			
375			656	658	
376	636.3	3.36			
377	795.4	3.29			
378			761.1	763	
379	645	3.16			
380	649.5	1.97			
381	731.4	3.3			
382			758.3	760.1	
383	612.1	3.2			
384	629.4	2.78			
385			504.1	506	
386			504.1	506	
387	716	3.13			
388	656	3.42			
389	723.4	2.3			
390			696.1	698	
391			583	584.8	
392	660	3.05			
393	696	4.05			
394	703.351	3.3			
395	730	3.55			
396	698.25	3.89			
397	650.5	1.73			
398			696.1	698	
399	742.7	2.16			
400			583	584.8	
401	698.3	3.9			
402	764.1	3.4			
403			716.9	719	
404	702.5	2.07			
405					
406			670.1	672.2	
407	632	3.35			
408	735.7	3.14			
409					
410	714.5	2.12			
411			566.10	568	
412	652	3			
413			717.9	719.8	

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
414			769.1	771	
415			639.5	641.5	
416	734.5	3.83			
417				653.3	
418	686.3	3.1			
419	714	3.26			
420	704	3.56			
421	591.6	1.87			
422			693.4	695.4	
423			714.2	716.1	
424	706	3.31			
425			691.8	693.8	
426	714.2	3.48			
427	666	3.38			
428			702.1	704	
429	760.9	2.38			
430	753.2	3.86			
431	691.9	3.3			
432	645	3.05			
433	835.7	4.16			
434	720	3.5			
435	744	3.56			
436			774	776.2	
437	730.2	3.7			
438			548.2	549.9	
439	603.4	3.34			
440			602.6	604.4	
441	724.9	2.22			
442			712.6	714.5	
443	615.6	3.25			
444			676.2	678.2	
445	742.35	3.2			
446	756.2	3.68			
447	749.7	1.78			
448			608.1	610	
449	756	351			
450			698.3	700.2	
451				630	
452	700.3	3.94			
453	694.3	3.64			
454	761.1	3.3			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
455				724.4	
456			710.5	712.2	
457	602.4	3.12			
458	803.7	3.97			
459	684.2	3.6			
460	587.5	3.01			
461	735.7	1.8			
462			610.1	611.9	
463	708.4	3.7			
464			706.1	708.2	
465			740.4	742.2	
466	738.6	3.63			
467	696.345	3.7			
468	686.2	2.98			
469	681.3	3.39			
470			610.1	612.05	
471	708.2	3.5			
472			837	839.1	
473			706	708.1	
474	710.2	3.2			
475	714	3.3			
476	734.4	3.67			
477	717.37	3.3			
478	690.2	3.73			
479	690.2	3.66			
480			672	673.9	
481			718	720	
482			698.2	700.1	
483	734.6	1.87			
484	660	1.44			
485	676	3.38			
486			803.6	805.4	
487	762	3.26			
488	794.7	4.07			
489	716.5	3.59			
490			709.4	711.4	
491				754	
492	659	3.39	734.4	736.2	
493	658.3	3.61			
494	744.2	3.71			
495	688.2	3.3			
496	698.3	3.83			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
497	694	2.16			
498			670.3	672.2	
499	726.2	3.65			
500	694.3	3.64			
501	720.5	3.62			
502			724.1	725.9	
503	700	3.36			
504	692.8	2.13			
505	713.8	2.73			
506	718	1.87			
507	854.7	4.15			
508	686.7	2.21			
509				724.3	
510	756.2	2.95			
511			680.5	682.54	
512			746.4	748.3	
513			726.4	728.2	
514	635	3.68			
515	688.4	3.2			
516	700	2.98			
517			744.1	746.1	
518	775.2	3.3			
519				636	
520	660	3.5			
521	720.1	3.84			
522	670	3.59			
523	672	3.1			
524					
525			735.2	737	
526	694	3.64			
527			746.1	748.1	
528	731.9	3.38			
529	732	2.89			
530				722	
531	650	3.46			
532	644	3.39			
533			694	696	
534	730.5	3.67			
535	668.2	3			
536			705.8	707.9	
537	742.7	2.25			
538	731.2	3.7			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
539			743.2	744.2	
540	778.9	4.15			
541	700.34	3.2			
542	685.34	3.5			
543			695.7	697.7	
544	746.2	2.3			
545			696.1	697.9	
546	748.7	2.38			
547			726.4	728.25	
548			682	684	
549			696	697.9	
550			653.3	654	
551				609.3	
552	692.3	3.51			
553	712.2	2.6			
554	670.5	2.9			
555					
556					
557	725.8	3.4			
558	679	3.46			
559			702	704	
560			696.2	698	
561	730.4	3.7			
562	716	3.41			
563	695	2.46			
564				707.9	
565	762.2	3.55			
566			628	630	
567	774.7	3.19			
568	712.7	2.3			
569				671.9	
570			656.1	658.2	
571	730.2	3.4			
572			639.1	641.2	
573				694.1	
574	634.5	1.7			
575	714.4	3.1			
576	680	2.2			
577	718	3.51			
578			680.5	682.4	
579	698	3.72			

Compound No.	LCMS (M+1)	LCMS RT	FIA-MS (M-1)	FIA-MS (M+1)	NMR
580	597	2.87			
581	720	3.51			
582	714.4	3.6			
583	693	3.35			
584				744	
585	762	3.68			
586	707	3.2			
587	730.5	3.68			
588	745.7	4.09			
589	735.20	3.70			
590	694.30	3.65			
591	651.30	3.26			
592	651.30	3.24			
593	685.20	3.53			
594	700.20	3.72			

[0538] Additional compounds, some of their physical data and method of synthesis are provided in Table 13.

Table 13

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
595	715.23		5B
596	689.24		5B
597	727.20		6
598	775.10		6
599	685.20		6
600		729.1	6
601	729.10		6
602	687.20		5B
603	673.17		5B
604	703.00		5B
605	716.00		5B
606	742.00		5B
607	676.00		5B
608	702.00		5B
609	676.00		5B
610	690.00		5B
611	716.00		5B
612	650.00		5B
613	676.00		5B
614	724.20		6
615	708.20		6
616		744.1	5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
617		684.1	5B
618	704.70		5B
619	677.60		5B
620		688	5B
621		662	5B
622		702	5B
623		676	5B
624		662	5B
625		636	5B
626		664	5B
627		638	5B
628		704	5B
629		678	5B
630		678	5B
631		652	5B
632	688.20		5B
633	686.10		5B
634	702.20		5B
635	700.10		5B
636	674.10		5B
637	688.20		5B
638	735.10		6

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
639	723.10		6
640	688.20		5B
641	702.20		5B
642	714.20		5B
643	728.10		6
644	651.21		5B
645	704.00		5B
646	678.00		5B
647	663.70		5B
648	689.70		5B
649	728.70		5B
650		690	5B
651		676	5B
652		690	5B
653		664	5B
654		650	5B
655	732.70		5B
656	718.70		5B
657	730.60		5B
658	700.10		5B
659	701.00		6
660	689.00		6
661		714	5B
662		728	5B
663		688	5B
664		702	5B
665		716	5B
666	673.10		6
667	701.19		5B
668	684.12		5B
669		708.2	6
670	687.70		5B
671	665.20		6
672	742.00		6
673	716.00		6
674	728.00		6
675	742.00		6
676	728.00		6
677	700.40		6
678	700.40		6
679	688.10		5B
680	662.10		5B
681	648.20		5B
682	690.10		5B
683	664.10		5B
684	650.10		5B
685	702.10		5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
686	676.10		5B
687	662.10		5B
688	674.10		5B
689	704.00		6
690	690.00		6
691		692.1	5A
692		706.1	5A
693		692.1	5A
694	690.04; Lot 2: 690.10		5B
695	690.04; Lot 2: 690.10		5B
696	704.62		5B
697	704.04		5B
698	676.06		5B
699	676.07		5B
700	675.00		6
701	663.00		6
702	634.00		5B
703	672.00		5B
704	686.10		5B
705	700.10		5B
706		740.1	5B
707	628.10		5B
708		740	5B
709		754	5B
710		728	5B
711		714	5B
712		682.2	5B
713		744	5B
714	696.10		5B
715		682.2	5B
716		670.1	5B
717	684.60		5B
718	658.80		5B
719	644.60		5B
720	670.60		5B
721	656.60		5B
722	682.60		5B
723	668.70		5B
724	694.70		5B
725	680.70		5B
726	696.70		5B
727	682.70		5B
728	708.70		5B
729	694.80		5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
730	682.70		5B
731	656.60		5B
732	642.70		5B
733	668.70		5B
734	654.70		5B
735	674.80		6
736	702.10		6
737	714.10		6
738	696.00		5B
739	696.00		5B
740		684.6	5A
741		708	5B
742	744.60		5B
743	718.70		5B
744	732.70		5B
745	706.60		5B
746	694.50		5B
747	650.10		5B
748	628.10		5B
749	628.10		5B
750	706.00		5B
751	684.10		5B
752	727.00		5B
753	716.00		5B
754	656.10		5B
755	656.10		5B
756	697.00		5B
757	697.00		5B
758	670.90		5B
759	656.90		5B
760	709.00		5B
761	682.90		5B
762	668.90		5B
763	721.00		5B
764	671.00		5B
765	656.90		5B
766	709.00		5B
767	682.90		5B
768	668.90		5B
769	700.00		5B
770	713.00		6
771	674.00		5B
772	660.10		5B
773	648.10		5B
774	634.10		5B
775	696.10		5B
776	682.10		5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
777	670.10		5B
778	656.10		5B
779	720.00		5B
780	734.00		5B
781		718.1	6
782		692.1	6
783		730.2	6
784		704	6
785	685.80		6
786	724.60		5B
787	710.41		5B
788	712.60		5B
789	698.60		5B
790	736.50		5B
791	750.60		5B
792	724.50		5B
793	764.60		5B
794	700.50		5B
795	714.50		5B
796	700.50		5B
797	714.50		5B
798	694.64		6
799	750.50		5B
800	724.50		5B
801	736.30		5B
802	727.80		6
803	772.00		6
804	693.60		5B
805	693.60		5B
806	707.70		5B
807	707.60		5B
808		698	5B
809		684	5B
810	720.00		5B
811	734.00		5B
812	652.90		6
813	658.00		5B
814	742.00		5B
815	728.00		5B
816	716.00		5B
817	758.60		6
818	758.50		6
819	728.60		6
820	640.80		6
821		722.7	5B
822		653.4	5B
823		684.6	5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
824		710.6	5B
825		698.5	5B
826	742.60		5B
827	716.50		5B
828	742.60		5B
829	728.60		5B
830	714.50		5B
831	680.60		5B
832	680.50		5B
833	738.50		5B
834	764.33		5B
835	684.00		5B
836	716.00		5B
837	704.00		5B
838	639.30		5B
839	670.50		5B
840	696.60		5B
841	684.50		5B
842	642.30		5B
843	628.30		5B
844	728.6		5B
845	728.70		5B
846	702.60		5B
847	714.30		6
848	728.30		6
849	682.30		5B
850	644.30		5B
851	682.30		5B
852	646.30		5B
853	682.30		5B
854	724.30		5B
855	748.00		5B
856	742.00		5B
857	672.00		5B
858	698.00		5B
859		710	5B
860		696	5B
861		684	5B
862	708.50		5B
863		698.3	11
864	731.38		6
865	726.30		5B
866	720.50		5B
867	680.60		5B
868	691.20		5B
869	691.20		5B
870	698.60		5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
871	710.60		5B
872	724.70		5B
873	718.50		5B
874	744.50		5B
875	730.40		5B
876	744.50		5B
877	686.50		5B
878	712.50		5B
879	698.50		5B
880	712.50		5B
881	696.50		5B
882	716.40		5B
883	684.50		5B
884	691.20		5B
885	691.20		5B
886	690.20		5B
887	690.10		5B
888	748.00		5B
889	722.00		5B
890	709.10		5B
891	720.40		5B
892	674.20		5B
893	688.30		5B
894	662.10		5B
895	687.70		5B
896	670.30		5B
897	694.20		5B
898	680.50		5B
899	720.10		5B
900	706.10		5B
901	706.20		5B
902	720.10		5B
903		679.3	11
904	654.20		11
905	704.50		5B
906	718.60		5B
907	718.60		5B
908	704.60		5B
909	656.20		5B
910	642.20		5B
911	748.00		5B
912	762.00		5B
913	736.00		5B
914	761.00		5B
915	672.20		5B
916	658.50		5B
917	694.50		5B

