COMPOSITIONS FOR USE IN INHIBITING SRC KINASE AND TREATING AND PREVENTING ASSOCIATED DISORDERS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of U.S. provisional patent application no. 62/901,540, filed on September 17 2019, which is incorporated herein by reference in its entirety.

FIELD OF THE INVENTION

[0002] The present invention encompasses compounds and composition that inhibit Src kinase and methods of treating or preventing disorders associated therewith. In some embodiments, the invention encompasses compositions and combinations of agents that act synergistically inhibit the growth of cancer cells and particularly the compositions and combinations can be used for the treatment of cancer.

BACKGROUND OF THE INVENTION

[0003] The National Cancer Institute has estimated that in the United States alone, 1 in 3 people will be struck with cancer during their lifetime. The widespread occurrence of this disease underscores the need for improved anticancer regimens for the treatment of malignancy.

[0004] *Vernonia cinerea* (family Asteracea) also commonly called as Sahadevi, a species native to tropical Asia and Africa. This branched herb is about 0.5 to 3 feet high, found throughout India. The different parts of the plant such as stems, seed, leaves, roots and flowers have been mentioned in ancient texts and in alternative medicines. Several studies from Asian sub-continent have reported the use of Sahadevi as anthelmintic, antibacterial, antiviral, antifungal, anti-inflammatory, diuretic, and stomachic.

[0005] The phytochemical screening of the whole plant extract revealed the presence of triterpene compounds such as beta amyrin acetate, lupeol acetate; sterols such as beta-sitosterol, stigmasterol and alpha-spinasterol and phenolic resins. Recently, the anti-tumour activity of the plant extract has also been established. However, the identification of the biologically active compound(s) responsible for the anti-tumor activity has been elusive.

[0006] The inventors have identified the active compound and certain derivatives responsible for the anti-tumor activity from the crude extract of Sahadevi by utilizing various extraction and bio-analytical tools.

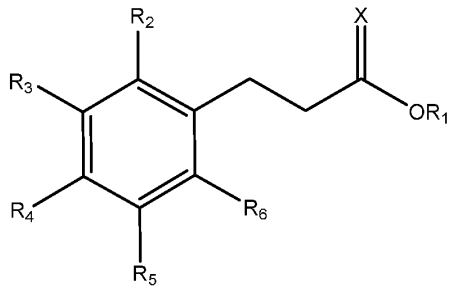
SUMMARY OF THE INVENTION

[0007] The invention generally encompasses compounds that inhibit a specific tyrosine kinase (*i.e.,* Src kinase) and compositions including such compounds as well as combinations of such compositions with known cancer drugs to treat and prevent cancer.

[0008] In another embodiment, the invention encompasses methods of use of the compositions including active compounds or derivatives and metabolites thereof in any disease conditions where Src tyrosine kinase or Src family kinases (SFKs) activity has been implicated, for example, but not limited to, the development, maintenance, progression, and metastatic spread of cancers.

[0009] In certain embodiments, the compound and compositions are effective against human cancer and tumor cells including, but not limited to, those associated with chronic myeloid leukemia (CML), acute myeloid leukemia (AML), acute lymphocytic leukemia (ALL), breast cancer (e.g., Triple negative, HER2+ etc), and colon cancer.

[0010] In one embodiment, the Compounds Of The Invention includes compounds of the following structure:



Formula I

[0011] wherein X is O or S;

[0012] R_1 is a hydrogen, or a substituted or unsubstituted substituent including but not limited to lower alkyl, a lower alkenyl, a lower alkynyl, $-(CH_2)_mR_7$, $(CH_2)_m$-OH, $-(CH_2)_m$-O-lower alkyl, $-(CH_2)_m$-O-lower alkenyl, $-(CH_2)_n$-O-$(CH_2)_m$-R_7, $-(CH_2)_m$-SH, $-(CH_2)_m$-S-lower alkyl, $-(CH_2)_m$-S-lower alkenyl, $-(CH_2)_n$-S-$(CH_2)_m$-R_7

[0013] each of R_2-R_6 is independently a hydrogen, a hydroxyl, a halogen, a lower alkyl, a lower alkenyl, a lower alkynyl, an amino, a nitro, an azido, a sulfate, a sulfonate, a sulfonamido, $-(CH_2)_mR_7$, $(CH_2)_m$-OH, $-(CH_2)_m$-O-lower alkyl, $-(CH_2)_m$-O-lower alkenyl, $-(CH_2)_n$-O-$(CH_2)_m$-R_7, $--(CH_2)_m$-SH, $-(CH_2)_m$-S-lower alkyl, $-(CH_2)_m$-S-lower alkenyl, $-(CH_2)_n$-S-$(CH_2)_m$-R_7

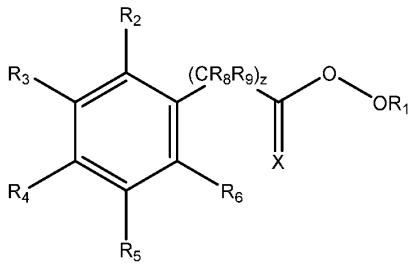
[0014] R_7 represents, for each occurrence, hydrogen, hydroxyl, or a substituted or unsubstituted

alkyl, alkenyl, aryl, aralkyl, benzyl, cycloalkyl, cycloalkenyl, or heterocycle; and

[0015] wherein each occurrence of m is independently an integer ranging from 1 to 9, and each occurrence of n is independently an integer ranging from 1 to 9;

[0016] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

[0017] In another embodiment, the Compounds Of The Invention includes compounds of the following structure:



Formula II

[0018] wherein X is O or S;

[0019] R_1 is a hydrogen, or a substituted or unsubstituted substituent including but not limited to lower alkyl, a lower alkenyl, a lower alkynyl, -$(CH_2)_mR_7$, $(CH_2)_m$-OH, -$(CH_2)_m$-O-lower alkyl, -$(CH_2)_m$-O-lower alkenyl, -$(CH_2)_n$-O-$(CH_2)_m$-R_7, -$(CH_2)_m$-SH, -$(CH_2)_m$-S-lower alkyl, -$(CH_2)_m$-S-lower alkenyl, -$(CH_2)_n$-S-$(CH_2)_m$-R_7

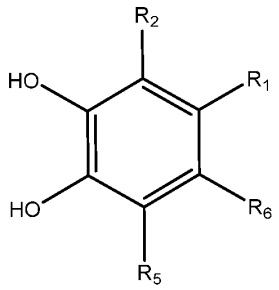
[0020] each of R_2-R_6 is independently a hydrogen, a hydroxyl, a halogen, a lower alkyl, a lower alkenyl, a lower alkynyl, an amino, a nitro, an azido, a sulfate, a sulfonate, a sulfonamido, -$(CH_2)_mR_7$, $(CH_2)_m$-OH, -$(CH_2)_m$-O-lower alkyl, -$(CH_2)_m$-O-lower alkenyl, -$(CH_2)_n$-O-$(CH_2)_m$-R_7, --$(CH_2)_m$-SH, -$(CH_2)_m$-S-lower alkyl, -$(CH_2)_m$-S-lower alkenyl, -$(CH_2)_n$-S-$(CH_2)_m$-R_7

[0021] R_7, R_8, and R_9 each independently represents, for each occurrence, hydrogen, hydroxyl, or a substituted or unsubstituted alkyl, alkenyl, aryl, aralkyl, benzyl, cycloalkyl, cycloalkenyl, or heterocycle; and

[0022] wherein each occurrence of m and n is separately and independently an integer ranging from 1 to 9, and each occurrence of z is independently an integer ranging from 1 to 9;

[0023] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

[0024] In another embodiment, , the Compounds Of The Invention includes compounds of the following structure:



Formula III

[0025] R_1 is a hydrogen, or a substituted or unsubstituted substituent including but not limited to lower alkyl, a lower alkenyl, a lower alkynyl, $-(CH_2)_mR_7$, $-(CH_2)_m$-OH, $-(CH_2)_m(=X)XR_7$, $-(CH_2)_m(=X)R_7$, $-(CH_2)_m$-X-lower alkyl, $-(CH_2)_m$-X-lower alkenyl, $-(CH_2)_n$-X-$(CH_2)_m$-R_7, $-(CH_2)_m$-XR_7

[0026] wherein X is O or S;

[0027] each of R_2, R_5, and R_6 is independently a hydrogen, a hydroxyl, a halogen, a lower alkyl, a lower alkenyl, a lower alkynyl, an amino, a nitro, an azido, a sulfate, a sulfonate, a sulfonamido, $-(CH_2)_mR_7$, $(CH_2)_m$-OH, $-(CH_2)_m$-O-lower alkyl, $-(CH_2)_m$-O-lower alkenyl, $-(CH_2)_n$-O-$(CH_2)_m$-R_7, $--(CH_2)_m$-SH, $-(CH_2)_m$-S-lower alkyl, $-(CH_2)_m$-S-lower alkenyl, $-(CH_2)_n$-S-$(CH_2)_m$-R_7

[0028] R'_7 represents, for each occurrence, hydrogen, hydroxyl, or a substituted or unsubstituted alkyl, acyl, alkenyl, aryl, aralkyl, benzyl, cycloalkyl, cycloalkenyl, or heterocycle; and

[0029] wherein each occurrence of m is independently an integer ranging from 1 to 9, and each occurrence of n is independently an integer ranging from 1 to 9;

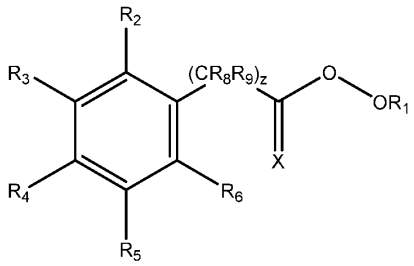
[0030] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

[0031] In certain embodiments, R_1 is a straight chain or branched alkyl having six or fewer carbon atoms (e.g., C_1-C_6 for straight chain, C_3-C_6 for branched chain), and in another embodiment, a straight chain or branched alkyl has four or fewer carbon atoms.

[0032] The present invention relates to a method of treating or preventing cancer by administering a composition including a compound of Formula (I) or (II) or a (III) pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, to a subject in need thereof, where administration of the composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, results in

one or more of the following: prevention of cancer cell proliferation by accumulation of cells in one or more phases of the cell cycle (e.g. G1, G1/S, G2/M), or induction of cell senescence, or promotion of tumor cell differentiation; promotion of cell death in cancer cells via cytotoxicity, necrosis or apoptosis, without a significant amount of cell death in normal cells, antitumor activity in animals with a therapeutic index of at least 2. As used herein, "therapeutic index" is the maximum tolerated dose divided by the efficacious dose.

[0033] In certain embodiments, the invention encompasses methods of modulating a Src kinase comprising administering a following structure:



Formula II

[0034] wherein X is O or S;

[0035] R_1 is a hydrogen, or a substituted or unsubstituted substituent including but not limited to lower alkyl, a lower alkenyl, a lower alkynyl, -$(CH_2)_mR_7$, $(CH_2)_m$-OH, -$(CH_2)_m$-O-lower alkyl, -$(CH_2)_m$-O-lower alkenyl, -$(CH_2)_n$-O-$(CH_2)_m$-R_7, -$(CH_2)_m$-SH, -$(CH_2)_m$-S-lower alkyl, -$(CH_2)_m$-S-lower alkenyl, -$(CH_2)_n$-S-$(CH_2)_m$-R_7

[0036] each of R_2-R_6 is independently a hydrogen, a hydroxyl, a halogen, a lower alkyl, a lower alkenyl, a lower alkynyl, an amino, a nitro, an azido, a sulfate, a sulfonate, a sulfonamido, -$(CH_2)_mR_7$, $(CH_2)_m$-OH, -$(CH_2)_m$-O-lower alkyl, -$(CH_2)_m$-O-lower alkenyl, -$(CH_2)_n$-O-$(CH_2)_m$-R_7, --$(CH_2)_m$-SH, -$(CH_2)_m$-S-lower alkyl, -$(CH_2)_m$-S-lower alkenyl, -$(CH_2)_n$-S-$(CH_2)_m$-R_7

[0037] R_7, R_8, and R_9 each independently represents, for each occurrence, hydrogen, hydroxyl, or a substituted or unsubstituted alkyl, alkenyl, aryl, aralkyl, benzyl, cycloalkyl, cycloalkenyl, or heterocycle; and

[0038] wherein each occurrence of m and n is separately and independently an integer ranging from 1 to 9, and each occurrence of z is independently an integer ranging from 1 to 9;

[0039] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

[0040] In certain embodiments, R_3 and R_4 are each -OH.

[0041] In certain embodiments, R_2 and R_3 are each -OH.

[0042] In certain embodiments, z is 2 and R_8 and R_9 are each -H.