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
918	694.50		5B
919		710.2	5B
920		710.3	5B
921		684.2	5B
922		698.2	5B
923		696.2	5B
924		670.3	5B
925		696.2	5B
926		684.2	5B
927		684.2	5B
928		684.2	5B
929		658.4	5B
930		672.2	5B
931		670.4	5B
932		670.2	5B
933		644.3	5B
934		658.5	5B
935		605.3	11
936		667.3	11

Cmpd No.	LC/MASS PLUS	FIA MS+	Syn. Method
937	734.00		5B
938	722.00		5B
939		708	5B
940		734	5B
941		720	5B
942		732	5B
943		744	5B
944		720	5B
945		746	5B
946		732	5B
947		696	5B
948	744.60		5B
949	730.60		5B
950	708.20		5B
951	700.60		5B
952	696.20		5B
953	696.20		5B
954	694.10		5B
955	738.67		5B

Example No.	FIA MASS MINUS	FIA MASS PLUS	LC MASS MINUS	LC MASS PLUS	Syn. Method
957			686.41	688.06	5B
958	663.50	665.10		665.10	5B
959				706.60	5B
960				720.60	5B
961				704.00	5B
962			691.70	693.30	11
963				645.30	11
964				679.20	11
965				665.20	11
966				690.00	5B
967				709.60	5B
968				734.10	5B
969				708.10	5B
970				734.00	5B
971		706.00			5B
972		730.00			5B
973		718.00			5B
974		670.00			5B
975		694.00			5B
976		682.00			5B
977				672.00	5B
978				658.00	5B
979	770.70	772.10			5B
980	741.70	743.20			5B
981		743.10			5B

Example No.	FIA MASS MINUS	FIA MASS PLUS	LC MASS MINUS	LC MASS PLUS	Syn. Method
982	741.60	743.20			5B
983	706.60	708.20			5B
984	746.50	748.20			5B
985	756.60	758.20			5B
986	727.60	729.10			5B
987	727.50	729.20			5B
988	727.60	729.10			5B
989	692.50	694.20			5B
990	732.60	734.30			5B
991	744.50	746.10			5B
992	680.60	682.30			5B
993	720.60	722.30			5B
994			759.80; 759.60	760.20	5B
995				734.20	5B
996				760.20	5B
997				734.20	5B
998				746.20	5B
999			744.50	746.10	5B
1000	689.50	691.10		691.10	5B
1001	703.60	705.10		705.10	5B
1002	716.40	718.30		718.30	5B
1003	718.60	720.00		720.00	5B
1004	718.60	720.00		720.00	5B
1005				772.10	5B
1006			744.80	746.00	5B
1007	710.70	712.30			5B
1008	724.60	726.20			5B
1009	724.70	726.20			5B
1010	696.50	698.20			5B
1011	710.60	712.20			5B
1012	710.60	712.20			5B
1013	698.70	700.20			5B
1014		876.30			5B
1015	679.70	681.20			11
1016	713.50	715.10			11
1017	693.60	695.10			11
1018			743.60	745.70	5B
1019			676.60	678.00	11
1020	627.70	629.20			11
1021				629.20	11
1022	702.60	704.20			11
1023	626.60	628.30			11
1024				712.30	5B
1025				686.20	5B
1026				698.40	5B
1027				712.20	5B
1028				686.30	5B

Example No.	FIA MASS MINUS	FIA MASS PLUS	LC MASS MINUS	LC MASS PLUS	Syn. Method
1029				698.30	5B
1030				712.20	5B
1031				686.20	5B
1032				698.30	5B
1033				740.00	5B
1034				714.30	5B
1035				726.30	5B
1036				702.00	5B
1037				670.00	5B
1038				698.00	5B
1039				710.00	5B
1040				684.20	5B
1041	696.70	698.20			5B
1042	696.60	698.20			5B
1043	684.70	686.20			5B
1044	698.70	700.20			5B

V. ASSAYS FOR DETECTING AND MEASURING INHIBITION PROPERTIES OF COMPOUNDS

A. HCV Enzyme Assays

1. Construction and Expression of the HCV NS3 Serine Protease Domain

[0539] A DNA fragment encoding residues Ala^{1}-Ser^{181} of the HCV NS3 protease (GenBank CAB46913) was obtained by PCR from the HCV Con1 replicon plasmid, I_{377}neo/NS3-3'/wt (re-named as pBR322-HCV-Neo in this study) [V. Lohmann et al., Science, 285, pp. 110-113 (1999)] and inserted into pBEV11 (S. Chamber, et al., personal communication) for expression of the HCV proteins with a C-terminal hexa-histidine tag in E. coli. All constructs were confirmed by sequencing.

[0540] The expression constructs for the HCV NS3 serine protease domain was transformed into BL21/DE3 pLysS E. coli cells (Stratagene). Freshly transformed cells were grown at 37° C in a BHI medium (Difco Laboratories) pplemented with 100 μg/mL carbenicillin and 35 μg/mL chloramphenicol to an optical density of 0.75 at 600 nm. Induction with 1 mM IPTG was performed for four hours at 24 °C. The cell paste was harvested by centrifugation and flash frozen at -80 °C prior to protein purification. All purification steps were performed at 4 °C. Next, 100 g of cell paste was lysed in 1.5 L of buffer A (50 mM HEPES (pH 8.0), 300 mM NaCl, 0.1% n-octyl-β-D-glucopyranoside, 5 mM β-mercaptoethanol, 10% (v/v) glycerol) and stirred for 30 minutes. The lysate was homogenized using a Microfluidizer (Microfluidics, Newton, Mass.), followed by ultra-centrifugation at 54,000 x g for 45 minutes. Imidazole was added to the supernatant to a final concentration of 5 mM along with 2 mL of Ni-NTA resin pre-equilibrated with buffer A containing 5 mM imidazole. The mixture was rocked for three hours and washed with 20 column volumes of buffer A plus 5 mM imidazole. The HCV NS3 protein was eluted in buffer A containing 300 mM imidazole. The eluate was concentrated and loaded onto a Hi-Load 16/60 Superdex 200 column, pre-equilibrated with buffer A. The appropriate fractions of the purified HCV protein were pooled and stored at -80° C.

2. HCV NS3 Protease Domain Peptide Cleavage Assay

[0541] This assay is a modification of that described by Landro, et al. (Landro JA, Raybuck SA, Luong YC, O'Malley ET, Harbeson SL, Morgenstern KA, Rao G and Livingston DL. Biochemistry 1997, 36, 9340-9348), and uses a peptide substrate (NS5AB), based on the NS5A/NS5B cleavage site for genotype 1a HCV. The substrate stock solution (25 mM) was prepared in DMSO containing 0.2 M DTT and stored at -20° C. A synthetic peptide cofactor (KK4A) was used as a substitute for the central core region of NS4A. Peptide sequences are

shown in the table below. The reaction was performed in a 96-well microtiter plate format using 25 ηM to 50 ηM HCV NS3 protease domain in buffer containing 50 mM HEPES pH 7.8, 100 mM NaCl, 20% glycerol, 5 mM DTT and 25 μM KK4A. The final DMSO concentration was no greater than 2% v/v. Reactions were quenched by addition of trifluoroacetic acid (TFA) to yield a final concentration of 2.5%.

Peptide Sequences Used with HCV NS3 Protease Domain

Peptide	Sequence
NS5AB	NH_2-EDVV-(alpha)Abu-CSMSY-COOH [SEQ ID NO:2]
KK4A	NH_2-KKGSVVIVGRIVLSGK-COOH [SEQ ID NO:3]

[0542] The SMSY product was separated from substrate and KK4A using a microbore separation method. The instrument used was a Agilent 1100 with a G1322A degasser, either a G1312A binary pump or a G1311A quaternary pump, a G1313A autosampler, a G1316A column thermostated chamber and a G1315A diode array detector. The column was a Phenomenex Jupiter, 5 μm C18, 300 Å, 150x2 mm, P/O 00F-4053-B0, with a flow-rate of 0.2 mL/min. The column thermostat was at 40° C. Mobile phases were HPLC grade H_2O/0.1% TFA (solvent A) and HPLC grade CH_3CN/0.1% TFA (solvent B). The SMSY product peak was quantitated using the data collected at 210 ηM.

3. Construction and Expression of NS3•4A Protease

[0543] Using standard recombinant DNA techniques, a cDNA fragment encoding the sequence for NS3 and NS4A, residues Ala_{1027} to Cys_{1711} from the HCV sub-type strain 1a, containing an N-terminal hexa-histidine sequence, was cloned into the baculoviral transfer vector pVL1392 (Webb NR and Summers MD (1990) Expression of proteins using recombinant baculoviruses, Techniques 2:173-188). Recombinant baculovirus containing NS3•4A was produced by co-transfection of pVL1392-His–NS3•4A with linearized Autographa californica nuclear polyhedrosis virus (AcMNPV) DNA into Spodoptera frugoperda (Sf9) insect cells. The transfected insect cells containing recombinant baculovirus clones were subsequently isolated by plaque purification. High-titer clonal baculovirus was routinely used to infect Sf9 insect cells for protein production. In production, Sf9 cells were grown at 27 °C until they reached a density of 2.0-x10^6 cells/ml. At this point, the insect cells were infected with virus. After 72 hours or when the cell viability was between 70-80% the culture was harvested and the cells were ready for purification.

4. Purification of NS3•4A Protein

[0544] The NS3•4A protein (SEQ ID NO:1) was purified as follows. Cell paste was thawed in at least five volumes of Lysis Buffer (50 mM Na_2HPO_4 pH 8.0, 10% Glycerol, 300 mM NaCl, 5 mM β-mercaptoethanol, 0.2 mM PMSF, 2.5 μg/ml Leupeptin, 1.0 μg/ml E64, 2.0 μg/ml Pepstatin) per gram of cell paste. The cell paste was then homogenized on ice using a Dounce homogenizer. The cells were next mechanically disrupted by passing once through a microfluidizer (Microfluidics Corporation, Newton, MA), and the cell lysate was collected on ice. The cell lysates was centrifuged at 100,000 x g for 30 minutes at 4° C and the supernatants were decanted. Optionally, the pellets were resuspended in wash buffer (Lysis Buffer + 0.1% β-octyl glucopyranoside), homogenized using a Dounce homogenizer and centrifuged at 100,000 x g for 30 minutes at 4° C. Insoluble NS3•4A was extracted from the pellets by resuspending in Extraction Buffer (Lysis Buffer + 0.5% lauryl maltoside) using 2.5 ml/g cell paste. The mixture was homogenized using a Dounce homogenizer and mixed at 4 °C for three hours or more. The mixture was centrifuged at 100,000 x g for 30 minutes at 4 °C. The supernatants were decanted and pooled.