[0043] In certain embodiments, z is 2; R_1, R_2, R_5, R_6, R_8 and R_9 are each -H; and R_3 and R_4 are each -OH.

[0044] In certain embodiments, X is O.

[0045] In certain embodiments,

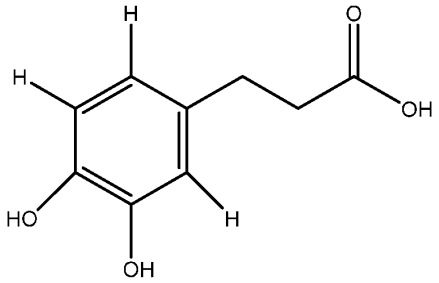
[0046] R_1, R_2, R_3, R_6, R_8, and R_9 are each -H;

[0047] R_4 and R_5 are each -OH;

[0048] X is O; and

[0049] Z is 2.

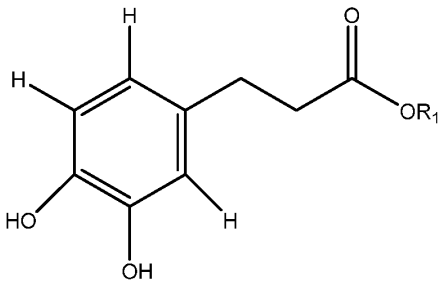
[0050] In certain embodiments, the compound of formula II has the following structure:



Exemplary Compound E05

[0051] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

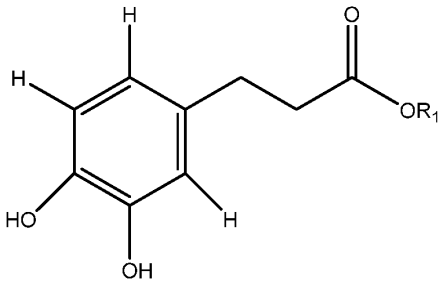
[0052] In certain embodiments, the compound of formula II is a prodrug of the following structure:



Formula IV

[0053] wherein R_1 comprises esters including ethyl esters, morpholinoethanol esters, acetate, dialkylaminoacetates, formates, phosphates, sulfates and benzoate derivatives; carbamates including N,N-dimethylaminocarbonyl of hydroxy functional groups, and N-acyl derivatives.

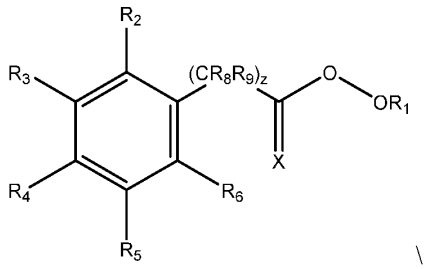
[0054] The invention also encompasses prodrugs of formula:



Formula IV

[0055] wherein R_1 comprises esters including alkyl esters, morpholinoethanol esters, acetate, dialkylaminoacetates, formates, phosphates, sulfates and benzoate derivatives; carbamates including N,N-dimethylaminocarbonyl of hydroxy functional groups, and N-acyl derivatives.

[0056] In other embodiments, the invention encompasses methods of treating cancer including chronic myeloid leukemia (CML), acute myeloid leukemia (AML), acute lymphocytic leukemia (ALL), breast cancer, and colon cancer comprising administering a following structure:



Formula II

[0057] wherein X is O or S;

[0058] R_1 is a hydrogen, or a substituted or unsubstituted substituent including but not limited to lower alkyl, a lower alkenyl, a lower alkynyl, $-(CH_2)_mR_7$, $(CH_2)_m$-OH, $-(CH_2)_m$-O-lower alkyl, $-(CH_2)_m$-O-lower alkenyl, $-(CH_2)_n$-O-$(CH_2)_m$-R_7, $-(CH_2)_m$-SH, $-(CH_2)_m$-S-lower alkyl, $-(CH_2)_m$-S-lower alkenyl, $-(CH_2)_n$-S-$(CH_2)_m$-R_7

[0059] each of R_2-R_6 is independently a hydrogen, a hydroxyl, a halogen, a lower alkyl, a lower alkenyl, a lower alkynyl, an amino, a nitro, an azido, a sulfate, a sulfonate, a sulfonamido, $-(CH_2)_mR_7$, $(CH_2)_m$-OH, $-(CH_2)_m$-O-lower alkyl, $-(CH_2)_m$-O-lower alkenyl, $-(CH_2)_n$-O-$(CH_2)_m$-R_7, $--(CH_2)_m$-SH, $-(CH_2)_m$-S-lower alkyl, $-(CH_2)_m$-S-lower alkenyl, $-(CH_2)_n$-S-$(CH_2)_m$-R_7

[0060] R_7, R_8, and R_9 each independently represents, for each occurrence, hydrogen, hydroxyl, or a substituted or unsubstituted alkyl, alkenyl, aryl, aralkyl, benzyl, cycloalkyl, cycloalkenyl,

or heterocycle; and

[0061] wherein each occurrence of m and n is separately and independently an integer ranging from 1 to 9, and each occurrence of z is independently an integer ranging from 1 to 9;

[0062] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

[0063] In certain embodiments, R_3 and R_4 are each -OH.

[0064] In certain embodiments, R_2 and R_3 are each -OH.

[0065] In certain embodiments, z is 2 and R_8 and R_9 are each -H.

[0066] In certain embodiments, z is 2; R_1, R_2, R_5, R_6, R_8 and R_9 are each -H; and R_3 and R_4 are each -OH.

[0067] In certain embodiments, X is O.

[0068] In certain embodiments,

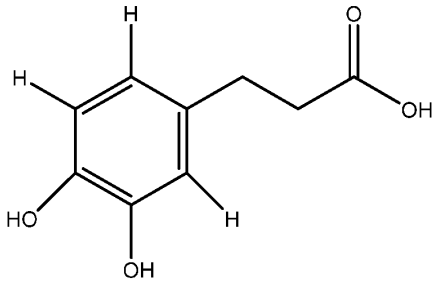
[0069] R_1, R_2, R_3, R_6, R_8, and R_9 are each -H;

[0070] R_4 and R_5 are each -OH;

[0071] X is O; and

[0072] Z is 2.

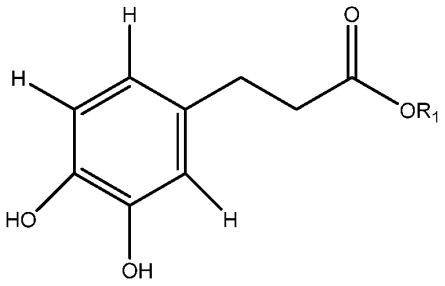
[0073] In certain embodiments, the compound of formula II has the following structure:



Exemplary Compound E05

[0074] or a pharmaceutically acceptable salt, hydrate, solvate, clathrate, enantiomer, diastereomer, racemate or mixture of steroisomers thereof.

[0075] In certain embodiments, the compound of formula II is a prodrug of the following structure:



Formula IV

[0076] wherein R1 comprises esters including ethyl esters, morpholinoethanol esters, acetate, dialkylaminoacetates, formates, phosphates, sulfates and benzoate derivatives; carbamates including N,N-dimethylaminocarbonyl of hydroxy functional groups, and N-acyl derivatives.

[0077] In certain embodiments, the invention further encompasses administration of one or more additional therapeutic agents comprising anticancer agents or chemotherapeutic agents.

[0078] One skilled in the art may refer to general reference texts for detailed descriptions of known techniques discussed herein or equivalent techniques. These texts include Ausubel et al., Current Protocols in Molecular Biology, John Wiley and Sons, Inc. (2005); Sambrook et al., Molecular Cloning, A Laboratory Manual (3.sup.th edition), Cold Spring Harbor Press, Cold Spring Harbor, N.Y. (2000); Coligan et al., Current Protocols in Immunology, John Wiley & Sons, N.Y.; Enna et al., Current Protocols in Pharmacology, John Wiley & Sons, N.Y.; Fingl et al., The Pharmacological Basis of Therapeutics (1975), Remington's Pharmaceutical Sciences, Mack Publishing Co., Easton, Pa., 18.sup.th edition (1990). These texts can, of course, also be referred to in making or using an aspect of the invention.

[0079] The present invention provides effective therapeutic methods for modulating tumor growth or metastasis wherein a combination of agents is employed. The methods of the present invention provide advantages such as greater overall efficacy, for example, in achieving synergy or avoiding antagonism, and allow, where desired, a reduction in the amount of one or more of the individual agents employed with a concomitant reduction in side effects. Further, where the tumor to be treated is not optimally responsive to a given anticancer agent, use of the present combination therapy methods can nonetheless provide effective treatment.

[0080] As used herein, the phrase "effective amount" of a compound or pharmaceutical composition refers to an amount sufficient to modulate tumor growth or metastasis in an animal, especially a human, including without limitation decreasing tumor growth or size or preventing formation of tumor growth in an animal lacking any tumor formation prior to

administration, i.e., prophylactic administration.

[0081] As used herein, the terms "tumor", "tumor growth" or "tumor tissue" can be used interchangeably, and refer to an abnormal growth of tissue resulting from uncontrolled progressive multiplication of cells and serving no physiological function. A solid tumor can be malignant, e.g. tending to metastasize and being life threatening, or benign. Examples of solid tumors that can be treated or prevented according to a method of the present invention include sarcomas and carcinomas such as, but not limited to: fibrosarcoma, myxosarcoma, liposarcoma, chondrosarcoma, osteogenic sarcoma, chordoma, angiosarcoma, endotheliosarcoma, lymphangiosarcoma, lymphangioendotheliosarcoma, synovioma, mesothelioma, Ewing's tumor, leiomyosarcoma, rhabdomyosarcoma, colon carcinoma, colorectal cancer, gastric cancer, pancreatic cancer, breast cancer, ovarian cancer, prostate cancer, squamous cell carcinoma, basal cell carcinoma, adenocarcinoma, sweat gland carcinoma, sebaceous gland carcinoma, papillary carcinoma, papillary adenocarcinomas, cystadenocarcinoma, medullary carcinoma, bronchogenic carcinoma, renal cell carcinoma, hepatoma, liver metastases, bile duct carcinoma, choriocarcinoma, seminoma, embryonal carcinoma, thyroid carcinoma such as anaplastic thyroid cancer, Wilms' tumor, cervical cancer, testicular tumor, lung carcinoma such as small cell lung carcinoma and non-small cell lung carcinoma, bladder carcinoma, epithelial carcinoma, glioma, astrocytoma, medulloblastoma, craniopharyngioma, ependymoma, pinealoma, hemangioblastoma, acoustic neuroma, oligodendroglioma, meningioma, melanoma, neuroblastoma, and retinoblastoma.

[0082] Moreover, tumors comprising dysproliferative changes (such as metaplasias and dysplasias) can be treated or prevented with a pharmaceutical composition or method of the present invention in epithelial tissues such as those in the cervix, colon, esophagus, and lung. Thus, the present invention provides for treatment of conditions known or suspected of preceding progression to neoplasia or cancer, in particular, where non-neoplastic cell growth consisting of hyperplasia, metaplasia, or most particularly, dysplasia has occurred (for review of such abnormal growth conditions, see Robbins and Angell, 1976, Basic Pathology, 2d Ed., W.B. Saunders Co., Philadelphia, pp. 68 to 79). Hyperplasia is a form of controlled cell proliferation involving an increase in cell number in a tissue or organ, without significant alteration in structure or function. For example, endometrial hyperplasia often precedes endometrial cancer. Metaplasia is a form of controlled cell growth in which one type of adult or fully differentiated cell substitutes for another type of adult cell. Metaplasia can occur in epithelial or connective tissue cells. Atypical metaplasia involves a somewhat disorderly

metaplastic epithelium. Dysplasia is frequently a forerunner of cancer, and is found mainly in the epithelia; it is the most disorderly form of non-neoplastic cell growth, involving a loss in individual cell uniformity and in the architectural orientation of cells. Dysplastic cells often have abnormally large, deeply stained nuclei, and exhibit pleomorphism. Dysplasia characteristically occurs where there exists chronic irritation or inflammation, and is often found in the cervix, respiratory passages, oral cavity, and gall bladder. For a review of such disorders, see Fishman et al., 1985, Medicine, 2d Ed., J. B. Lippincott Co., Philadelphia.