[0545] The NS3•4A protein was further purified using Nickel-NTA metal affinity chromatography. Imidazole from a 2 M stock, pH 8.0, solution was added to the pooled supernatants so that the final concentration of imidazole was 10 mM. The supernatants were incubated batchwise overnight at 4° C with Nickel-NTA affinity resin that had been pre-equilibrated with Lysis Buffer + 10 mM imidazole. 1 ml of resin per 5 μg of expected NS3-4A was used. The resin was next settled by gravity or by centrifugation at 500 x g for five minutes. The resin was next poured into a gravity flow column and washed with 10 or more column volumes of Nickel Wash Buffer (Lysis Buffer + 0.1% lauryl maltoside + 10 mM imidazole). The column was next eluted with three to four column volumes of Nickel Elution Buffer (Nickel Wash Buffer + 300 mM imidazole). The elution fractions were collected on ice and evaluated using SDS-PAGE. To prevent NS3-4A proteolysis, 100 μM DFP protease inhibitor was added to gel samples before adding SDS sample buffer and boiling. The peak fractions were pooled and protein concentration was determined by measuring absorbance at 280 ηm and by dividing by the extinction coefficient (e), which for NS3•4A is 1.01.

[0546] The NS3•4A was purified further using gel filtration chromatography. A Superdex 200 26/60 column was equilibrated with Superdex Buffer (20 mM HEPES pH 8.0, 10% glycerol, 300 mM NaCl, 10 mM β-mercaptoethanol, 0.05% lauryl maltoside) at a rate of 3 ml/min. The nickel purified NS3•4A was concentrated in a Centriprep 30 to greater than 2 mg/ml, if necessary, and was filtered through a 0.2 μm syringe filter and up to 10 ml was

loaded onto the Superdex 200 column. After 0.3 column volumes passed through, 4-5 ml fractions were collected. Fractions were evaluated by SDS-PAGE. NS3•4A protein elutes in two peaks. Peak 1 contains aggregated NS3•4A and peak 2 contains active protein. The fractions of peak 2 were pooled, aliquoted and frozen at -70° C.

Analysis of NS3•4A protein.

ANALYSIS	ENTIRE PROTEIN
Length	695 amino acids
Molecular Weight	74,347.78
1 microgram	13.450 picot moles
Molar Extinction Coefficient	73430
1 A_{280} corresponds to	1.01 mg/ml
Isoelectric Point	6.50
Charge at pH 7	-3.58

5. HCV NS3 Peptide Cleavage Assay

[0547] This assay follows the cleavage of a peptide substrate by full-length hepatitis C viral protein NS3•4A. One of three peptide substrates based on the NS5A/NS5B cleavage site for genotype 1a HCV is used to measure enzyme activity. All substrate stock solutions (25 mM) were prepared in DMSO containing 0.2M DTT and stored at –20 °C. A synthetic peptide cofactor (NS4A Peptide) was used to supplement NS4A. Peptide sequences are shown below. The hydrolysis reaction was performed in a 96-well microtiter plate format using 100 ηM to 125 ηM HCV NS3•4A in buffer containing 50 mM HEPES pH 7.8, 100 mM NaCl, 20% glycerol, 5 mM DTT and 25 μM NS4A Peptide. The final DMSO concentration was no greater than 2% v/v. Reactions using NS5AB or NS5AB-EDANS as substrate were quenched by the addition of 10% trifluoroacetic acid (TFA) to yield a final TFA concentration of 2.5%. Reactions using FITC-NS5AB-1 as substrate were quenched by the addition of 0.4M formic acid to yield a final concentration of 0.08M acid.

[0548] Enzymatic activity was assessed by separation of substrate and products by reverse phase HPLC. The instrument used was a Agilent 1100 with a G1322A degasser, either a G1312A binary pump or a G1311A quaternary pump, a G1313A autosampler, a G1316A column thermostated chamber, a G1321A fluorescence detector and a G1315A diode array detector. The column thermostat was at 40° C. For substrate NS5AB the column was a Phenomenex Jupiter, 5 μm C18, 300 Å, 150x2 mm, P/O 00F-4053-B0, with a flow-rate of 0.2 mL/min using HPLC grade H_2O/0.1% TFA (solvent A) and HPLC grade CH_3CN/0.1% TFA (solvent B) as mobile phases. The C-terminal product peak (NH2-SMSY -COOH) was quantitated using the absorbance data collected at 210 ηm. For substrate NS5AB-EDANS the column was a Phenomenex Aqua, 5 μm C18, 125 Å, 50x4.6 mm, P/O 00B-4299-E0, with

a flow-rate of 1.0 mL/min using HPLC grade H_2O/0.1% TFA (solvent A) and HPLC grade CH_3CN/0.1% TFA (solvent B) as mobile phases. The C-terminal product peak (NH2-SMSYT-Asp(EDANS)-KKK-COOH) was quantitated using the fluorescence data collected at 350 ηm excitation / 490 ηm emission. For substrate FITC-NS5AB-1 the column was a Phenomenex Prodigy, 5 μm ODS(2), 125 Å, 50x4.6 mm, P/O 00B-3300-E0, with a flow-rate of 1.0 mL/min using 10 mM sodium phosphate pH 7.0 in HPLC grade H_2O (solvent A) and 65% HPLC Grade CH_3CN / 35% 10 mM sodium phosphate pH 7.0 in HPLC grade H2O (solvent B) as mobile phases. The N-terminal product peak (FITC-Ahx-EDVV-(alpha)Abu-C-COOH) was quantitated using the fluorescence data collected at 440 nm excitation / 520 nm emission. Alternatively, the ratio of N-terminal product to unreacted FITC-NS5AB-1 substrate was determined using a Caliper LabChip 3000 with detection at 488 nm excitation / 530 nm emission, using a chip buffer of 100 mM Tris pH 7.0, 10 mM EDTA, 0.01% (v/v) Brij-35, and 0.1% (v/v) CR-3.

Peptide sequences used with HCV NS3.

Peptide	Sequence
NS4A Peptide	NH_2-KKGSVVIVGRIVLSGKPAIIPKK-COOH [SEQ ID NO:4]
NS5AB	NH_2-EDVV-(alpha)Abu-CSMSY-COOH [SEQ ID NO:2]
NS5AB-EDANS	NH_2-EDVV-(alpha)Abu-CSMSYT-Asp(EDANS)-KKK-COOH [SEQ ID NO:5]
FITC-NS5AB-1	FITC-Ahx-EDVV-(alpha)Abu-CSMSYTKK-NH_2 [SEQ ID NO:6]

6. Determination of Km and Vmax

[0549] To determine the kinetic parameters Km and Vmax, the HCV NS3 protease domain or HCV NS3•4A was reacted with peptide substrate under the assay conditions described above. Peptide substrate concentration was varied between 3 μM and 200 μM, with less than 20 percent conversion at all substrate concentrations. The ratio of the product peak area (as determined by reverse phase HPLC) to the reaction time yielded a rate of enzyme catalyzed hydrolysis. These *rate vs. substrate* concentration data points were fit to the Michaelis-Menten equation using non-linear regression. The value of k_{cat} was determined from Vmax using the nominal protease concentration and a fully cleaved substrate peptide as an instrument calibration standard.

Kinetic parameters for peptide substrates with HCV NS3 or NS3 protease domain.

Enzyme	Substrate	Km (μM)	k_{cat}/Km ($M^{-1}sec^{-1}$)

NS3 Protease Domain	NS5AB	25	3.0×10^4
NS3•4A	NS5AB	30	7.9×10^3
NS3•4A	NS5AB-EDANS	56	1.4×10^3
NS3•4A	FITC-NS5AB-1	15	1.2×10^3

7. Determination of Compound Potency

[0550] To evaluate apparent Ki values, all components except the test compound and substrate were pre-incubated for 5-10 minutes at room temperature. Then, test compound, dissolved in DMSO, was added to the mixture and incubated for either 15 minutes or 60 minutes at 30 °C. Neat DMSO was included as a no inhibitor control. The cleavage reaction was initiated by the addition of peptide substrate at a concentration either equal to Km or equal to one-half times Km, and allowed to proceed at 30 °C for 20 minutes. At the end of the reaction the mixture was quenched, and the extent of reaction was determined as described above. Eleven concentrations of compound were used to titrate enzyme activity for inhibition. Activity vs. inhibitor concentration data points were fit to the Morrison equation describing competitive tight-binding enzyme inhibition using non-linear regression (Sculley MJ and Morrison JF. *Biochim. Biophys. Acta*, 1986, 874, 44-53).

[0551] The tested compounds of formula I generally exhibited Ki values from about 0.008 to about 20 μM. In some embodiments, the compounds of formula I exhibited Ki values from about 0.008 to about 0.100 μM. In some other embodiments, the compounds of formula I exhibited Ki values from about 0.100 to about 0.500 μM. In still some other embodiments, the compounds of formula I exhibited Ki values from 0.500 to about 5.000 μM.

[0552] Examples of activities of the compounds of formulae (I, Ia, and Ib) on inhibiting serine protease receptors are shown below in Table 14. For compound activities for serine protease measured using the HCV Enzyme Assays, serine protease activity is illustrated with "+++" if activity was measured to be less than 0.1 μM, "++" if activity was measured to be from 0.1 μM to 0.5 μM, "+" if activity was measured to be greater than 0.5 μM, and "–" if no data was available. It should be noted that 0% efficacy is the minimum response obtained with the DMSO only control. The Enzyme Assay 1 refers to the HCV NS3 Protease Domain Peptide Cleavage Assay and Enzyme Assay 2 refers to the HCV NS3 Peptide Cleavage Assay.

Table 14. HCV Enzymatic Assay Activities and efficacies of exemplary compounds in accordance to Formulae I.

Compound No.	Enzyme Assay 1	Enzyme Assay 2
1	-	-
2	-	-
3	++	+++
4	++	
5	+++	+++
6	+	+
7	+	-
8	+	-
9	+	-
10	+	+
11	+	-
12	++	+++
13	+++	+++
14	+	-
15	++	-
16	+	-
17	+	++
18	+++	+++
19	++	+++
20	+	-
21	+	+
22	-	-
23	+++	++
24	++	-
25	+	-
26	++	+++
27	++	-
28	+	++
29	++	++
30	++	+++
31	-	++
32	++	-
33	+++	-
34	+	-
35	-	-
36	++	-
37	+	-
38	-	++
39	++	-
40	+	-
41	+++	-
42	++	+++
43	++	+++
44	++	-
45	+	++
46	+	+

Compound No.	Enzyme Assay 1	Enzyme Assay 2
47	+	-
48	-	-
49	-	+++
50	-	+++
51	+	-
52	++	-
53	+	-
54	+	++
55	++	-
56	++	-
57	-	-
58	+	+
59	-	+++
60	++	++
61	+	-
62	++	++
63	+	+
64	+	-
65	+	+
66	++	-
67	+++	+++
68	-	+++
69	+	++
70	+	-
71	+	-
72	++	-
73	+	++
74	++	+++
75	++	-
76	++	-
77	++	+++
78	++	-
79	++	+++
80	+	-
81	+	-
82	++	-
83	++	-
84	+	+
85	++	-
86	++	-
87	++	++
88	+	++
89	-	++
90	-	+++
91	+	+
92	++	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
93	++	+++
94	+++	-
95	++	-
96	+	++
97	-	+++
98	+	++
99	+	+
100	+	+
101	+	-
102	++	-
103	++	-
104	-	+++
105	-	+++
106	+	-
107	++	-
108	++	-
109	+++	-
110	++	-
111	++	++
112	++	+++
113	+	+
114	+++	+++
115	+	++
116	+	-
117	-	++
118	+	-
119	++	++
120	+	+
121	++	+++
122	++	-
123	+++	-
124	+	++
125	++	++
126	++	++
127	++	++
128	++	-
129	-	+++
130	++	+++
131	-	+++
132	++	++
133	++	++
134	++	+++
135	+	-
136	++	++
137	-	-
138	+	+