[0083] The present invention encompasses treating and/or preventing various types of leukemia. Leukemia is a cancer of the early blood-forming cells. Most often, leukemia is a cancer of the white blood cells, but some leukemias start in other blood cell types. There are several types of leukemia, which are divided based mainly on whether the leukemia is acute (fast growing) or chronic (slower growing), and whether it starts in myeloid cells or lymphoid cells. Different types of leukemia have different treatment options and outlooks. Acute lymphocytic (or lymphoblastic) leukemia is sometimes called ALL. It starts in the bone marrow where blood cells are made. It is more common in children than in adults. Acute myeloid leukemia is also called acute myelocytic leukemia, acute myelogenous leukemia, acute granulocytic leukemia, acute non-lymphocytic leukemia, or sometimes just AML. It is most common in older people. Chronic lymphocytic leukemia (CLL) is a type of cancer that starts in white blood cells (called lymphocytes) in the bone marrow. CLL mainly affects older adults and accounts for about one-third of all leukemias. Chronic myeloid leukemia (CML) is also known as chronic myelogenous leukemia. It's a type of cancer that starts in the blood-forming cells of the bone marrow and invades the blood. About 15% of leukemias in adults are CML. Chronic myelomonocytic leukemia (CMML) is a type of cancer that starts in blood-forming cells of the bone marrow and invades the blood. It affects mainly older adults.

[0084] The present methods can, for example, be carried out using a single pharmaceutical composition comprising both an Aur-A inhibitor and Src inhibitor (dasatinib) (when administration is to be simultaneous) or using two or more pharmaceutical compositions separately comprising the Src inhibitor and dasatinib (when administration is to be simultaneous or sequential). The phrase "pharmaceutically acceptable" refers to molecular entities and compositions that are physiologically tolerable and preferably do not produce an allergic or similar untoward reaction, such as gastric upset, dizziness and the like, when administered to a human.

[0085] A pharmaceutical composition of the present invention can be administered by any

suitable route, for example, by injection, by oral, pulmonary, nasal or other forms of administration. In general, pharmaceutical compositions contemplated to be within the scope of the invention, comprise, inter alia, pharmaceutically acceptable diluents, preservatives, solubilizers, emulsifiers, adjuvants and/or carriers. Such compositions can include diluents of various buffer content (e.g., Tris-HCl, acetate, phosphate), pH and ionic strength; additives such as detergents and solubilizing agents (e.g., Tween 80, Polysorbate 80), anti-oxidants (e.g., ascorbic acid, sodium metabisulfite), preservatives (e.g., Thimersol, benzyl alcohol) and bulking substances (e.g., lactose, mannitol); incorporation of the material into particulate preparations of polymeric compounds such as polylactic acid, polyglycolic acid, etc., or into liposomes. Such compositions may influence the physical state, stability, rate of in vivo release, and rate of in vivo clearance of components of a pharmaceutical composition of the present invention. See, e.g., Remington's Pharmaceutical Sciences, 18th Ed. (1990, Mack Publishing Co., Easton, Pa. 18042) pages 1435-1712 which are herein incorporated by reference. A pharmaceutical composition of the present invention can be prepared, for example, in liquid form, or can be in dried powder, such as lyophilized form. Particular methods of administering such compositions are described infra.

[0086] The present invention is directed towards methods for modulating tumor growth and metastasis comprising, the administration of an Aur-A inhibitor such as those listed above and at least one Src inhibitor, preferably dasatinib. The agents of the invention can be administered separately (e.g, formulated and administered separately), or in combination as a pharmaceutical composition of the present invention. Administration can be achieved by any suitable route, such as parenterally, transmucosally, e.g., orally, nasally, or rectally, or transdermally. Preferably, administration is parenteral, e.g., via intravenous injection. Alternative means of administration also include, but are not limited to, intra-arteriole, intramuscular, intradermal, subcutaneous, intraperitoneal, intraventricular, and intracranial administration, or by injection into the tumor(s) being treated or into tissues surrounding the tumor(s).

[0087] In yet another embodiment, a pharmaceutical composition of the present invention can be delivered in a controlled release system, such as using an intravenous infusion, an implantable osmotic pump, a transdermal patch, liposomes, or other modes of administration. In a particular embodiment, a pump may be used [see Langer, supra; Sefton, CRC Crit. Ref. Biomed. Eng. 14:201 (1987); Buchwald et al., Surgery 88:507 (1980); Saudek et al., N. Engl. J. Med. 321:574 (1989)]. In another embodiment, polymeric materials can be used [see Medical Applications of Controlled Release, Langer and Wise (eds.), CRC Press: Boca Raton, Fla.

(1974); Controlled Drug Bioavailability, Drug Product Design and Performance, Smolen and Ball (eds.), Wiley: New York (1984); Ranger and Peppas, J. Macromol. Sci. Rev. Macromol. Chem. 23:61 (1983); see also Levy et al., Science 228:190 (1985); During et al., Ann. Neurol. 25:351 (1989); Howard et al., J. Neurosurg. 71:105 (1989)]. In yet another embodiment, a controlled release system can be placed in proximity of the target tissues of the animal, thus requiring only a fraction of the systemic dose [see, e.g., Goodson, in Medical Applications of Controlled Release, supra, vol. 2, pp. 115-138 (1984)]. In particular, a controlled release device can be introduced into an animal in proximity of the site of inappropriate immune activation or a tumor. Other controlled release systems are discussed in the review by Langer [Science 249:1527-1533 (1990)].

BRIEF DESCRIPTION OF THE FIGURES

[0088] FIGURE 1 illustrates and exemplary embodiment of a chromatography profile of the fractions indentified in the initial Capture Step.

[0089] FIGURE 2 illustrates and exemplary embodiment of a chromatography profile of the fractions indentified in the Intermediate Step.

[0090] FIGURE 3 illustrates and exemplary embodiment of a chromatography profile of the fractions indentified in the Polishing Step.

[0091] FIGURE 4 illustrates and exemplary embodiment of a chromatography profile of the fractions indentified in the Desalting Step.

[0092] FIGURE 5 illustrates and exemplary embodiment of a Proton (^{1}H) NMR profile of the purified compound E05.

[0093] FIGURE 6 illustrates and exemplary embodiment of a Carbon (^{13}C) NMR profile of the purified compound E05.

[0094] FIGURE 7 illustrates and exemplary embodiment of a distortionless enhancement by polarization transfer (DEPT) profile of the purified compound E05.

[0095] FIGURE 8 illustrates and exemplary embodiment of a infrared (IR) profile of the purified compound E05.

[0096] FIGURE 9a illustrates and exemplary embodiment of a LC-MS spectra of purified compound E05.

[0097] FIGURE 9b illustrates and exemplary embodiment of a MS spectra of purified compound E05.

[0098] FIGURE 10 illustrates and exemplary embodiment of a ORTEP view of compound

E05 (3-(3,4-dihydroxy phenyl) propanoic acid) showing the atom-numbering scheme, Displacement ellipsoids are drawn at the 50% probability level and H atoms are shown as small spheres with arbitrary radii.

[0099] FIGURE 11 illustrates and exemplary embodiment of a Sahadevi water-soluble fraction induced anti-proliferative activity in different cell lines

[0100] FIGURE 12 illustrates and exemplary embodiment of anti-proliferative activity of pure compound (E05) in colon cancer cell line, HCT116. A dose dependent response can be seen upon treatment with the pure compound

[0101] FIGURE 13 illustrates and exemplary embodiment of anti-proliferative activity of pure compound (E05) in breast cancer cells, BT-474. A dose dependent response can be seen upon treatment with the pure compound

[0102] FIGURE 14 illustrates and exemplary embodiment of a comparison of IC_{50} values between synthetic E05 and the SRC kinase inhibitor Bosutinib in F-36E cells

[0103] FIGURE 15 illustrates and exemplary embodiment of IC_{50} value of synthetic E05 in triple-negative breast cancer cell line, MDA-MB-468

[0104] FIGURE 16 illustrates and exemplary embodiment of a comparison of anti-proliferative activity between synthetic E05 and lapatinib in triple-negative breast cancer cell line, MDA-MB-468

[0105] FIGURE 17 illustrates and exemplary embodiment of efficacy of exemplary compound E05 in the murine xenograft model of triple-negative breast cancer.

[0106] FIGURE 18 illustrates and exemplary embodiment of mean plasma concentration-time profile of small molecule following oral gavage administration of small molecule dose formulation in male Sprague Dawley rats (Dose: 10 mg/kg; G3).

DETAILED DESCRIPTION OF THE INVENTION

[0107] As used herein, the term "cycloalkyl" refers to a optionally substituted saturated or unsaturated nonaromatic hydrocarbon mono- or multi-ring (e.g., fused, bridged, or Spiro rings) system having 3 to 30 carbon atoms (e.g., C_3-C_{10}). Examples of cycloalkyl include, but are not limited to, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl, cyclopentenyl, cyclohexenyl, cycloheptenyl, and adamantyl. The term "heterocycloalkyl" refers to a saturated or unsaturated nonaromatic 3-8 membered monocyclic, 7-12 membered bicyclic (fused, bridged, or spiro rings), or 11-14 membered tricyclic ring system (fused, bridged, or spiro rings) having one or more heteroatoms (such as O, N, S, or Se), unless

specified otherwise. Examples of heterocycloalkyl groups include, but are not limited to, piperidinyl, piperazinyl, pyrrolidinyl, dioxanyl, tetrahydrofuranyl, isoindolinyl, indolinyl, imidazolidinyl, pyrazolidinyl, oxazolidinyl, isoxazolidinyl, triazolidinyl, tetrahyrofuranyl, oxiranyl, azetidinyl, oxetanyl, thietanyl, 1,2,3,6-tetrahydropyridinyl, tetrahydropyranyl, dihydropyranyl, pyranyl, morpholinyl, 1,4-diazepanyl, 1,4-oxazepanyl, 2-oxa-5-azabicyclo[2.2.1]heptanyl, 2,5-diazabicyclo[2.2.1]heptanyl, 2-oxa-6-azaspiro[3.3]heptanyl, 2,6-diazaspiro[3.3]heptanyl, 1,4-dioxa-8-azaspiro[4.5]decanyl and the like.

[0108] The term "optionally substituted alkyl" refers to unsubstituted alkyl or alkyl having designated substituents replacing one or more hydrogen atoms on one or more carbons of the hydrocarbon backbone. Such substituents can include, for example, alkyl, alkenyl, alkynyl, halogen, hydroxyl, alkylcarbonyloxy, arylcarbonyloxy, alkoxycarbonyloxy, aryloxycarbonyloxy, carboxylate, alkylcarbonyl, arylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkyl aminocarbonyl, dialkylaminocarbonyl, alkylthiocarbonyl, alkoxyl, phosphate, phosphonato, phosphinato, amino (including alkylamino, dialkylamino, arylamino, diarylamino and alkylarylamino), acylamino (including alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), amidino, imino, sulfhydryl, alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonato, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, azido, heterocyclyl, alkylaryl, or an aromatic or heteroaromatic moiety.

[0109] An "arylalkyl" or an "aralkyl" moiety is an alkyl substituted with an aryl (e.g., phenylmethyl (benzyl)). An "alkylaryl" moiety is an aryl substituted with an alkyl (e.g., methylphenyl).

[0110] "Alkenyl" includes unsaturated aliphatic groups analogous in length and possible substitution to the alkyls described above, but that contain at least one double bond. For example, the term "alkenyl" includes straight chain alkenyl groups (e.g., ethenyl, propenyl, butenyl, pentenyl, hexenyl, heptenyl, octenyl, nonenyl, decenyl), and branched alkenyl groups. In certain embodiments, a straight chain or branched alkenyl group has six or fewer carbon atoms in its backbone (e.g., C_1-C_6 for straight chain, C_3-C_6 for branched chain). The term "C_2-C_6" includes alkenyl groups containing two to six carbon atoms. The term "C_3-C_6" includes alkenyl groups containing three to six carbon atoms.

[0111] The term "optionally substituted alkenyl" refers to unsubstituted alkenyl or alkenyl having designated substituents replacing one or more hydrogen atoms on one or more hydrocarbon backbone carbon atoms. Such substituents can include, for example, alkyl, alkenyl, alkynyl, halogen, hydroxyl, alkylcarbonyloxy, aryl carbonyloxy, alkoxycarbonyloxy,

aryloxycarbonyloxy, carboxylate, alkylcarbonyl, arylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkyl aminocarbonyl, dialkylaminocarbonyl, alkylthiocarbonyl, alkoxyl, phosphate, phosphonato, phosphinato, amino (including alkylamino, dialkylamino, arylamino, diarylamino and alkylarylamino), acylamino (including alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), amidino, imino, sulfhydryl, alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonato, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, heterocyclyl, alkylaryl, or an aromatic or heteroaromatic moiety.