Compound No.	Enzyme Assay 1	Enzyme Assay 2
139	+	-
140	++	+++
141	+	++
142	+	-
143	++	++
144	+	+
145	++	-
146	+	-
147	+++	-
148	+	+
149	+	-
150	++	-
151	++	-
152	++	-
153	++	++
154	++	-
155	+	-
156	+++	-
157	+	+
158	+++	+++
159	++	-
160	+	-
161	+	-
162	++	+++
163	+++	+++
164	+	-
165	++	+++
166	++	-
167	++	-
168	++	++
169	-	++
170	-	+++
171	+	+
172	+	++
173	+++	-
174	++	-
175	-	-
176	++	++
177	+	++
178	++	++
179	++	-
180	+	-
181	+	+
182	+	+
183	++	-
184	-	+++

Compound No.	Enzyme Assay 1	Enzyme Assay 2
185	+	++
186	+	+
187	+	-
188	+	+
189	+	-
190	++	+++
191	+	-
192	-	-
193	++	-
194	+	-
195	-	+++
196	++	-
197	++	-
198	+	-
199	+	-
200	+	-
201	++	-
202	+	-
203	++	-
204	++	++
205	+	-
206	++	++
207	+	-
208	++	-
209	++	-
210	+	-
211	-	+++
212	++	+++
213	+	+
214	+	-
215	++	-
216	+	-
217	+++	-
218	+	+
219	-	+++
220	+	-
221	++	++
222	+	-
223	++	-
224	++	-
225	-	+++
226	++	-
227	+	+
228	+	-
229	+	-
230	++	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
231	++	-
232	-	-
233	++	+++
234	++	-
235	+	-
236	+	-
237	++	-
238	-	+
239	++	-
240	+	-
241	+	++
242	+++	-
243	+	-
244	++	-
245	++	-
246	+	+
247	+++	-
248	+	+
249	++	-
250	-	+++
251	+	-
252	++	+++
253	+	+
254	++	-
255	++	++
256	++	-
257	++	-
258	+	-
259	++	-
260	++	-
261	++	-
262	+	-
263	++	+++
264	-	+++
265	+	++
266	++	++
267	-	-
268	++	+
269	+	+
270	++	-
271	++	+++
272	+	+
273	++	-
274	+	-
275	++	++
276	+	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
277	+	++
278	+	-
279	++	-
280	+	+
281	+++	-
282	+	++
283	+	-
284	++	++
285	++	-
286	-	+++
287	++	-
288	++	-
289	+	++
290	++	-
291	+	-
292	++	+++
293	+	+
294	++	-
295	++	-
296	+	+
297	-	+++
298	++	++
299	+	-
300	++	++
301	++	-
302	++	++
303	+	-
304	++	+++
305	-	++
306	+	-
307	++	+++
308	++	-
309	+	+
310	+	+
311	+	-
312	+++	+++
313	+	-
314	++	-
315	+++	+++
316	++	-
317	+	-
318	++	-
319	++	-
320	+	+
321	++	+++
322	+	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
323	++	-
324	+	-
325	+	-
326	++	++
327	+	-
328	+	+
329	+	+
330	++	-
331	+	-
332	+	++
333	++	-
334	+	-
335	+	-
336	+	+
337	-	+++
338	+	++
339	++	+++
340	++	-
341	+	-
342	+	-
343	+	-
344	-	-
345	+	-
346	-	+++
347	+	+
348	+	-
349	++	-
350	+	+
351	+	+
352	+	-
353	+	-
354	+	-
355	+	+
356	-	+++
357	++	-
358	++	-
359	++	+++
360	+	-
361	++	+++
362	++	+++
363	+	-
364	+	-
365	++	-
366	++	+++
367	+	-
368	-	++

Compound No.	Enzyme Assay 1	Enzyme Assay 2
369	-	+++
370	++	+++
371	++	-
372	++	++
373	++	-
374	++	-
375	+	-
376	+	+
377	++	+++
378	++	-
379	+++	-
380	-	+
381	++	-
382	+++	-
383	+	-
384	+	+
385	+	-
386	-	-
387	++	-
388	+++	+++
389	+	-
390	++	+++
391	++	-
392	++	-
393	++	-
394	+++	-
395	+	+
396	-	+++
397	+	+
398	+	-
399	+	+
400	+++	-
401	++	++
402	++	-
403	+	-
404	+	++
405	+	-
406	-	+++
407	++	-
408	+	++
409	-	-
410	+	+
411	++	+++
412	++	++
413	+	-
414	+	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
415	++	-
416	+	++
417	+++	-
418	++	+++
419	++	++
420	+	+
421	-	+
422	++	-
423	+	-
424	++	-
425	+	-
426	+	++
427	+	-
428	++	-
429	+	+
430	++	+++
431	++	-
432	+	-
433	+	+
434	++	-
435	++	-
436	++	-
437	+	+
438	+	++
439	+	+
440	-	-
441	+	++
442	++	+++
443	+	+
444	++	-
445	++	+++
446	-	+++
447	+	++
448	++	-
449	++	-
450	+++	-
451	+	-
452	+	++
453	++	-
454	++	-
455	+	-
456	++	-
457	+	+
458	+	+
459	+	++
460	+	+

Compound No.	Enzyme Assay 1	Enzyme Assay 2
461	+	++
462	++	-
463	+	-
464	+	-
465	++	-
466	+	+
467	++	+++
468	-	+++
469	+	++
470	-	-
471	++	-
472	+	-
473	-	-
474	+	-
475	++	-
476	++	++
477	+++	-
478	++	-
479	-	-
480	+	-
481	+	-
482	+	-
483	+	+
484	++	-
485	-	+++
486	++	-
487	++	-
488	+	++
489	+	++
490	++	-
491	+	-
492	+++	-
493	++	+++
494	+++	-
495	+	-
496	+	+
497	++	-
498	++	-
499	+++	+++
500	++	+++
501	+	+
502	-	-
503	++	-
504	++	-
505	++	-
506	+	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
507	+	+
508	++	++
509	+	-
510	++	+++
511	++	-
512	++	-
513	-	+++
514	+	-
515	++	-
516	+	-
517	+	-
518	+++	-
519	++	-
520	+	-
521	++	+++
522	-	+++
523	++	-
524	+	-
525	+	-
526	++	-
527	++	-
528	++	-
529	-	+++
530	+	-
531	++	++
532	+	-
533	+	-
534	+	+
535	++	-
536	+	-
537	+	+
538	+	-
539	++	-
540	++	++
541	++	+++
542	+++	-
543	+	-
544	++	-
545	++	-
546	++	++
547	+++	-
548	++	-
549	+++	-
550	++	-
551	++	-
552	++	-

Compound No.	Enzyme Assay 1	Enzyme Assay 2
553	++	-
554	+	-
555	++	-
556	-	+++
557	++	-
558	++	-
559	+	+
560	++	-
561	++	-
562	-	+++
563	++	-
564	+	-
565	+	+
566	+	+
567	+	++
568	+	+
569	+++	-
570	++	-
571	++	-
572	++	-
573	+	+
574	+	+
575	+++	+++
576	++	-
577	-	+++
578	+	-
579	++	-
580	++	-
581	-	+++
582	++	-
583	++	-
584	+	-
585	++	-
586	+	+
587	+	++
588	+	+
589	-	-
590	-	-
591	-	-
592	-	-
593	-	-
594	-	-

Compound No.	Enzyme Assay 2
595	+++
596	++
597	++
598	++
599	++
600	++
601	+++
602	+++
603	+++
604	++
605	++
606	++
607	++
608	++
609	+++
610	+++
611	+++
612	+++
613	+++
614	+++
615	++
616	+++
617	++
618	+++
619	+++
620	+++
621	+++
622	++
623	+++
624	++
625	+++
626	+++
627	+++
628	++
629	+++
630	++
631	+++
632	+++
633	+++
634	+++
635	+++
636	+++
637	+++
638	+++
639	+++
640	+++
641	+++
642	+++

Compound No.	Enzyme Assay 2
643	+++
644	++
645	+++
646	+++
647	++
648	++
649	++
650	++
651	++
652	+++
653	++
654	+++
655	++
656	++
657	+++
658	++
659	-
660	+++
661	+++
662	++
663	++
664	+++
665	+++
666	+++
667	+++
668	+++
669	+++
670	+++
671	++
672	++
673	+++
674	+++
675	+++
676	+++
677	+++
678	++
679	+
680	+++
681	++
682	+++
683	+++
684	+++
685	+++
686	+++
687	+++
688	+++
689	+++
690	++

Compound No.	Enzyme Assay 2
691	++
692	++
693	++
694	++
695	+++
696	+++
697	+++
698	+++
699	+++
700	+++
701	++
702	++
703	+++
704	+++
705	+++
706	+++
707	++
708	+++
709	++
710	++
711	+++
712	+++
713	+++
714	-
715	+++
716	+++
717	+
718	+++
719	+++
720	+++
721	+++
722	+++
723	+++
724	+++
725	+++
726	+++
727	+++
728	+++
729	+++
730	+++
731	+++
732	++
733	+++
734	+++
735	+++
736	++
737	++
738	++

Compound No.	Enzyme Assay 2
739	+++
740	+++
741	+++
742	++
743	+++
744	+++
745	+++
746	+++
747	+++
748	+++
749	+++
750	+++
751	++
752	+++
753	+++
754	+++
755	+++
756	+++
757	+++
758	+++
759	+++
760	+++
761	+++
762	+++
763	+++
764	++
765	+
766	++
767	++
768	++
769	++
770	+++
771	+++
772	+++
773	++
774	++
775	+++
776	+++
777	+++
778	+++
779	+++
780	+++
781	+++
782	+++
783	+++
784	++
785	+++
786	++

Compound No.	Enzyme Assay 2
787	+++
788	+++
789	+++
790	+++
791	+++
792	+++
793	+++
794	+++
795	+++
796	+++
797	+++
798	+++
799	+++
800	+++
801	+++
802	+++
803	++
804	++
805	+++
806	+++
807	++
808	+++
809	+++
810	+++
811	+++
812	+++
813	+
814	+++
815	+++
816	+++
817	+++
818	+++
819	+++
820	+++
821	+
822	++
823	+++
824	+++
825	++
826	+++
827	+++
828	+++
829	+++
830	+++
831	+++
832	++
833	+++
834	+++

Compound No.	Enzyme Assay 2
835	+++
836	+++
837	++
838	+++
839	++
840	++
841	++
842	+++
843	++
844	++
845	+++
846	+++
847	+++
848	+++
849	+++
850	+++
851	+++
852	+++
853	+++
854	+++
855	+++
856	+++
857	+++
858	+++
859	+++
860	+++
861	+++
862	++
863	+++
864	++
865	+++
866	++
867	+++
868	+++
869	+++
870	++
871	+++
872	+++
873	+++
874	+++
875	+++
876	+++
877	+++
878	+++
879	+++
880	+++
881	+++
882	+++

Compound No.	Enzyme Assay 2
883	+++
884	+++
885	++
886	+++
887	++
888	+++
889	+++
890	+++
891	+++
892	+++
893	+++
894	+++
895	+++
896	+++
897	+++
898	+++
899	+++
900	+++
901	+++
902	+++
903	+++
904	+++
905	++
906	++
907	++
908	+++
909	+++
910	+++
911	+++
912	+++
913	+++
914	+++
915	+++
916	++
917	++
918	++
919	+++
920	++
921	+++
922	+++
923	+++
924	++
925	+++
926	+++
927	+++
928	++
929	+++
930	+++

Compound No.	Enzyme Assay 2
931	++
932	+++
933	+++
934	+++
935	+++
936	+
937	+++
938	+++
939	+++
940	++
941	+++
942	+++
943	+++
944	+++
945	+++
946	+++
947	+++
948	+++
949	+++
950	+++
951	+++
952	+++
953	+++
954	+++
955	+++
956	+++
957	++
958	+++
959	+++
960	+++
961	+
962	++
963	++
964	++
965	++
966	+
967	+++
968	+++
969	+++
970	+++
971	+++
972	+++
973	+++
974	+++
975	+++
976	+++
977	+++
978	+++

Compound No.	Enzyme Assay 2
979	-
980	++
981	+++
982	++
983	+++
984	+++
985	+++
986	++
987	-
988	-
989	-
990	-
991	+++
992	+++
993	+++
994	+++
995	+++
996	+++
997	+++
998	-
999	-
1000	-
1001	-
1002	-
1003	-
1004	-
1005	-
1006	-
1007	-
1008	-
1009	-
1010	-
1011	-
1012	-
1013	-
1014	-
1015	-
1016	-
1017	-
1018	-
1019	-
1020	-
1021	-
1022	-
1023	-
1024	-
1025	-
1026	-

Compound No.	Enzyme Assay 2
1027	-
1028	-
1029	-
1030	-
1031	-
1032	-
1033	-
1034	-
1035	-
1036	-
1037	-
1038	-
1039	-
1040	-
1041	-
1042	-
1043	-
1044	-

B. HCV Cell Assays

[0553] uh-7 cells were propagated in Dulbecco's modified Eagle's medium (DMEM, JRH Biosciences, Lenexa, Kansas) supplemented with 10% heat-inactivated FBS (fetal bovine serum), 2 mM L-glutamine, and nonessential amino acids (JRH). The cells were transfected with an in vitro transcribed HCV replicon RNA identical to replicon I377neo/NS3-3'/wt as described by Lohmann et al. (1999). Stable cell clones were selected and maintained in the presence of 250 μg/mL G418 (Invitrogen, Carlsbad, California). One of the clones, 24-2, was used in the subsequent HCV replicon assays. The replicon cells were propagated in DMEM supplemented with 10% FBS, 2 mM L-glutamine, nonessential amino acids, and 250 μg/mL G418. The cells were split twice per week in fresh media upon reaching confluence. There are approximately 200 – 300 copies of HCV RNA per replicon cell.

[0554] HCV replicon RNA from cells was measured using the Quantigene Discover XL kit (Panomics Inc., Fremont California) as per the manufacturer's instructions. Briefly, compound-treated replicon cells were lysed and immobilized on to capture plates using HCV specific oligonucleotides over night and the relative amounts of captured RNA was measured using oligonucleotide probe sets as per the manufacturer's instructions.

1. 2-Day HCV Replicon IC_{50} Assay

[0555] On the day prior to the assay, 104 replicon cells were plated per well of a 96-well plate and allowed to attach and grow overnight in DMEM (Invitrogen, Carlsbad, California) supplemented with 10 % heat-inactivated FBS (JRH Biosciences, Lenexa, Kansas), 2 mM L-glutamine (Invitrogen), nonessential amino acids (Invitrogen) and 250 μg/ml G418 (Invitrogen). Compounds were serially diluted in DMEM plus 2 % FBS and 0.5% DMSO (Sigma Chemical Co., St. Louis, MO) without G418. HCV replicon RNA from cells was measured using the Quantigene Discover XL kit (Panomics Inc., Fremont California) as per the manufacturer's instructions. Briefly, compound-treated replicon cells were lysed and immobilized on to capture plates using HCV specific oligonucleotides overnight and the relative amounts of captured RNA was measured using oligonucleotide probe sets as per the manufacturer's instructions. Unless indicated otherwise, each data point represents the average of three replicates. The IC_{50} is the concentration of the compound at which the HCV replicon RNA level in cells is reduced by 50 % as compared to the untreated replicon cell controls. To monitor the effect of compounds on cell proliferation or cell viability, replicon cells were treated with serially diluted compounds for 48 h, after which cell viability was determined using a CellTiter Glo assay (Promega, Madison, Wisconsin). Each CC_{50} is derived from three replicates and is the concentration of the compound at which the number

of viable cells is reduced by 50 % as compared to untreated cell controls. The IC_{50} and CC_{50} was determined using 4 parameter curve fitting in the SoftMax Pro program (Molecular Devices, Sunnyvale, California).

2. 5-Day HCV Replicon IC_{99} Assay

[0556] On the day prior to the assay, HCV replicon cells were plated at a low density of 2500 cells per well in a 96-well plate so the cells would not reach confluence during 5 days in culture. Compounds were serially diluted in DMEM containing 10 % FBS and 0.5 % DMSO in the absence of G418. Fresh media and compounds were added to the cells on day 1 and day 3. After the cells were treated with antiviral compounds for 5 days, HCV replicon RNA from cells was measured using the Quantigene Discover XL kit (Panomics Inc., Fremont California) as per the manufacturer's instructions. Briefly, compound-treated replicon cells were lysed and immobilized onto to capture plates using HCV specific oligonucleotides overnight and the relative amounts of captured replicon RNA was measured using oligonucleotide probe sets (Panomics) as per manufacturer's instructions. Each data point represents the average of two replicates. The IC_{99} is the concentration of the compound at which the HCV replicon RNA level in cells is reduced by 2 logs as compared to the untreated cell controls. To monitor the effect of compounds on cell proliferation or cell viability, replicon cells were treated with serially diluted compounds for 5 days, after which cell viability was determined using a CellTiter Glo assay (Promega, Madison, Wisconsin). Each CC_{50} is derived from two replicates and is the concentration of the compound at which the number of viable cells is reduced by 50% as compared to untreated cell controls. The IC_{99} and CC_{50} were determined by 4 parameter curve fitting method using the Prism software (GraphPad Software Inc., San Diego, California) and Excel program (Microsoft Corporation, Redmond, Washington).

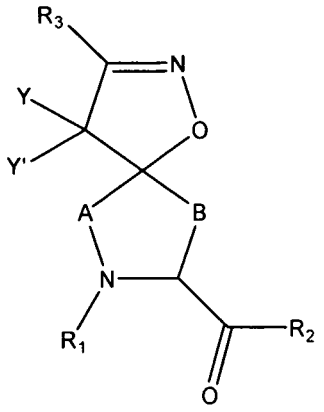
[0557] Using the assays above, compounds of the present invention are determined to be useful serine protease inhibitors.

OTHER EMBODIMENTS

[0558] It is to be understood that while the invention has been described in conjunction with the detailed description thereof, the foregoing description is intended to illustrate and not limit the scope of the invention, which is defined by the scope of the appended claims. Other aspects, advantages, and modifications are within the scope of the following claims.

WHAT IS CLAIMED IS:

1. A compound of formula (I),



(I)

or a pharmaceutically acceptable salt thereof, wherein:

each A is $-CH_2-$;

each B is $-CH_2-$;

each R_1 is $-Z^AR_4$, wherein Z^A is -C(O)- and R_4 is a substituted alkyl;

each R_2 is $-Z^BR_5$, wherein each Z^B is independently a bond or an optionally substituted aliphatic wherein up to four carbon units of Z^B are optionally and independently replaced by -C(O)-, $-C(O)NR^B-$, $-C(O)C(O)-NR^B-$, or $-NR^B-$;

each R_5 is independently R^B, halo, -OH, -CN, $-NO_2$, $-NH_2$, -alkoxy, or haloalkoxy;

each R^B is independently hydrogen, an optionally substituted aliphatic, or an optionally substituted cycloaliphatic; or

R_1 and R_2, together with the atoms to which they are attached, form an optionally substituted 6-membered heterocycloaliphatic ring;

each R_3 is an optionally substituted amino, $-O-R_{3A}$, an optionally substituted heterocycloaliphatic, an optionally substituted aryl, or an optionally substituted heteroaryl;

each R_{3A} is independently an optionally substituted aryl or an optionally substituted heteroaryl; and

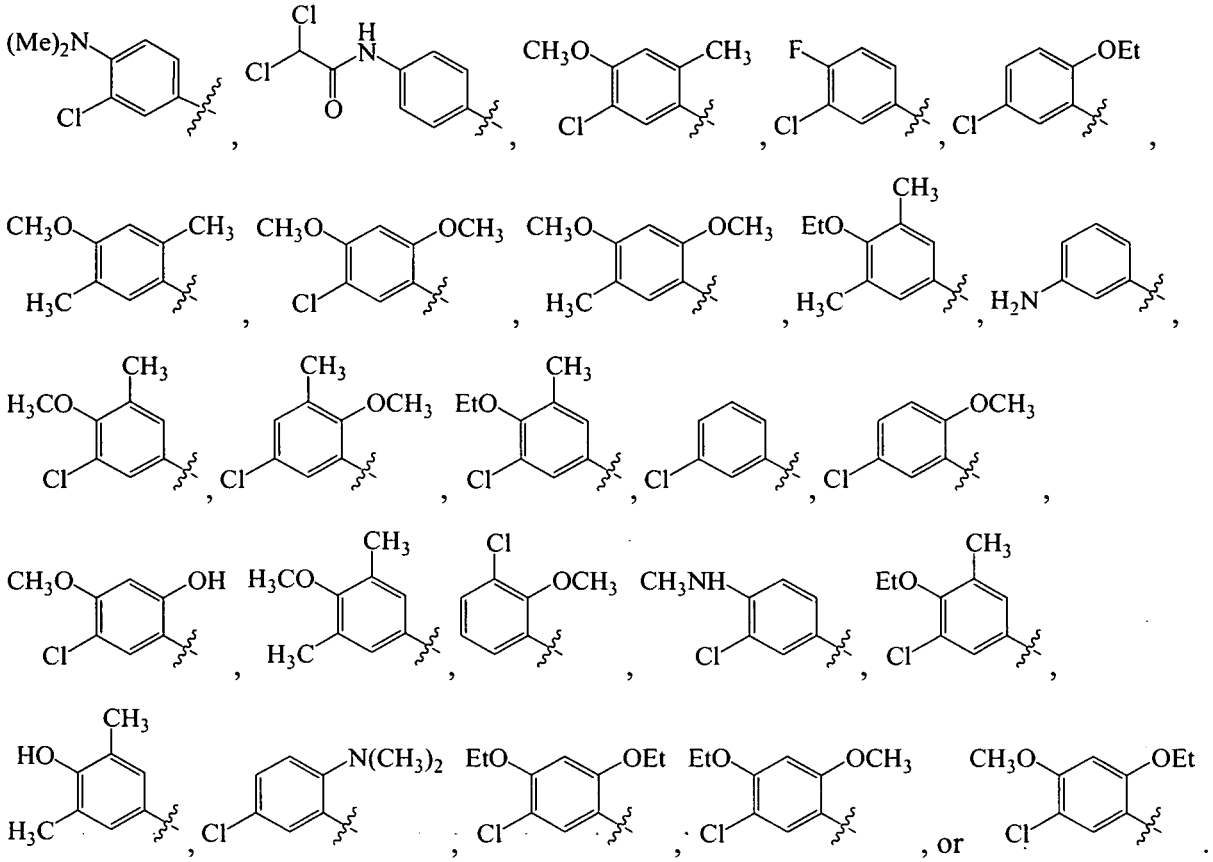
each of Y and Y' is H.