[0112] "Alkynyl" includes unsaturated aliphatic groups analogous in length and possible substitution to the alkyls described above, but which contain at least one triple bond. For example, "alkynyl" includes straight chain alkynyl groups (e.g., ethynyl, propynyl, butynyl, pentynyl, hexynyl, heptynyl, octynyl, nonynyl, decynyl), and branched alkynyl groups. In certain embodiments, a straight chain or branched alkynyl group has six or fewer carbon atoms in its backbone (e.g., C_2-C_6 for straight chain, C_3-C_6 for branched chain). The term " C_2-C_6" includes alkynyl groups containing two to six carbon atoms. The term " C_3-C_6" includes alkynyl groups containing three to six carbon atoms. The term "optionally substituted alkynyl" refers to unsubstituted alkynyl or alkynyl having designated substituents replacing one or more hydrogen atoms on one or more hydrocarbon backbone carbon atoms. Such substituents can include, for example, alkyl, alkenyl, alkynyl, halogen, hydroxyl, alkylcarbonyloxy, arylcarbonyloxy, alkoxycarbonyloxy, aryloxycarbonyloxy, carboxylate, alkylcarbonyl, arylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkyl aminocarbonyl, dialkylaminocarbonyl, alkylthiocarbonyl, alkoxyl, phosphate, phosphonato, phosphinato, amino (including alkylamino, dialkylamino, arylamino, diarylamino and alkylarylamino), acylamino (including alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), amidino, imino, sulfhydryl, alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonato, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, azido, heterocyclyl, alkylaryl, or an aromatic or heteroaromatic moiety.

[0113] Other optionally substituted moieties (such as optionally substituted cycloalkyl, heterocycloalkyl, aryl, or heteroaryl) include both the unsubstituted moieties and the moieties having one or more of the designated substituents. For example, substituted heterocycloalkyl includes those substituted with one or more alkyl groups, such as 2,2,6,6-tetramethyl-piperidinyl and 2,2,6,6-tetramethyl-1,2,3,6-tetrahydropyridinyl.

[0114] "Aryl" includes groups with aromaticity, including "conjugated," or multicyclic systems with at least one aromatic ring and do not contain any heteroatom in the ring structure.

Examples include phenyl, benzyl, 1,2,3,4-tetrahydronaphthalenyl, etc.

[0115] "Heteroaryl" groups are aryl groups, as defined above, except having from one to four heteroatoms in the ring structure, and may also be referred to as "aryl heterocycles" or "heteroaromatics." As used herein, the term "heteroaryl" is intended to include a stable 5-, 6-, or 7-membered monocyclic or 7-, 8-, 9-, 10-, 11- or 12-membered bicyclic aromatic heterocyclic ring which consists of carbon atoms and one or more heteroatoms, e.g., 1 or 1-2 or 1-3 or 1-4 or 1-5 or 1-6 heteroatoms, or e.g., 1, 2, 3, 4, 5, or 6 heteroatoms, independently selected from the group consisting of nitrogen, oxygen and sulfur. The nitrogen atom may be substituted or unsubstituted (i.e., N or NR wherein R is H or other substituents, as defined). The nitrogen and sulfur heteroatoms may optionally be oxidized (i.e., N.fwdarw.O and S(O).sub.p, where p=1 or 2). It is to be noted that total number of S and O atoms in the aromatic heterocycle is not more than 1. Examples of heteroaryl groups include pyrrole, furan, thiophene, thiazole, isothiazole, imidazole, triazole, tetrazole, pyrazole, oxazole, isoxazole, pyridine, pyrazine, pyridazine, pyrimidine, and the like.

[0116] Furthermore, the terms "aryl" and "heteroaryl" include multicyclic aryl and heteroaryl groups, e.g., tricyclic, bicyclic, e.g., naphthalene, benzoxazole, benzodioxazole, benzothiazole, benzoimidazole, benzothiophene, methylenedioxyphenyl, quinoline, isoquinoline, naphthrydine, indole, benzofuran, purine, benzofuran, deazapurine, indolizine.

[0117] In the case of multicyclic aromatic rings, only one of the rings needs to be aromatic (e.g., 2,3-dihydroindole), although all of the rings may be aromatic (e.g., quinoline). The second ring can also be fused or bridged.

[0118] The cycloalkyl, heterocycloalkyl, aryl, or heteroaryl ring can be substituted at one or more ring positions (e.g., the ring-forming carbon or heteroatom such as N) with such substituents as described above, for example, alkyl, alkenyl, alkynyl, halogen, hydroxyl, alkoxy, alkyl carbonyloxy, aryl carbonyloxy, alkoxycarbonyloxy, aryloxycarbonyloxy, carboxylate, alkylcarbonyl, alkylaminocarbonyl, aralkylaminocarbonyl, alkenylaminocarbonyl, alkylcarbonyl, arylcarbonyl, aralkylcarbonyl, alkenylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkylthiocarbonyl, phosphate, phosphonato, phosphinato, amino (including alkylamino, dialkylamino, arylamino, diarylamino and alkylarylamino), acylamino (including alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), amidino, imino, sulfhydryl, alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonato, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, azido, heterocyclyl, alkylaryl, or an aromatic or heteroaromatic moiety. Aryl and heteroaryl groups can also be fused or

bridged with alicyclic or heterocyclic rings, which are not aromatic so as to form a multicyclic system (e.g., tetralin, methylenedioxyphenyl).

[0119] As used herein, "carbocycle" or "carbocyclic ring" is intended to include any stable monocyclic, bicyclic or tricyclic ring having the specified number of carbons, any of which may be saturated, unsaturated, or aromatic. Carbocycle includes cycloalkyl and aryl. For example, a C.sub.3-C.sub.14 carbocycle is intended to include a monocyclic, bicyclic or tricyclic ring having 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13 or 14 carbon atoms. Examples of carbocycles include, but are not limited to, cyclopropyl, cyclobutyl, cyclobutenyl, cyclopentyl, cyclopentenyl, cyclohexyl, cycloheptenyl, cycloheptyl, cycloheptenyl, adamantyl, cyclooctyl, cyclooctenyl, cyclooctadienyl, fluorenyl, phenyl, naphthyl, indanyl, adamantyl and tetrahydronaphthyl. Bridged rings are also included in the definition of carbocycle, including, for example, [3.3.0]bicyclooctane, [4.3.0]bicyclononane, [4.4.0]bicyclodecane and [2.2.2]bicyclooctane. A bridged ring occurs when one or more carbon atoms link two non-adjacent carbon atoms. In one embodiment, bridge rings are one or two carbon atoms. It is noted that a bridge always converts a monocyclic ring into a tricyclic ring. When a ring is bridged, the substituents recited for the ring may also be present on the bridge. Fused (e.g., naphthyl, tetrahydronaphthyl) and Spiro rings are also included.

[0120] As used herein, "heterocycle" or "heterocyclic group" includes any ring structure (saturated, unsaturated, or aromatic) which contains at least one ring heteroatom (e.g., N, O or S). Heterocycle includes heterocycloalkyl and heteroaryl. Examples of heterocycles include, but are not limited to, morpholine, pyrrolidine, tetrahydrothiophene, piperidine, piperazine, oxetane, pyran, tetrahydropyran, azetidine, and tetrahydrofuran. Examples of heterocyclic groups include, but are not limited to, acridinyl, azocinyl, benzimidazolyl, benzofuranyl, benzothiofuranyl, benzothiophenyl, benzoxazolyl, benzoxazolinyl, benzthiazolyl, benztriazolyl, benztetrazolyl, benzisoxazolyl, benzisothiazolyl, benzimidazolinyl, carbazolyl, 4aH-carbazolyl, carbolinyl, chromanyl, chromenyl, cinnolinyl, decahydroquinolinyl, 2H,6H-1,5,2-dithiazinyl, dihydrofuro[2,3-b]tetrahydrofuran, furanyl, furazanyl, imidazolidinyl, imidazolinyl, imidazolyl, 1H-indazolyl, indolenyl, indolinyl, indolizinyl, indolyl, 3H-indolyl, isatinoyl, isobenzofuranyl, isochromanyl, isoindazolyl, isoindolinyl, isoindolyl, isoquinolinyl, isothiazolyl, isoxazolyl, methylenedioxyphenyl, morpholinyl, naphthyridinyl, octahydroisoquinolinyl, oxadiazolyl, 1,2,3-oxadiazolyl, 1,2,4-oxadiazolyl, 1,2,5-oxadiazolyl, 1,3,4-oxadiazolyl, 1,2,4-oxadiazol5(4H)-one, oxazolidinyl, oxazolyl, oxindolyl, pyrimidinyl, phenanthridinyl, phenanthrolinyl, phenazinyl, phenothiazinyl, phenoxathinyl, phenoxazinyl,

phthalazinyl, piperazinyl, piperidinyl, piperidonyl, 4-piperidonyl, piperonyl, pteridinyl, purinyl, pyranyl, pyrazinyl, pyrazolidinyl, pyrazolinyl, pyrazolyl, pyridazinyl, pyridooxazole, pyridoimidazole, pyridothiazole, pyridinyl, pyridyl, pyrimidinyl, pyrrolidinyl, pyrrolinyl, 2H-pyrrolyl, pyrrolyl, quinazolinyl, quinolinyl, 4H-quinolizinyl, quinoxalinyl, quinuclidinyl, tetrahydrofuranyl, tetrahydroisoquinolinyl, tetrahydroquinolinyl, tetrazolyl, 6H-1,2,5-thiadiazinyl, 1,2,3-thiadiazolyl, 1,2,4-thiadiazolyl, 1,2,5-thiadiazolyl, 1,3,4-thiadiazolyl, thianthrenyl, thiazolyl, thienyl, thienothiazolyl, thienooxazolyl, thienoimidazolyl, thiophenyl, triazinyl, 1,2,3-triazolyl, 1,2,4-triazolyl, 1,2,5-triazolyl, 1,3,4-triazolyl and xanthenyl.

[0121] When any variable (e.g., R_2) occurs more than one time in any constituent or formula for a compound, its definition at each occurrence is independent of its definition at every other occurrence. Thus, for example, if a group is shown to be substituted with 0-2 R.sub.1 moieties, then the group may optionally be substituted with up to two R.sub.1 moieties and R.sub.1 at each occurrence is selected independently from the definition of R.sub.1. Also, combinations of substituents and/or variables are permissible, but only if such combinations result in stable compounds.

[0122] The term "hydroxy" or "hydroxyl" includes groups with an --OH.

[0123] As used herein, "halo" or "halogen" refers to fluoro, chloro, bromo and iodo. The term "perhalogenated" generally refers to a moiety wherein all hydrogen atoms are replaced by halogen atoms. The term "haloalkyl" or "haloalkoxyl" refers to an alkyl or alkoxyl substituted with one or more halogen atoms.

[0124] The term "carbonyl" includes compounds and moieties which contain a carbon connected with a double bond to an oxygen atom. Examples of moieties containing a carbonyl include, but are not limited to, aldehydes, ketones, carboxylic acids, amides, esters, anhydrides, etc.

[0125] The term "carboxyl" refers to --COOH or its C_1-C_6 alkyl ester.

[0126] "Acyl" includes moieties that contain the acyl radical (R--C(O)--) or a carbonyl group. "Substituted acyl" includes acyl groups where one or more of the hydrogen atoms are replaced by, for example, alkyl groups, alkynyl groups, halogen, hydroxyl, alkylcarbonyloxy, arylcarbonyloxy, alkoxycarbonyloxy, aryloxycarbonyloxy, carboxylate, alkylcarbonyl, arylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkylaminocarbonyl, dialkylaminocarbonyl, alkylthiocarbonyl, alkoxyl, phosphate, phosphonato, phosphinato, amino (including alkylamino, dialkylamino, arylamino, diarylamino and alkylarylamino), acylamino (including alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), amidino, imino, sulfhydryl,

alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonato, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, azido, heterocyclyl, alkylaryl, or an aromatic or heteroaromatic moiety. The term "alkoxy" or "alkoxyl" includes substituted and unsubstituted alkyl, alkenyl and alkynyl groups covalently linked to an oxygen atom. Examples of alkoxy groups or alkoxyl radicals include, but are not limited to, methoxy, ethoxy, isopropyloxy, propoxy, butoxy and pentoxy groups. Examples of substituted alkoxy groups include halogenated alkoxy groups. The alkoxy groups can be substituted with groups such as alkenyl, alkynyl, halogen, hydroxyl, alkylcarbonyloxy, arylcarbonyloxy, alkoxycarbonyloxy, aryloxycarbonyloxy, carboxylate, alkylcarbonyl, arylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkylaminocarbonyl, dialkylaminocarbonyl, alkylthiocarbonyl, alkoxyl, phosphate, phosphonato, phosphinato, amino (including alkylamino, dialkylamino, arylamino, diarylamino, and alkylarylamino), acylamino (including alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), amidino, imino, sulfhydryl, alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonato, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, azido, heterocyclyl, alkylaryl, or an aromatic or heteroaromatic moieties. Examples of halogen substituted alkoxy groups include, but are not limited to, fluoromethoxy, difluoromethoxy, trifluoromethoxy, chloromethoxy, dichloromethoxy and trichloromethoxy.