2. The compound of claim 1, wherein R_3 is an optionally substituted aryl.

3. The compound of claim 2, wherein R_3 is a monocyclic, bicyclic, or tricyclic aryl, and is optionally substituted with 1 to 3 substituents each independently selected from the group consisting of halo, hydroxy, cyano, amino, nitro, aliphatic, haloaliphatic, (aliphatic)oxy,

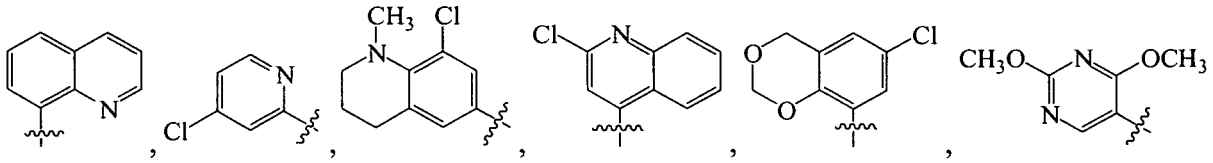
(halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, or heterocycloaliphatic.

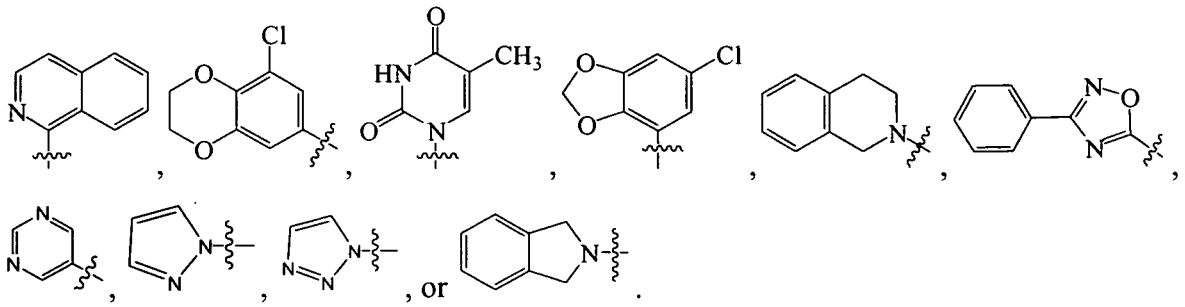
4. The compound of claim 3, wherein R_3 is



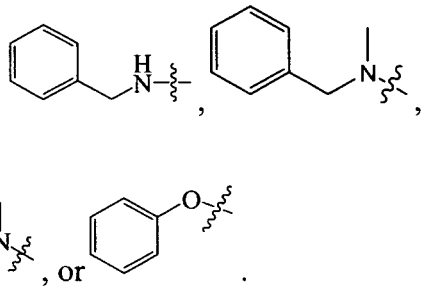
5. The compound of claim 1, wherein R_3 is a monocyclic, bicyclic, or tricyclic heteroaryl, and is optionally substituted with 1 to 3 substituents each independently selected from the group consisting of halo, hydroxy, cyano, amino, nitro, aliphatic, haloaliphatic, (aliphatic)oxy, (halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, or heterocycloaliphatic.

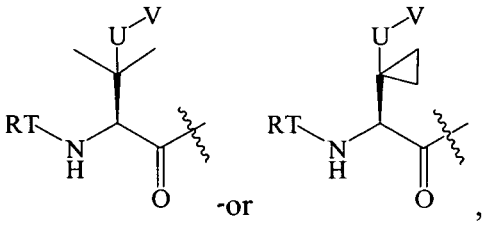
6. The compound according to claim 5, wherein R_3 is





7. The compound of claim 1, wherein R_3 is an optionally substituted amino, an optionally substituted heterocycloaliphatic, or an optionally substituted aryloxy.

8. The compound according to claim 7, wherein R_3 is 

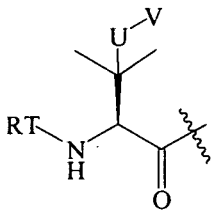
9. The compound of claim 1, wherein R_1 is  wherein:

U is a bond or -O-;

V is an alkyl;

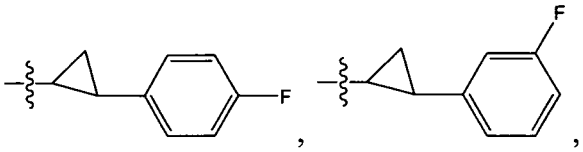
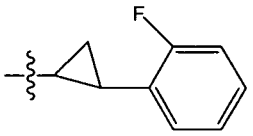
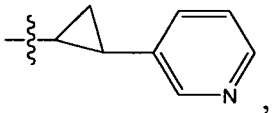
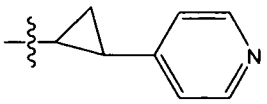
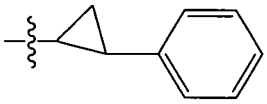
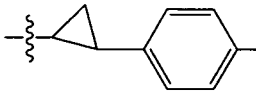
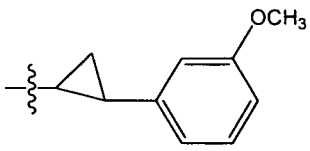
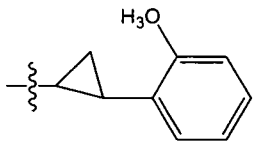
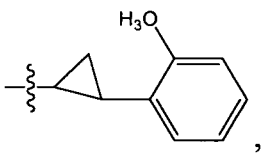
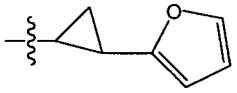
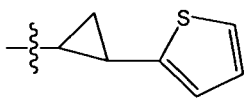
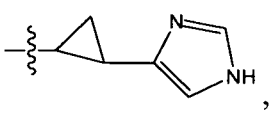
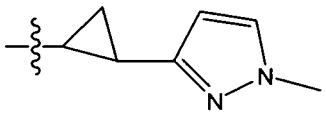
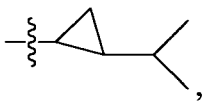
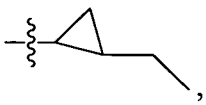
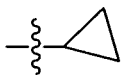
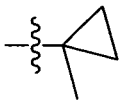
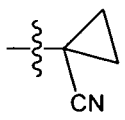
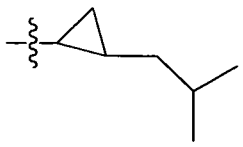
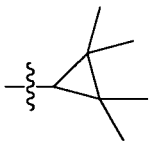
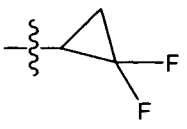
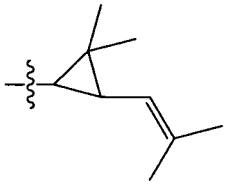
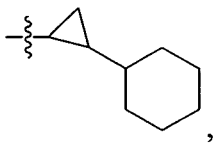
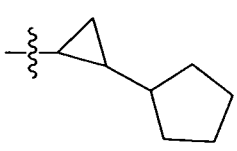
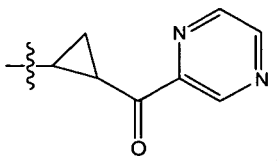
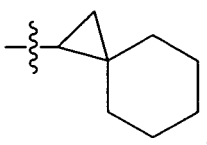
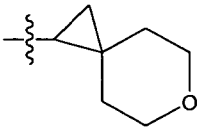
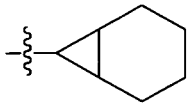
T is -C(O)-; and

R is an alkyl, cycloalkyl, heterocycloalkyl, aryl, heteroaryl, alkoxy, or (cycloalkyl)oxy, each of which is optionally substituted with 1 to 3 substituents each independently selected from the group consisting of halo, hydroxy, cyano, amino, nitro, aliphatic, haloaliphatic, (aliphatic)oxy, (halo(aliphatic))oxy, (aliphatic(oxy(aryl)))oxy, aryl, heteroaryl, haloaryl, cycloaliphatic, or heterocycloaliphatic.

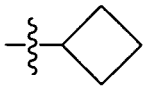
10. The compound of claim 9, wherein R_1 is  , and V is $-CH_3$.

11. The compound of claim 10, wherein R is cyclopropyl with 1-4 optional substituents.

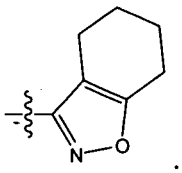
12. The compound of claim 10, wherein each of the 1-4 optional substituent on cycloproyl is independently aryl, heteroaryl, heterocycloalkyl, cycloalykl, alkyl, alkenyl, cyano, or halo, and attached to cyclopropyl via an optional carbonyl linker; or two of the optional substituents, together with the atom or atoms to which they are attached, form heterocycloalkyl or cycloalkyl.

13. The compound of claim 10, wherein R is  , ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  ,  , , ,  ,  ,  ,  ,  ,  ,  , or .

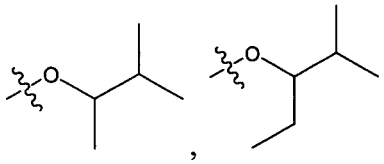
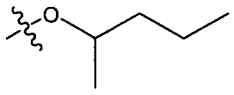
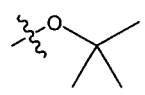
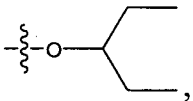
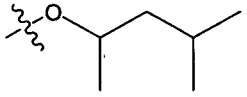
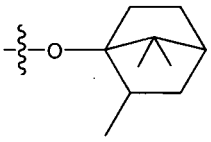
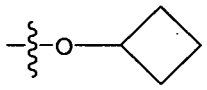
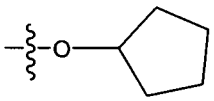
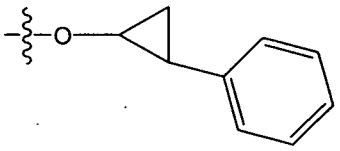
14. The compound of claim 10, wherein R is optionally substituted cyclobutyl or optionally substituted heterocyclobutyl.

15. The compound of claim 14, wherein R is  .

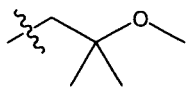
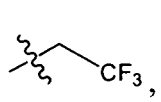
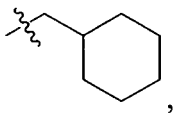
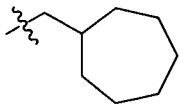
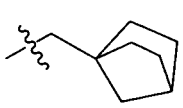
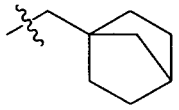
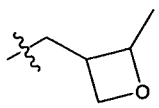
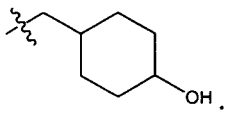
16. The compound of claim 10, wherein R is an optionally suibstituted heteroaryl.

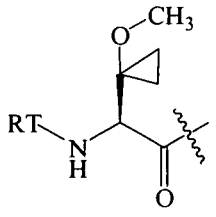
17. The compound of claim 16, wherein R is  .

18. The compound of claim 10, wherein R is an optionally substituted alkoxy or optionally substituted cycloalkyloxy.

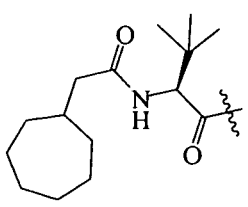
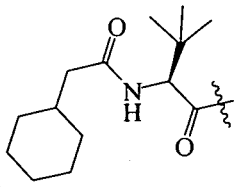
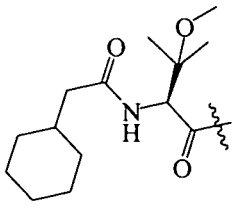
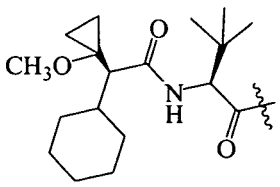
19. The compound of claim 18, wherein R is  ,  ,  ,  ,  ,  ,  ,  , or  .

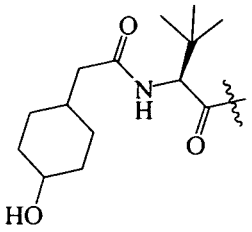
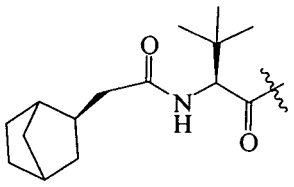
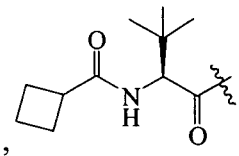
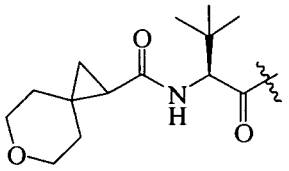
20. The compound of claim 10, wherein R is optionally substituted alkyl.

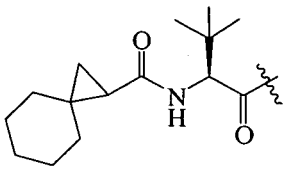
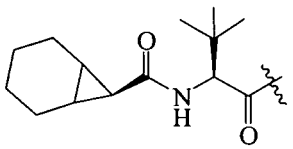
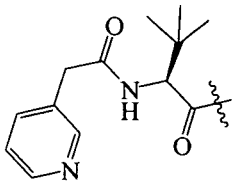
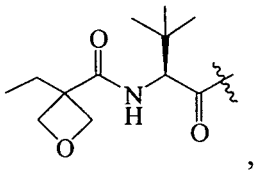
21. The compound of claim 20, wherein R is  , ,  ,  ,  ,  ,  ,  , or  .

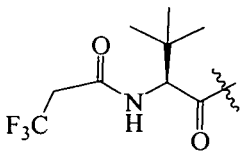
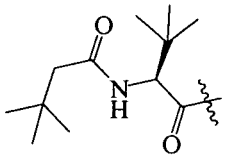
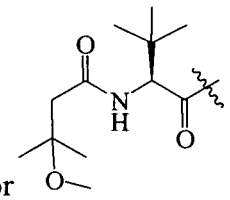
22. The compound of claim 9, wherein R_1 is  .

23. The compound of claim 9, wherein R_1 is

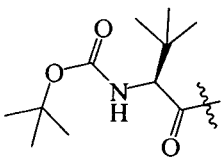
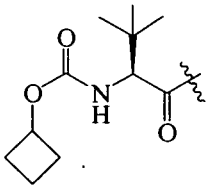
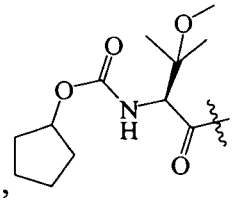
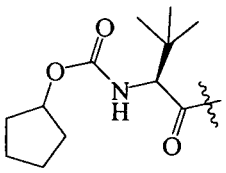
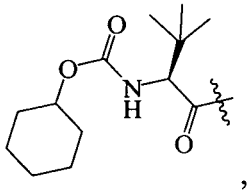
 ,  ,  ,  ,

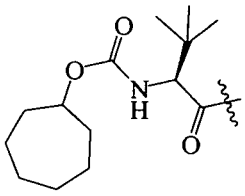
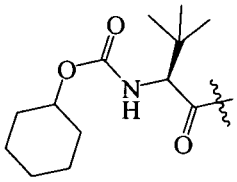
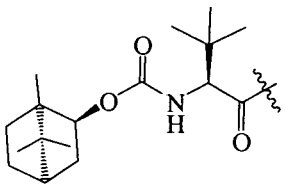
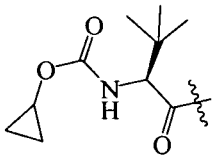
 ,  ,  ,  ,

 ,  ,  ,  ,

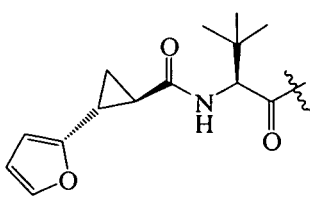
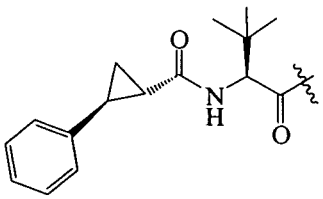
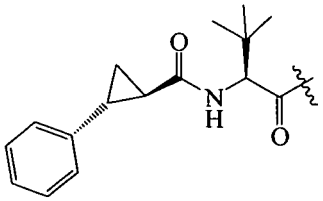
 ,  , or  .

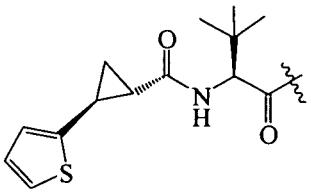
24. The compound of claim 9, wherein R_1 is

 ,  ,  ,  ,  ,

 ,  ,  , or  .