[0127] The term "ether" or "alkoxy" includes compounds or moieties which contain an oxygen bonded to two carbon atoms or heteroatoms. For example, the term includes "alkoxyalkyl," which refers to an alkyl, alkenyl, or alkynyl group covalently bonded to an oxygen atom which is covalently bonded to an alkyl group.

[0128] The term "ester" includes compounds or moieties which contain a carbon or a heteroatom bound to an oxygen atom which is bonded to the carbon of a carbonyl group. The term "ester" includes alkoxycarboxy groups such as methoxycarbonyl, ethoxycarbonyl, propoxycarbonyl, butoxycarbonyl, pentoxycarbonyl, etc.

[0129] As used herein, "amine" or "amino" refers to unsubstituted or substituted --NH.sub.2. "Alkylamino" includes groups of compounds wherein nitrogen of --NH.sub.2 is bound to at least one alkyl group. Examples of alkylamino groups include benzylamino, methylamino, ethylamino, phenethylamino, etc. "Dialkylamino" includes groups wherein the nitrogen of --NH.sub.2 is bound to at least two additional alkyl groups. Examples of dialkylamino groups include, but are not limited to, dimethylamino and diethylamino. "Arylamino" and "diarylamino" include groups wherein the nitrogen is bound to at least one or two aryl groups, respectively. "Aminoaryl" and "aminoaryloxy" refer to aryl and aryloxy substituted with

amino. "Alkylarylamino," "alkylaminoaryl" or "arylaminoalkyl" refers to an amino group which is bound to at least one alkyl group and at least one aryl group. "Alkaminoalkyl" refers to an alkyl, alkenyl, or alkynyl group bound to a nitrogen atom which is also bound to an alkyl group. "Acylamino" includes groups wherein nitrogen is bound to an acyl group. Examples of acylamino include, but are not limited to, alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido groups.

[0130] The term "amide" or "aminocarboxy" includes compounds or moieties that contain a nitrogen atom that is bound to the carbon of a carbonyl or a thiocarbonyl group. The term includes "alkaminocarboxy" groups that include alkyl, alkenyl or alkynyl groups bound to an amino group which is bound to the carbon of a carbonyl or thiocarbonyl group. It also includes "arylaminocarboxy" groups that include aryl or heteroaryl moieties bound to an amino group that is bound to the carbon of a carbonyl or thiocarbonyl group. The terms "alkylaminocarboxy", "alkenylaminocarboxy", "alkynylaminocarboxy" and "arylaminocarboxy" include moieties wherein alkyl, alkenyl, alkynyl and aryl moieties, respectively, are bound to a nitrogen atom which is in turn bound to the carbon of a carbonyl group. Amides can be substituted with substituents such as straight chain alkyl, branched alkyl, cycloalkyl, aryl, heteroaryl or heterocycle. Substituents on amide groups may be further substituted.

[0131] Compounds of the present invention that contain nitrogens can be converted to N-oxides by treatment with an oxidizing agent (e.g., 3-chloroperoxybenzoic acid (mCPBA) and/or hydrogen peroxides) to afford other compounds of the present invention. Thus, all shown and claimed nitrogen-containing compounds are considered, when allowed by valency and structure, to include both the compound as shown and its N-oxide derivative (which can be designated as N.fwdarw.O or N.sup.+--O.sup.-). Furthermore, in other instances, the nitrogens in the compounds of the present invention can be converted to N-hydroxy or N-alkoxy compounds. For example, N-hydroxy compounds can be prepared by oxidation of the parent amine by an oxidizing agent such as m-CPBA. All shown and claimed nitrogen-containing compounds are also considered, when allowed by valency and structure, to cover both the compound as shown and its N-hydroxy (*i.e.,* N--OH) and N-alkoxy (i.e., N--OR, wherein R is substituted or unsubstituted C_1-C_6 alkyl, C_1-C_6 alkenyl, C_1-C_6 alkynyl, 3-14-membered carbocycle or 3-14-membered heterocycle) derivatives.

[0132] In the present specification, the structural formula of the compounds of Formula (I) or (II) represent a certain isomer for convenience in some cases, but the present invention includes

all isomers, such as geometrical isomers, optical isomers based on an asymmetrical carbon, stereoisomers, tautomers, and the like. In addition, a crystal polymorphism may be present for the compounds represented by the formula. It is noted that any crystal form, crystal form mixture, or anhydride or hydrate thereof is included in the scope of the present invention. Furthermore, so-called metabolite which is produced by degradation of the present compound in vivo is included in the scope of the present invention.

[0133] It is to be understood that the structures and other compounds discussed in this invention include all atropic isomers thereof. "Atropic isomers" are a type of stereoisomer in which the atoms of two isomers are arranged differently in space. Atropic isomers owe their existence to a restricted rotation caused by hindrance of rotation of large groups about a central bond. Such atropic isomers typically exist as a mixture, however as a result of recent advances in chromatography techniques, it has been possible to separate mixtures of two atropic isomers in select cases.

[0134] "Tautomer" is one of two or more structural isomers that exist in equilibrium and is readily converted from one isomeric form to another. This conversion results in the formal migration of a hydrogen atom accompanied by a switch of adjacent conjugated double bonds. Tautomers exist as a mixture of a tautomeric set in solution. In solutions where tautomerization is possible, a chemical equilibrium of the tautomers will be reached. The exact ratio of the tautomers depends on several factors, including temperature, solvent and pH. The concept of tautomers that are interconvertable by tautomerizations is called tautomerism.

[0135] The term "crystal polymorphs", "polymorphs" or "crystal forms" means crystal structures in which a compound (or a salt or solvate thereof) can crystallize in different crystal packing arrangements, all of which have the same elemental composition. Different crystal forms usually have different X-ray diffraction patterns, infrared spectral, melting points, density hardness, crystal shape, optical and electrical properties, stability and solubility. Recrystallization solvent, rate of crystallization, storage temperature, and other factors may cause one crystal form to dominate. Crystal polymorphs of the compounds can be prepared by crystallization under different conditions.

[0136] The term “Compounds of the Invention” includes compounds of Formula (I), (II), (III), or (IV) disclosed herein include the compounds themselves, as well as their salts, their esters, their solvates, and their prodrugs, if applicable.

[0137] A salt, for example, can be formed between an anion and a positively charged group (e.g., amino) on an aryl- or heteroaryl-substituted benzene compound. Suitable anions include

chloride, bromide, iodide, sulfate, bisulfate, sulfamate, nitrate, phosphate, citrate, methanesulfonate, trifluoroacetate, glutamate, glucuronate, glutarate, malate, maleate, succinate, fumarate, tartrate, tosylate, salicylate, lactate, naphthalenesulfonate, and acetate (e.g., trifluoroacetate). The term "pharmaceutically acceptable anion" refers to an anion suitable for forming a pharmaceutically acceptable salt. Likewise, a salt can also be formed between a cation and a negatively charged group (e.g., carboxylate) on an aryl- or heteroaryl-substituted benzene compound. Suitable cations include sodium ion, potassium ion, magnesium ion, calcium ion, and an ammonium cation such as tetramethylammonium ion. The aryl- or heteroaryl-substituted benzene compounds also include those salts containing quaternary nitrogen atoms. In the salt form, it is understood that the ratio of the compound to the cation or anion of the salt can be 1:1, or any ration other than 1:1, e.g., 3:1, 2:1, 1:2, or 1:3.

[0138] Examples of prodrugs include esters and other pharmaceutically acceptable derivatives, which, upon administration to a subject, are capable of providing active aryl- or heteroaryl-substituted benzene compounds.

[0139] Additionally, the compounds of the present invention, for example, the salts of the compounds, can exist in either hydrated or unhydrated (the anhydrous) form or as solvates with other solvent molecules. Nonlimiting examples of hydrates include monohydrates, dihydrates, etc. Nonlimiting examples of solvates include ethanol solvates, acetone solvates, etc.

[0140] "Solvate" means solvent addition forms that contain either stoichiometric or non stoichiometric amounts of solvent. Some compounds have a tendency to trap a fixed molar ratio of solvent molecules in the crystalline solid state, thus forming a solvate. If the solvent is water the solvate formed is a hydrate; and if the solvent is alcohol, the solvate formed is an alcoholate. Hydrates are formed by the combination of one or more molecules of water with one molecule of the substance in which the water retains its molecular state as H_2O.

[0141] As used herein, the term "analog" refers to a chemical compound that is structurally similar to another but differs slightly in composition (as in the replacement of one atom by an atom of a different element or in the presence of a particular functional group, or the replacement of one functional group by another functional group). Thus, an analog is a compound that is similar or comparable in function and appearance, but not in structure or origin to the reference compound.

[0142] As defined herein, the term "derivative" refers to compounds that have a common core structure, and are substituted with various groups as described herein. For example, all of the compounds represented by Formula (I) and (II) are aryl- or heteroaryl-substituted benzene

compounds, and have Formula (I) and (II) as a common core.

[0143] The term "bioisostere" refers to a compound resulting from the exchange of an atom or of a group of atoms with another, broadly similar, atom or group of atoms. The objective of a bioisosteric replacement is to create a new compound with similar biological properties to the parent compound. The bioisosteric replacement may be physicochemically or topologically based. Examples of carboxylic acid bioisosteres include, but are not limited to, acyl sulfonimides, tetrazoles, sulfonates and phosphonates. See, e.g., Patani and LaVoie, Chem. Rev. 96, 3147-3176, 1996.

[0144] The present invention is intended to include all isotopes of atoms occurring in the present compounds. Isotopes include those atoms having the same atomic number but different mass numbers. By way of general example and without limitation, isotopes of hydrogen include tritium and deuterium, and isotopes of carbon include C-13 and C-14.

[0145] The inhibition is a measurable inhibition compared to a suitable control. In one embodiment, inhibition is at least 10 percent inhibition compared to a suitable control. That is, the rate of enzymatic activity or the amount of product with the inhibitor is less than or equal to 90 percent of the corresponding rate or amount made without the inhibitor. In various other embodiments, inhibition is at least 20, 25, 30, 40, 50, 60, 70, 75, 80, 90, or 95 percent inhibition compared to a suitable control. In one embodiment, inhibition is at least 99 percent inhibition compared to a suitable control. That is, the rate of enzymatic activity or the amount of product with the inhibitor is less than or equal to 1 percent of the corresponding rate or amount made without the inhibitor.

[0146] The term “Composition of the Invention” comprises a compound of Formula (I) or (II), (III), or (IV), or a pharmaceutically acceptable salt thereof as well as their esters, their solvates, and their prodrugs, if applicable.

[0147] The present invention provides for the administration of a compound of Formula (I), (II), (III), or (IV) or a pharmaceutically acceptable salt thereof, and one or more therapeutic agents or a pharmaceutically acceptable salt thereof, as a co-formulation or separate formulations, wherein the administration of formulations is simultaneous, sequential, or in alternation. In certain embodiments, the other therapeutic agents can be an agent that is recognized in the art as being useful to treat the disease or condition being treated by the composition of the present invention. In other embodiment, the other therapeutic agent can be an agent that is not recognized in the art as being useful to treat the disease or condition being treated by the composition of the present invention. In one aspect, the other therapeutic agents

can be an agent that imparts a beneficial attribute to the composition of the present invention (e.g., an agent that affects the viscosity of the composition). The beneficial attribute to the composition of the present invention includes, but is not limited to, pharmacokinetic or pharmacodynamic co-action resulting from the combination of a compound of Formula (I), (II), (III), or (IV) and one or more other therapeutic agents. For example, the one or more other therapeutic agents can be anticancer agents or chemotherapeutic agents. For example, the one or more other therapeutic agents can be glucocorticoids. For example, the one or more other therapeutic agents can be selected from prednisone, prednisolone, cyclophosphamide, vincristine, doxorubicin, mafosfamide, cisplatin, AraC, everolimus, decitabine, dexamethasone, or functional analogs, derivatives, produgs, and metabolites thereof. In another aspect, the other therapeutic agent can be Prednisone or its active metabolite, Prednisolone.

COMBINATIONS OF THE INVENTION

[0148] A composition of the present invention comprises a compound of Formula (I), (II), (III), or (IV), or a pharmaceutically acceptable salt thereof, and one or more additional therapeutic agents, or a pharmaceutically acceptable salt thereof.