25. The compound of claim 9, wherein R_1 is

 ,  ,  ,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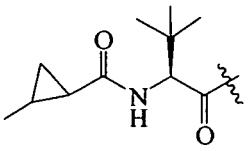
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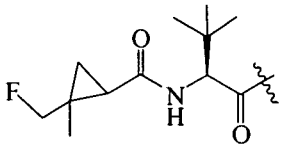
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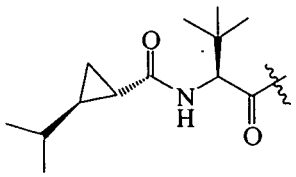
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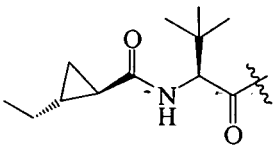
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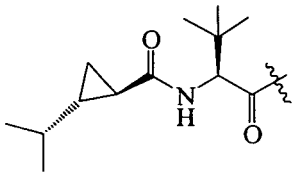
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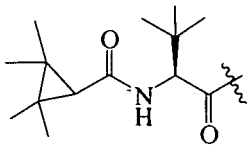
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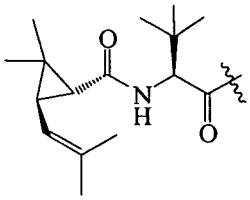
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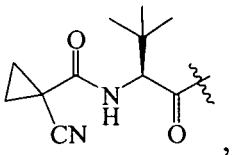
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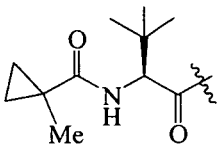
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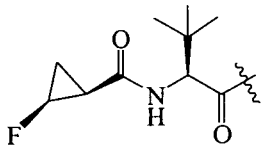
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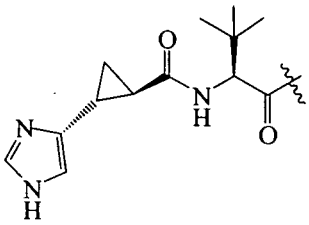
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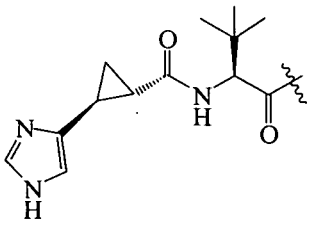
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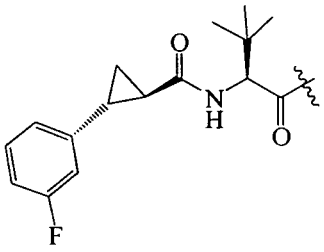
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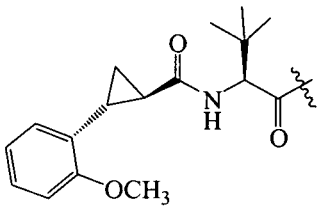
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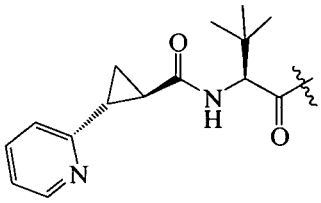
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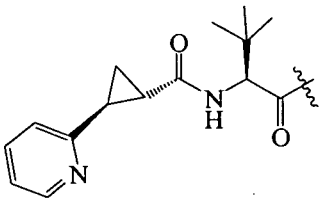
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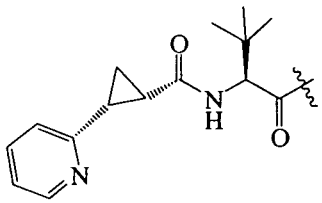
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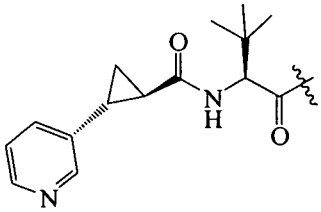
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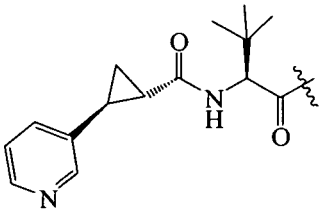
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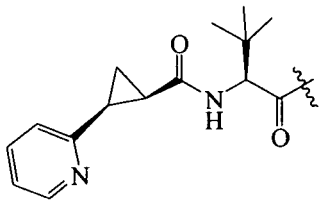
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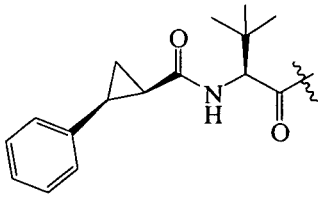
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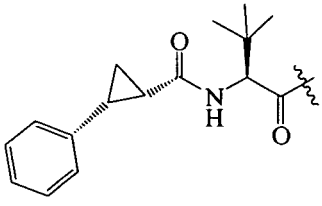
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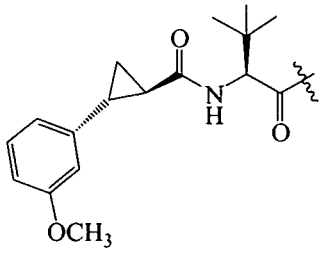
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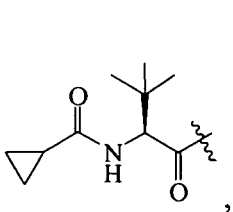
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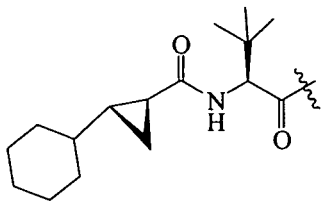
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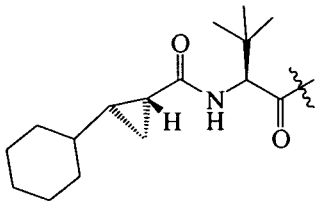
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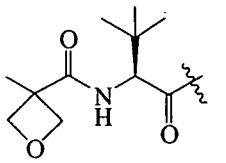
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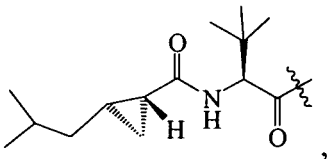
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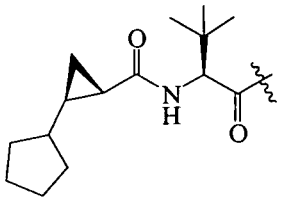
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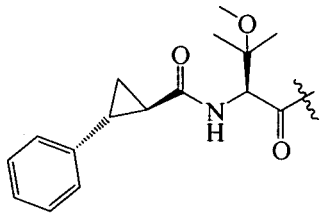
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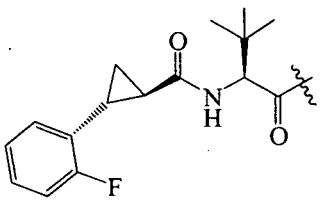
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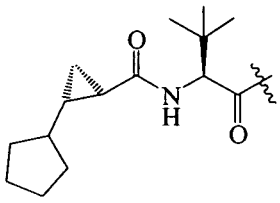
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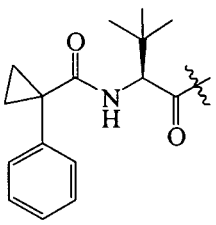
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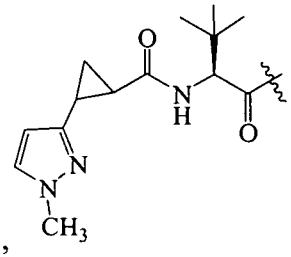
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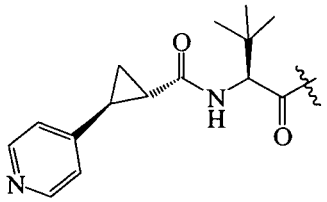
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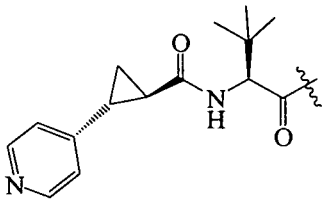
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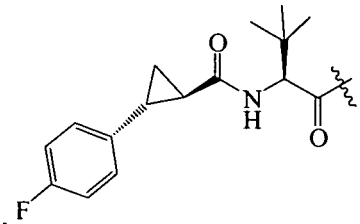
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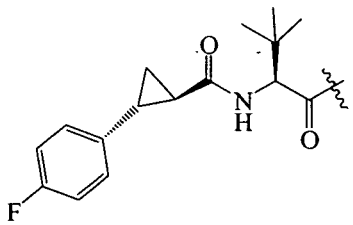
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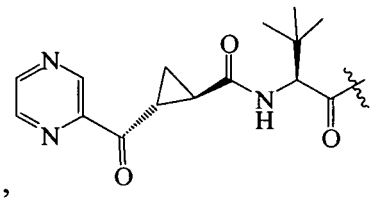
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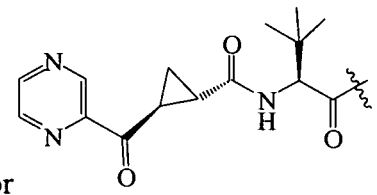
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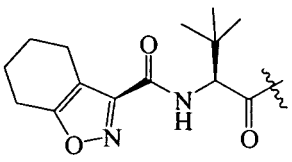
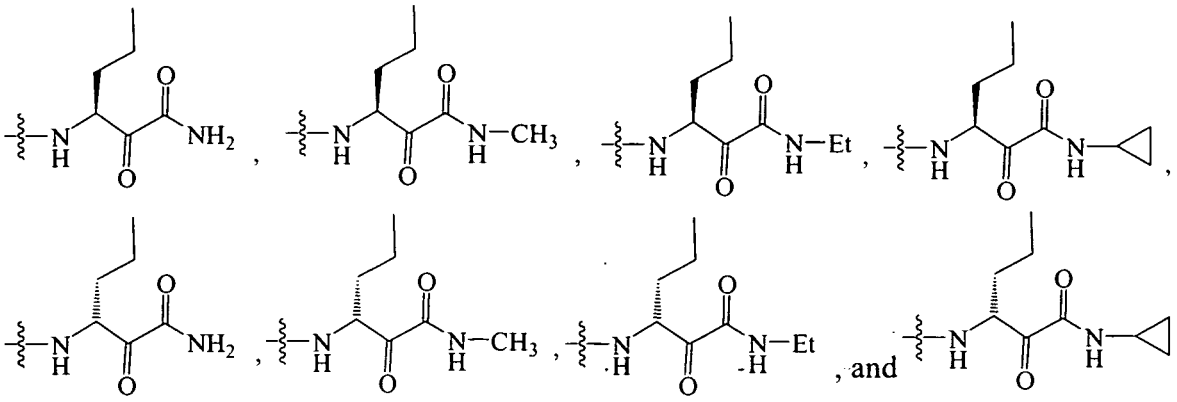
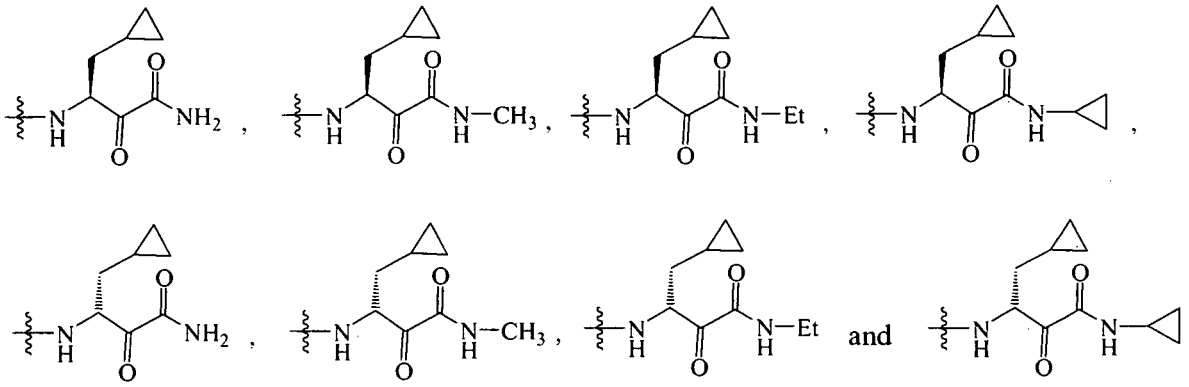
26. The compound of claim 9, wherein R_1 is  .

27. The compound of claim 1, wherein R_2 is a (cycloaliphatic(carbonyl(carbonyl(aliphatic))))amino, (amino(carbonyl(carbonyl(aliphatic))))amino, or (aliphatic(carbonyl(carbonyl(aliphatic))))amino, each of which is optionally substituted.

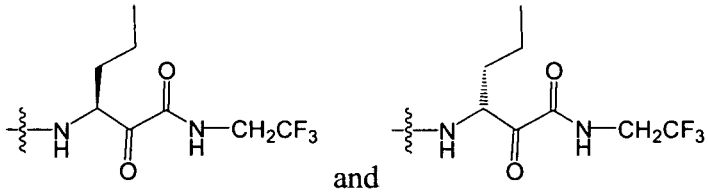
28. The compound of claim 27, wherein R_2 is selected from the group consisting of



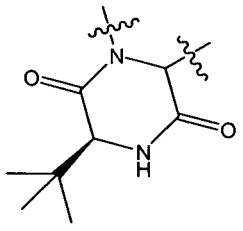
29. The compound of claim 27, wherein R_2 is selected from the group consisting of



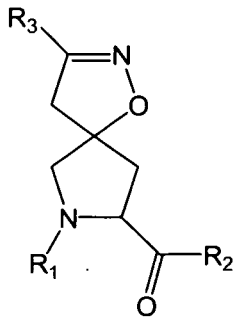
30. The compound of claim 27, wherein R_2 is selected from the group consisting of



31. The compound of claim 1, wherein R_1 and R_2, together with the atoms to which they are attached, form



32. A compound of formula (II), wherein



(II)

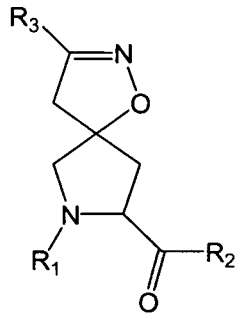
each R_1 is a (cycloalkyl(alkyl)carbonyl))amino)(alkyl)carbonyl;

each R_2 is a (cycloalkylamino(carbonyl))(carbonyl)alkyl))amino or (alkylamino(carbonyl))(carbonyl)alkyl))amino; and

each R_3 is an optioanlly substituted aryl.

33. The compound of claim 32, wherein R_3 is phenyl substituted with 1 to 3 substituents each independently selected from the group consisting of C_{1-4} alkoxy, C_{1-4} alkyl, and halo.

34. A compound of formula (II), wherein



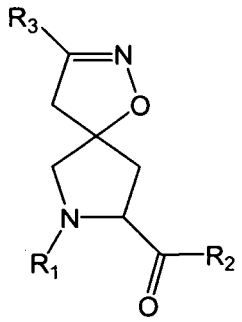
(II)

each R_1 is a (cycloalkyl(alkyl)carbonyl))amino)(alkyl)carbonyl;

each R_2 is a (cycloalkylamino(carbonyl))(carbonyl)alkyl))amino or (alkylamino(carbonyl))(carbonyl)alkyl))amino; and

each R_3 is a heteroaryl optionally substituted with 1 to 3 substituents each independently selected from the group consisting of C_{1-4} alkoxy, C_{1-4} alkyl, and halo.

35. A compound of formula (II), wherein



(II)

each R_1 is a (cycloalkyl(alkyl)carbonyl))amino)(alkyl)carbonyl;

each R_2 is a (cycloalkylamino(carbonyl))(carbonyl)alkyl))amino or (alkylamino(carbonyl))(carbonyl)alkyl))amino; and

each R_3 is an optionally substituted amino, optionally substituted heterocycloaliphatic, or optionally substituted aryloxy.

36. A compound selected from the group consisting of compound numbers 595 through 1044.

37. A pharmaceutical composition comprising a compound of claim 1 or claim 36, or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier, adjuvant or vehicle.

38. The composition according to claim 37, further comprising an additional agent selected from an immunomodulatory agent; an antiviral agent; a second inhibitor of HCV protease; an inhibitor of another target in the HCV life cycle; and a cytochrome P-450 inhibitor.

39. The composition according to claim 38, wherein said immunomodulatory agent is α–, β–, or γ–interferon or thymosin; said antiviral agent is ribavirin, amantadine, or telbivudine; and said inhibitor of another target in the HCV life cycle is an inhibitor of HCV helicase, polymerase, or metalloprotease.

40. The composition according to claim 38, wherein said cytochrome P-450 inhibitor is ritonavir.

41. A method of inhibiting a serine protease in a cell, comprising contacting the cell with a compound of claim 1 or claim 36.

42. The method according to claim 41, wherein said serine protease is an HCV NS3 protease.

43. A method of treating an HCV-infected patient, comprising administering to said patient a compound of claim 1 or claim 36.

44. The method according to claim 43, further comprising administering to said patient an additional agent selected from an immunomodulatory agent, an antiviral agent, a second inhibitor of HCV protease, and an inhibitor of another target in the HCV life cycle.

45. The method according to claim 44, wherein said immunomodulatory agent is α–, β–, or γ–interferon or thymosin; said antiviral agent is ribavarin or amantadine; and said inhibitor of another target in the HCV life cycle is an inhibitor of HCV helicase, polymerase, or metalloprotease.

46. A method of eliminating or reducing HCV contamination of a biological sample or medical or laboratory equipment, comprising the step of contacting said biological sample or medical or laboratory equipment with a compound of claim 1 or claim 36.