[0149] The present invention provides for the administration of a compound of Formula (I), (II), (III), or (IV) or a pharmaceutically acceptable salt thereof, and one or more therapeutic agents or a pharmaceutically acceptable salt thereof, as a co-formulation or separate formulations, wherein the administration of formulations is simultaneous, sequential, or in alternation. In certain embodiments, the other therapeutic agents can be an agent that is recognized in the art as being useful to treat the disease or condition being treated by the composition of the present invention. In other embodiment, the other therapeutic agent can be an agent that is not recognized in the art as being useful to treat the disease or condition being treated by the composition of the present invention. In one aspect, the other therapeutic agents can be an agent that imparts a beneficial attribute to the composition of the present invention (e.g., an agent that affects the viscosity of the composition). The beneficial attribute to the composition of the present invention includes, but is not limited to, pharmacokinetic or pharmacodynamic co-action resulting from the combination of a compound of Formula (I), (II), (III), or (IV) and one or more other therapeutic agents. For example, the one or more other therapeutic agents can be anticancer agents or chemotherapeutic agents. For example, the one or more other therapeutic agents can be glucocorticoids. For example, the one or more other therapeutic agents can be selected from prednisone, prednisolone, cyclophosphamide,

vincristine, doxorubicin, mafosfamide, cisplatin, AraC, everolimus, decitabine, dexamethasone, or functional analogs, derivatives, produgs, and metabolites thereof. In another aspect, the other therapeutic agent can be Prednisone or its active metabolite, Prednisolone.

[0150] The therapeutic agents set forth below are for illustrative purposes and not intended to be limiting. The present invention includes at least one other therapeutic agent selected from the lists below. The present invention can include more than one other therapeutic agent, e.g., two, three, four, or five other therapeutic agents such that the composition of the present invention can perform its intended function.

[0151] In one embodiment, the other therapeutic agent is an anticancer agent.

[0152] In one embodiment, the anticancer agent is selected from the group consisting of chemotherapeutics (such as 2CdA, 5-FU, 6-Mercaptopurine, 6-TG, Abraxane™, Accutane®, Actinomycin-D, Adriamycin®, Alimta®, all-trans retinoic acid, amethopterin, Ara-C, Azacitadine, BCNU, Blenoxane®, Camptosar®, CeeNU®, Clofarabine, Clolar™, Cytoxan®, daunorubicin hydrochloride, DaunoXome®, Dacogen®, DIC, Doxil®, Ellence®, Eloxatin®, Emcyt®, etoposide phosphate, Fludara®, FUDR®, Gemzar®, Gleevec®, hexamethylmelamine, Hycamtin®, Hydrea®, Idamycin®, Ifex®, ixabepilone, Ixempra®, L-asparaginase, Leukeran®, liposomal Ara-C, L-PAM, Lysodren, Matulane®, mithracin, Mitomycin-C, Myleran®, Navelbine®, Neutrexin®, nilotinib, Nipent®, Nitrogen Mustard, Novantrone®, Oncaspar®, Panretin®, Paraplatin®, Platinol®, prolifeprospan 20 with carmustine implant, Sandostatin®, Targretin®, Tasigna®, Taxotere®, Temodar®, TESPA, Trisenox®, Valstar®, Velban®, Vidaza™, vincristine sulfate, VM 26, Xeloda® and Zanosar®); biologics (such as Alpha Interferon, Bacillus Calmette-Guerin, Bexxar®, Campath®, Ergamisol®, Erlotinib, Herceptin®, Interleukin-2, Iressa®, lenalidomide, Mylotarg®, Ontak®, Pegasys®, Revlimid®, Rituxan®, Tarceva™, Thalomid®, Tykerb®, Velcade® and Zevalin™); corticosteroids, (such as dexamethasone sodium phosphate, DeltaSone® and Delta-Cortef®); hormonal therapies (such as Arimidex®, Aromasin®, Casodex®, Cytadren®, Eligard®, Eulexin®, Evista®, Faslodex®, Femara®, Halotestin®, Megace®, Nilandron®, Nolvadex®, Plenaxis™ and Zoladex®); and radiopharmaceuticals (such as Iodotope®, Metastron®, Phosphocol® and Samarium SM-153).

[0153] In another embodiment, the other therapeutic agent is a chemotherapeutic agent (also referred to as an anti-neoplastic agent or anti-proliferative agent), selected from the group including an alkylating agent; an antibiotic; an anti-metabolite; a detoxifying agent; an interferon; a polyclonal or monoclonal antibody; an EGFR inhibitor; a HER2 inhibitor; a

histone deacetylase inhibitor; a hormone; a mitotic inhibitor; an MTOR inhibitor; a multi-kinase inhibitor; a serine/threonine kinase inhibitor; a tyrosine kinase inhibitors; a VEGF/VEGFR inhibitor; a taxane or taxane derivative, an aromatase inhibitor, an anthracycline, a microtubule targeting drug, a topoisomerase poison drug, an inhibitor of a molecular target or enzyme (e.g., a kinase or a protein methyltransferase), a cytidine analogue drug or any chemotherapeutic, anti-neoplastic or anti-proliferative agent listed in www.cancer.org/docroot/cdg/cdg_0.asp.

[0154] Exemplary alkylating agents include, but are not limited to, cyclophosphamide (Cytoxan; Neosar); chlorambucil (Leukeran); melphalan (Alkeran); carmustine (BiCNU); busulfan (Busulfex); lomustine (CeeNU); dacarbazine (DTIC-Dome); oxaliplatin (Eloxatin); carmustine (Gliadel); ifosfamide (Ifex); mechlorethamine (Mustargen); busulfan (Myleran); carboplatin (Paraplatin); cisplatin (CDDP; Platinol); temozolomide (Temodar); .sub.thiotepa (Thioplex); .sub.bendamustine (Treanda); or .sub.streptozocin (Zanosar).

[0155] Exemplary anti-metabolites include, but are not limited to, fluorouracil (Adrucil); capecitabine (Xeloda); hydroxyurea (Hydrea); mercaptopurine (Purinethol); pemetrexed (Alimta); fludarabine (Fludara); nelarabine (Arranon); cladribine (Cladribine Novaplus); clofarabine (Clolar); cytarabine (Cytosar-U); decitabine (Dacogen); cytarabine liposomal (DepoCyt); hydroxyurea (Droxia); pralatrexate (Folotyn); floxuridine (FUDR); gemcitabine (Gemzar; cladribine (Leustatin); fludarabine (Oforta); methotrexate (MTX; Rheumatrex); methotrexate (Trexall); thioguanine (Tabloid); TS-1 or cytarabine (Tarabine PFS).

[0156] Exemplary detoxifying agents include, but are not limited to, amifostine (Ethyol) or .sub.mesna (Mesnex).

[0157] Exemplary interferons include, but are not limited to, interferon alfa-2b (Intron A) or interferon alfa-2a (Roferon-A).

[0158] Exemplary polyclonal or monoclonal antibodies include, but are not limited to, trastuzumab (Herceptin); ofatumumab (Arzerra); bevacizumab (Avastin); rituximab (Rituxan); cetuximab (Erbitux); panitumumab (Vectibix); tositumomab/iodine131 tositumomab (Bexxar); alemtuzumab (Campath); ibritumomab (Zevalin; In-111; Y-90 Zevalin); .sub.gemtuzumab (Mylotarg); .sub.eculizumab (Soliris) ordenosumab.

[0159] Exemplary EGFR inhibitors include, but are not limited to, gefitinib (Iressa); lapatinib (Tykerb); cetuximab (Erbitux); erlotinib (Tarceva); panitumumab (Vectibix); PKI-166; canertinib (CI-1033); matuzumab (Emd7200) or EKB-569.

[0160] Exemplary HER2 inhibitors include, but are not limited to, trastuzumab (Herceptin);

lapatinib (Tykerb) or AC-480.

[0161] Histone Deacetylase Inhibitors include, but are not limited to, vorinostat (Zolinza).

[0162] Exemplary hormones include, but are not limited to, tamoxifen (Soltamox; Nolvadex); raloxifene (Evista); megestrol (Megace); leuprolide (Lupron; Lupron Depot; Eligard; Viadur); fulvestrant (Faslodex); letrozole (Femara); triptorelin (Trelstar LA; Trelstar Depot); exemestane (Aromasin); goserelin (Zoladex); bicalutamide (Casodex); anastrozole (Arimidex); fluoxymesterone (Androxy; Halotestin); medroxyprogesterone (Provera; Depo-Provera); estramustine (Emcyt); flutamide (Eulexin); toremifene (Fareston); .sub.degarelix (Firmagon); .sub.nilutamide (Nilandron), .sub.abarelix (Plenaxis); or testolactone (Teslac).

[0163] Exemplary mitotic inhibitors include, but are not limited to, paclitaxel (Taxol; Onxol; Abraxane); docetaxel (Taxotere); vincristine (Oncovin; Vincasar PFS); vinblastine (Velban); etoposide (Toposar; Etopophos; VePesid); teniposide (Vumon); ixabepuone (Ixempra); nocodazole; epothilone; .sub.vinorelbine (Navelbine); camptothecin (CPT); .sub.irinotecan (Camptosar); topotecan (Hycamtin); amsacrine or lamellarin D (LAM-D).

[0164] Exemplary MTOR inhibitors include, but are not limited to, everolimus (Afinitor) or temsirolimus (Torisel); rapamune, ridaforolimus; or AP23573.

[0165] Exemplary VEGFNEGFR inhibitors include, but are not limited to, bev.sub.acizumab (Avastin); .sub.sorafenib (Nexavar); .sub.sunitinib (Sutent); ranibizumab; pegaptanib; or vandetinib.

[0166] Exemplary microtubule targeting drugs include, but are not limited to, paclitaxel, docetaxel, vincristine, vinblastin, nocodazole, epothilones and navelbine.

[0167] Exemplary topoisomerase poison drugs include, but are not limited to, teniposide, etoposide, adriamycin, camptothecin, daunorubicin, dactinomycin, mitoxantrone, amsacrine, epirubicin and idarubicin.

[0168] Exemplary taxanes or taxane derivatives include, but are not limited to, paclitaxel and docetaxol.

[0169] Exemplary general chemotherapeutic, anti-neoplastic, anti-proliferative agents include, but are not limited to, altretamine (Hexalen); isotretinoin (Accutane; Amnesteem; Claravis; Sotret); tretinoin (Vesanoid); azacitidine (Vidaza); bortezomib (Velcade) asparaginase (Elspar); levamisole (Ergamisol); mitotane (Lysodren); procarbazine (Matulane); pegaspargase (Oncaspar); denileukin diftitox (Ontak), porfimer (Photofrin); .sub.aldesleukin (Proleukin); lenalidomide (Revlimid); bexarotene (Targretin); thalidomide (Thalomid); temsirolimus (Torisel); arsenic trioxide (Trisenox); verteporfin (Visudyne); mimosine

(Leucenol); (1M tegafur-0.4 M 5-chloro-2,4-dihydroxypyrimidine-1 M potassium oxonate), or lovastatin.

[0170] In another aspect, the other therapeutic agent is a chemotherapeutic agent or a cytokine such as G-CSF (granulocyte colony stimulating factor).

[0171] In yet another aspect, the other therapeutic agents can be standard chemotherapy combinations such as, but not restricted to, CMF (cyclophosphamide, methotrexate and 5-fluorouracil), CAF (cyclophosphamide, adriamycin and 5-fluorouracil), AC (adriamycin and cyclophosphamide), FEC (5-fluorouracil, epirubicin, and cyclophosphamide), ACT or ATC (adriamycin, cyclophosphamide, and paclitaxel), rituximab, Xeloda (capecitabine), Cisplatin (CDDP), Carboplatin, TS-1 (tegafur, gimestat and otastat potassium at a molar ratio of 1:0.4:1), Camptothecin-11 (CPT-11, Irinotecan or Camptosar™), CHOP (cyclophosphamide, hydroxydaunorubicin, oncovin, and prednisone or prednisolone), R-CHOP (rituximab, cyclophosphamide, hydroxydaunorubicin, oncovin, prednisone or prednisolone), or CMFP (cyclophosphamide, methotrexate, 5-fluorouracil and prednisone).

[0172] In another aspect, the other therapeutic agents can be an inhibitor of an enzyme, such as a receptor or non-receptor kinase. Receptor and non-receptor kinases are, for example, tyrosine kinases or serine/threonine kinases. Kinase inhibitors described herein are small molecules, polynucleic acids, polypeptides, or antibodies.

[0173] Exemplary kinase inhibitors include, but are not limited to, Bevacizumab (targets VEGF), BIBW 2992 (targets EGFR and Erb2), Cetuximab/Erbitux (targets Erb1), Imatinib/Gleevic (targets Bcr-Abl), Trastuzumab (targets Erb2), Gefitinib/Iressa (targets EGFR), Ranibizumab (targets VEGF), Pegaptanib (targets VEGF), Erlotinib/Tarceva (targets Erb1), Nilotinib (targets Bcr-Abl), Lapatinib (targets Erb1 and Erb2/Her2), GW-572016/lapatinib ditosylate (targets HER2/Erb2), Panitumumab/Vectibix (targets EGFR), Vandetinib (targets RET/VEGFR), E7080 (multiple targets including RET and VEGFR), Herceptin (targets HER2/Erb2), PKI-166 (targets EGFR), Canertinib/CI-1033 (targets EGFR), Sunitinib/SU-11464/Sutent (targets EGFR and FLT3), Matuzumab/Emd7200 (targets EGFR), EKB-569 (targets EGFR), Zd6474 (targets EGFR and VEGFR), PKC-412 (targets VEGR and FLT3), Vatalanib/Ptk787/ZK222584 (targets VEGR), CEP-701 (targets FLT3), SU5614 (targets FLT3), MLN518 (targets FLT3), XL999 (targets FLT3), VX-322 (targets FLT3), Azd0530 (targets SRC), BMS-354825 (targets SRC), SKI-606 (targets SRC), CP-690 (targets JAK), AG-490 (targets JAK), WHI-P154 (targets JAK), WHI-P131 (targets JAK), sorafenib/Nexavar (targets RAF kinase, VEGFR-1, VEGFR-2, VEGFR-3, PDGFR-.beta., KIT,

FLT-3, and RET), Dasatinib/Sprycel (BCR/ABL and Src), AC-220 (targets Flt3), AC-480 (targets all HER proteins, "panHER"), Motesanib diphosphate (targets VEGF1-3, PDGFR, and c-kit), Denosumab (targets RANKL, inhibits SRC), AMG888 (targets HER3), and AP24534 (multiple targets including Flt3).

[0174] Exemplary serine/threonine kinase inhibitors include, but are not limited to, Rapamune (targets mTOR/FRAP1), Deforolimus (targets mTOR), Certican/Everolimus (targets mTOR/FRAP1), AP23573 (targets mTORJFRAP1), Eril/Fasudil hydrochloride (targets RHO), Flavopiridol (targets CDK), Seliciclib/CYC202/Roscovitrine (targets CDK), SNS-032/BMS-387032 (targets CDK), Ruboxistaurin (targets PKC), Pkc412 (targets PKC), Bryostatin (targets PKC), KAI-9803 (targets PKC), SF1126 (targets PI3K), VX-680 (targets Aurora kinase), Azd1152 (targets Aurora kinase), Arry-142886/AZD-6244 (targets MAP/MEK), SCIO-469 (targets MAP/MEK), GW681323 (targets MAP/MEK), CC-401 (targets JNK), CEP-1347 (targets JNK), and PD 332991 (targets CDK).

[0175] Exemplary tyrosine kinase inhibitors include, but are not limited to, erlotinib (Tarceva); gefitinib (Iressa); imatinib (Gleevec); sorafenib (Nexavar); sunitinib (Sutent); trastuzumab (Herceptin); bevacizumab (Avastin); rituximab (Rituxan); lapatinib (Tykerb); cetuximab (Erbitux); panitumumab (Vectibix); everolimus (Afinitor); alemtuzumab (Campath), .sub.gemtuzumab (Mylotarg); .sub.temsimlimus (Torisel), .sub.pazopanib (Votrient); .sub.dasatinib (Sprycel); .sub.niotinib (Tasigna), vatalanib (Ptk787; ZK222584); CEP-701; SU5614; MLN518, XL999; VX-322; Azd0530; BMS-354825; SKI-606 CP-690; AG-490; WHI-P154; WHI-P131; AC-220; or AMG888.

[0176] The present invention provides methods for combination therapy in which a composition comprising a compound of Formula (I) or (II)or a pharmaceutically acceptable salt thereof, and one or more other therapeutic agents are administered to a subject in need for treatment of a disease or cancer. The combination therapy can also be administered to cancer cells to inhibit proliferation or induce cell death. In one aspect, a compound of Formula (I) or (II) or a pharmaceutically acceptable salt thereof is administered prior to administration of the composition of the present invention comprising a compound of Formula (I) or a pharmaceutically acceptable salt thereof, and one or more other therapeutic agents. In one aspect, a compound of Formula (I) or (II) or a pharmaceutically acceptable salt thereof is administered prior to administration of one or more therapeutic agents, such that the other therapeutic agents are administered either in a single composition or in two or more compositions, e.g. administered simultaneously, sequentially, or in alternation.

[0177] In one embodiment, a composition of the present invention includes a compound of Formula (I) or (II) or a pharmaceutically acceptable salt thereof, and one or more anticancer agents, e.g., CHOP (cyclophosphamide, hydroxydaunorubicin, oncovin, and prednisone or prednisolone) or R-CHOP (rituximab, cyclophosphamide, hydroxydaunorubicin, oncovin, prednisone or prednisolone). In one embodiment, a composition of the present invention includes a compound of Formula (I) or (II) or a pharmaceutically acceptable salt thereof, and prednisone or prednisolone. Methods of the present invention include the combination therapy of administering a compound of Formula (I) or (II) or a pharmaceutically acceptable salt thereof, and anticancer agents, wherein the anticancer agents are CHOP, R-CHOP, prednisone, or prednisolone.

[0178] In certain embodiments, "combination therapy" is intended to embrace administration of these therapeutic agents in a sequential manner, wherein each therapeutic agent is administered at a different time, as well as administration of these therapeutic agents, or at least two of the therapeutic agents concurrently, or in a substantially simultaneous manner. Simultaneous administration can be accomplished, for example, by administering to the subject a single capsule having a fixed ratio of each therapeutic agent or in multiple, single capsules for each of the therapeutic agents. Sequential or substantially simultaneous administration of each therapeutic agent can be effected by any appropriate route including, but not limited to, oral routes, intravenous routes, intramuscular routes, and direct absorption through mucous membrane tissues. The therapeutic agents can be administered by the same route or by different routes. For example, a first therapeutic agent of the combination selected may be administered by intravenous injection while the other therapeutic agents of the combination may be administered orally. Alternatively, for example, all therapeutic agents may be administered orally or all therapeutic agents may be administered by intravenous injection. Therapeutic agents may also be administered in alternation.

[0179] In certain aspects of the invention, the combination therapies featured in the present invention can result in a synergistic effect in the treatment of a disease or cancer. A "synergistic effect" is defined as where the efficacy of a combination of therapeutic agents is greater than the sum of the effects of any of the agents given alone. A synergistic effect may also be an effect that cannot be achieved by administration of any of the compounds or other therapeutic agents as single agents. The synergistic effect may include, but is not limited to, an effect of treating cancer by reducing tumor size, inhibiting tumor growth, or increasing survival of the subject. The synergistic effect may also include reducing cancer cell viability, inducing cancer

cell death, and inhibiting or delaying cancer cell growth.

[0180] In certain aspects of the invention "combination therapy" also embraces the administration of the therapeutic agents as described above in further combination with other biologically active ingredients and non-drug therapies (e.g., surgery or radiation treatment). Where the combination therapy further comprises a non-drug treatment, the non-drug treatment may be conducted at any suitable time so long as a beneficial effect from the co-action of the combination of the therapeutic agents and non-drug treatment is achieved. For example, in appropriate cases, the beneficial effect is still achieved when the non-drug treatment is temporally removed from the administration of the therapeutic agents, perhaps by days or even weeks.

[0181] In another aspect, a composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, analog or derivative thereof, may be administered in combination with radiation therapy. Radiation therapy can also be administered in combination with a composition of the present invention and another chemotherapeutic agent described herein as part of a multiple agent therapy.

COMPOSITIONS OF THE INVENTION

[0182] The present invention also provides pharmaceutical compositions comprising a compound of Formula (I), (II), (III), or (IV) or pharmaceutically acceptable salts thereof mixed with pharmaceutically suitable carriers or excipient(s) at doses to treat or prevent a disease or condition as described herein. In one aspect, the present invention also provides pharmaceutical compositions comprising any compound of Formula (I) or (II) or pharmaceutically acceptable salts thereof, mixed with pharmaceutically suitable carriers or excipient (s) at doses to treat or prevent a disease or condition as described herein. In another aspect, the present invention also provides pharmaceutical compositions comprising compounds of Formula (I), (II), (III), or (IV) or pharmaceutically acceptable salts thereof, mixed with pharmaceutically suitable carriers or excipient(s) at doses to treat or prevent a disease or condition as described herein. The pharmaceutical compositions of the present invention can also be administered in combination with other therapeutic agents or therapeutic modalities simultaneously, sequentially, or in alternation.

[0183] Mixtures of compositions of the present invention can also be administered to the patient as a simple mixture or in suitable formulated pharmaceutical compositions. For example, one aspect of the invention relates to a pharmaceutical composition comprising a

therapeutically effective dose of an SRC inhibitor of Formula (I), (II), (III), or (IV) or a pharmaceutically acceptable salt, hydrate, enantiomer or stereoisomer thereof; one or more other therapeutic agent, and a pharmaceutically acceptable diluent or carrier.

[0184] A "pharmaceutical composition" is a formulation containing the compounds of Formula Formula (I), (II), (III), or (IV) in a form suitable for administration to a subject. In one embodiment, the pharmaceutical composition is in bulk or in unit dosage form. The unit dosage form is any of a variety of forms, including, for example, a capsule, an IV bag, a tablet, a single pump on an aerosol inhaler or a vial. The quantity of active ingredient (e.g., a formulation of the disclosed compound or salt, hydrate, solvate or isomer thereof) in a unit dose of composition is an effective amount and is varied according to the particular treatment involved. One skilled in the art will appreciate that it is sometimes necessary to make routine variations to the dosage depending on the age and condition of the patient. The dosage will also depend on the route of administration. A variety of routes are contemplated, including oral, pulmonary, rectal, parenteral, transdermal, subcutaneous, intravenous, intramuscular, intraperitoneal, inhalational, buccal, sublingual, intrapleural, intrathecal, intranasal, and the like. Dosage forms for the topical or transdermal administration of a compound of this invention include powders, sprays, ointments, pastes, creams, lotions, gels, solutions, patches and inhalants. In one embodiment, the active compound is mixed under sterile conditions with a pharmaceutically acceptable carrier, and with any preservatives, buffers, or propellants that are required.

[0185] As used herein, the phrase "pharmaceutically acceptable" refers to those compounds, anions, cations, materials, compositions, carriers, and/or dosage forms which are, within the scope of sound medical judgment, suitable for use in contact with the tissues of human beings and animals without excessive toxicity, irritation, allergic response, or other problem or complication, commensurate with a reasonable benefit/risk ratio.

[0186] "Pharmaceutically acceptable excipient" means an excipient that is useful in preparing a pharmaceutical composition that is generally safe, non-toxic and neither biologically nor otherwise undesirable, and includes excipient that is acceptable for veterinary use as well as human pharmaceutical use. A "pharmaceutically acceptable excipient" as used in the specification and claims includes both one and more than one such excipient.

[0187] A pharmaceutical composition of the invention is formulated to be compatible with its intended route of administration. Examples of routes of administration include parenteral, e.g., intravenous, intradermal, subcutaneous, oral (e.g., inhalation), transdermal (topical), and transmucosal administration. Solutions or suspensions used for parenteral, intradermal, or

subcutaneous application can include the following components: a sterile diluent such as water for injection, saline solution, fixed oils, polyethylene glycols, glycerine, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl parabens; antioxidants such as ascorbic acid or sodium bisulfite; chelating agents such as ethylenediaminetetraacetic acid; buffers such as acetates, citrates or phosphates, and agents for the adjustment of tonicity such as sodium chloride or dextrose. The pH can be adjusted with acids or bases, such as hydrochloric acid or sodium hydroxide. The parenteral preparation can be enclosed in ampoules, disposable syringes or multiple dose vials made of glass or plastic.

[0188] A composition of the invention can be administered to a subject in many of the well-known methods currently used for chemotherapeutic treatment. For example, for treatment of cancers, a compound of the invention may be injected directly into tumors, injected into the blood stream or body cavities or taken orally or applied through the skin with patches. The dose chosen should be sufficient to constitute effective treatment but not so high as to cause unacceptable side effects. The state of the disease condition (e.g., cancer, precancer, and the like) and the health of the patient should preferably be closely monitored during and for a reasonable period after treatment.

[0189] The term "therapeutically effective amount", as used herein, refers to an amount of a pharmaceutical agent to treat, ameliorate, or prevent an identified disease or condition, or to exhibit a detectable therapeutic or inhibitory effect. The effect can be detected by any assay method known in the art. The precise effective amount for a subject will depend upon the subject's body weight, size, and health; the nature and extent of the condition; and the therapeutic or combination of therapeutics selected for administration. Therapeutically effective amounts for a given situation can be determined by routine experimentation that is within the skill and judgment of the clinician. In a preferred aspect, the disease or condition to be treated is cancer. In another aspect, the disease or condition to be treated is a cell proliferative disorder.

[0190] In certain embodiments the therapeutically effective amount of each pharmaceutical agent used in combination will be lower when used in combination in comparison to monotherapy with each agent alone. Such lower therapeutically effective amount could afford for lower toxicity of the therapeutic regimen.

[0191] For any compound, the therapeutically effective amount can be estimated initially either in cell culture assays, e.g., of neoplastic cells, or in animal models, usually rats, mice, rabbits, dogs, or pigs. The animal model may also be used to determine the appropriate concentration

range and route of administration. Such information can then be used to determine useful doses and routes for administration in humans. Therapeutic/prophylactic efficacy and toxicity may be determined by standard pharmaceutical procedures in cell cultures or experimental animals, e.g., ED_{50} (the dose therapeutically effective in 50% of the population) and LD_{50} (the dose lethal to 50% of the population). The dose ratio between toxic and therapeutic effects is the therapeutic index, and it can be expressed as the ratio, LD_{50}/ED_{50}. Pharmaceutical compositions that exhibit large therapeutic indices are preferred. The dosage may vary within this range depending upon the dosage form employed, sensitivity of the patient, and the route of administration.

[0192] Dosage and administration are adjusted to provide sufficient levels of the active agent(s) or to maintain the desired effect. Factors which may be taken into account include the severity of the disease state, general health of the subject, age, weight, and gender of the subject, diet, time and frequency of administration, drug combination(s), reaction sensitivities, and tolerance/response to therapy. Long-acting pharmaceutical compositions may be administered every 3 to 4 days, every week, or once every two weeks depending on half-life and clearance rate of the particular formulation.

[0193] The pharmaceutical compositions containing active compounds of the present invention may be manufactured in a manner that is generally known, e.g., by means of conventional mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping, or lyophilizing processes. Pharmaceutical compositions may be formulated in a conventional manner using one or more pharmaceutically acceptable carriers comprising excipients and/or auxiliaries that facilitate processing of the active compounds into preparations that can be used pharmaceutically. Of course, the appropriate formulation is dependent upon the route of administration chosen.

[0194] Pharmaceutical compositions suitable for injectable use include sterile aqueous solutions (where water soluble) or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersion. For intravenous administration, suitable carriers include physiological saline, bacteriostatic water, Cremophor EL™ (BASF, Parsippany, N.J.) or phosphate buffered saline (PBS). In all cases, the composition must be sterile and should be fluid to the extent that easy syringeability exists. It must be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol (for example, glycerol, propylene

glycol, and liquid polyethylene glycol, and the like), and suitable mixtures thereof. The proper fluidity can be maintained, for example, by the use of a coating such as lecithin, by the maintenance of the required particle size in the case of dispersion and by the use of surfactants. Prevention of the action of microorganisms can be achieved by various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, ascorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, polyalcohols such as manitol and sorbitol, and sodium chloride in the composition. Prolonged absorption of the injectable compositions can be brought about by including in the composition an agent which delays absorption, for example, aluminum monostearate and gelatin.

[0195] Sterile injectable solutions can be prepared by incorporating the active compound in the required amount in an appropriate solvent with one or a combination of ingredients enumerated above, as required, followed by filtered sterilization. Generally, dispersions are prepared by incorporating the active compound into a sterile vehicle that contains a basic dispersion medium and the required other ingredients from those enumerated above. In the case of sterile powders for the preparation of sterile injectable solutions, methods of preparation are vacuum drying and freeze-drying that yields a powder of the active ingredient plus any additional desired ingredient from a previously sterile-filtered solution thereof.

[0196] Oral compositions generally include an inert diluent or an edible pharmaceutically acceptable carrier. They can be enclosed in gelatin capsules or compressed into tablets. For the purpose of oral therapeutic administration, the active compound can be incorporated with excipients and used in the form of tablets, troches, or capsules. Oral compositions can also be prepared using a fluid carrier for use as a mouthwash, wherein the compound in the fluid carrier is applied orally and swished and expectorated or swallowed. Pharmaceutically compatible binding agents, and/or adjuvant materials can be included as part of the composition. The tablets, pills, capsules, troches and the like can contain any of the following ingredients, or compounds of a similar nature: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a disintegrating agent such as alginic acid, Primogel, or corn starch; a lubricant such as magnesium stearate or Sterotes; a glidant such as colloidal silicon dioxide; a sweetening agent such as sucrose or saccharin; or a flavoring agent such as peppermint, methyl salicylate, or orange flavoring.

[0197] For administration by inhalation, the compounds are delivered in the form of an aerosol spray from pressured container or dispenser, which contains a suitable propellant, e.g., a gas such as carbon dioxide, or a nebulizer.

[0198] Systemic administration can also be by transmucosal or transdermal means. For transmucosal or transdermal administration, penetrants appropriate to the barrier to be permeated are used in the formulation. Such penetrants are generally known in the art, and include, for example, for transmucosal administration, detergents, bile salts, and fusidic acid derivatives. Transmucosal administration can be accomplished through the use of nasal sprays or suppositories. For transdermal administration, the active compounds are formulated into ointments, salves, gels, or creams as generally known in the art.

[0199] The active compounds can be prepared with pharmaceutically acceptable carriers that will protect the compound against rapid elimination from the body, such as a controlled release formulation, including implants and microencapsulated delivery systems. Biodegradable, biocompatible polymers can be used, such as ethylene vinyl acetate, polyanhydrides, polyglycolic acid, collagen, polyorthoesters, and polylactic acid. Methods for preparation of such formulations will be apparent to those skilled in the art. The materials can also be obtained commercially from Alza Corporation and Nova Pharmaceuticals, Inc. Liposomal suspensions (including liposomes targeted to infected cells with monoclonal antibodies to viral antigens) can also be used as pharmaceutically acceptable carriers. These can be prepared according to methods known to those skilled in the art, for example, as described in U.S. Pat. No. 4,522,811.

[0200] It is especially advantageous to formulate oral or parenteral compositions in dosage unit form for ease of administration and uniformity of dosage. Dosage unit form as used herein refers to physically discrete units suited as unitary dosages for the subject to be treated; each unit containing a predetermined quantity of active compound calculated to produce the desired therapeutic effect in association with the required pharmaceutical carrier. The specification for the dosage unit forms of the invention are dictated by and directly dependent on the unique characteristics of the active compound and the particular therapeutic effect to be achieved.

[0201] In therapeutic applications, the dosages of the SRC inhibitor compounds of Formula (I), (II), (III), or (IV) described herein, other therapeutic agents described herein, compositions comprising a compound of Formula (I), (II), (III), or (IV) and optionally one or more other therapeutic agents, or the pharmaceutical compositions used in accordance with the invention vary depending on the agent, the age, weight, and clinical condition of the recipient patient, and the experience and judgment of the clinician or practitioner administering the therapy, among other factors affecting the selected dosage. Generally, the dose should be sufficient to result in slowing, and preferably regressing, the growth of the tumors and also preferably causing complete regression of the cancer. Dosages can range from about 0.01 mg/kg per day

to about 5000 mg/kg per day. In preferred aspects, dosages can range from about 1 mg/kg per day to about 1000 mg/kg per day. In an aspect, the dose will be in the range of about 0.1 mg/day to about 50 g/day; about 0.1 mg/day to about 25 g/day; about 0.1 mg/day to about 10 g/day; about 0.1 mg to about 3 g/day; or about 0.1 mg to about 1 g/day, in single, divided, or continuous doses (which dose may be adjusted for the patient's weight in kg, body surface area in m^2, and age in years). An effective amount of a pharmaceutical agent is that which provides an objectively identifiable improvement as noted by the clinician or other qualified observer. For example, regression of a tumor in a patient may be measured with reference to the diameter of a tumor. Decrease in the diameter of a tumor indicates regression. Regression is also indicated by failure of tumors to reoccur after treatment has stopped. As used herein, the term "dosage effective manner" refers to amount of an active compound to produce the desired biological effect in a subject or cell.

[0202] The pharmaceutical compositions can be included in a container, pack, or dispenser together with instructions for administration.

[0203] The composition of the present invention is capable of further forming salts. The composition of the present invention is capable of forming more than one salt per molecule, e.g., mono-, di-, tri-. All of these forms are also contemplated within the scope of the claimed invention.

[0204] As used herein, "pharmaceutically acceptable salts" refer to derivatives of the compounds of the present invention wherein the parent compound is modified by making acid or base salts thereof. Examples of pharmaceutically acceptable salts include, but are not limited to, mineral or organic acid salts of basic residues such as amines, alkali or organic salts of acidic residues such as carboxylic acids, and the like. The pharmaceutically acceptable salts include the conventional non-toxic salts or the quaternary ammonium salts of the parent compound formed, for example, from non-toxic inorganic or organic acids. For example, such conventional non-toxic salts include, but are not limited to, those derived from inorganic and organic acids selected from 2-acetoxybenzoic, 2-hydroxyethane sulfonic, acetic, ascorbic, benzene sulfonic, benzoic, bicarbonic, carbonic, citric, edetic, ethane disulfonic, 1,2-ethane sulfonic, fumaric, glucoheptonic, gluconic, glutamic, glycolic, glycollyarsanilic, hexylresorcinic, hydrabamic, hydrobromic, hydrochloric, hydroiodic, hydroxymaleic, hydroxynaphthoic, isethionic, lactic, lactobionic, lauryl sulfonic, maleic, malic, mandelic, methane sulfonic, napsylic, nitric, oxalic, pamoic, pantothenic, phenylacetic, phosphoric, polygalacturonic, propionic, salicyclic, stearic, subacetic, succinic, sulfamic, sulfanilic,

sulfuric, tannic, tartaric, toluene sulfonic, and the commonly occurring amine acids, e.g., glycine, alanine, phenylalanine, arginine, etc.

[0205] Other examples of pharmaceutically acceptable salts include hexanoic acid, cyclopentane propionic acid, pyruvic acid, malonic acid, 3-(4-hydroxybenzoyl)benzoic acid, cinnamic acid, 4-chlorobenzenesulfonic acid, 2-naphthalenesulfonic acid, 4-toluenesulfonic acid, camphorsulfonic acid, 4-methylbicyclo-[2.2.2]-oct-2-ene-1-carboxylic acid, 3-phenylpropionic acid, trimethylacetic acid, tertiary butylacetic acid, muconic acid, and the like. The present invention also encompasses salts formed when an acidic proton present in the parent compound either is replaced by a metal ion, e.g., an alkali metal ion, an alkaline earth ion, or an aluminum ion; or coordinates with an organic base such as ethanolamine, diethanolamine, triethanolamine, tromethamine, N-methylglucamine, and the like.

[0206] It should be understood that all references to pharmaceutically acceptable salts include solvent addition forms (solvates) or crystal forms (polymorphs) as defined herein, of the same salt.

[0207] The composition of the present invention may also be prepared as esters, for example, pharmaceutically acceptable esters. For example, a carboxylic acid function group in a compound of Formula (I), (II), (III), or (IV) can be converted to its corresponding ester, e.g., a methyl, ethyl or other ester. Also, an alcohol group in a compound can be converted to its corresponding ester, e.g., acetate, propionate or other ester.

[0208] The composition of the present invention can also be prepared as prodrugs, for example, pharmaceutically acceptable prodrugs. The terms "pro-drug" and "prodrug" are used interchangeably herein and refer to any compound which releases an active parent drug in vivo. Since prodrugs are known to enhance numerous desirable qualities of pharmaceuticals (e.g., solubility, bioavailability, manufacturing, etc.), the compounds of the present invention can be delivered in prodrug form. Thus, the present invention is intended to cover prodrugs of the presently claimed compounds, methods of delivering the same and compositions containing the same. "Prodrugs" are intended to include any covalently bonded carriers that release an active parent drug of the present invention in vivo when such prodrug is administered to a subject. Prodrugs in the present invention are prepared by modifying functional groups present in the compound in such a way that the modifications are cleaved, either in routine manipulation or in vivo, to the parent compound. Prodrugs include compounds of the present invention wherein a hydroxy, amino, sulfhydryl, carboxy or carbonyl group is bonded to any group that may be cleaved in vivo to form a free hydroxyl, free amino, free sulfhydryl, free

carboxy or free carbonyl group, respectively.

[0209] Examples of prodrugs include, but are not limited to, esters (e.g., acetate, dialkylaminoacetates, formates, phosphates, sulfates and benzoate derivatives) and carbamates (e.g., N,N-dimethylaminocarbonyl) of hydroxy functional groups, esters (e.g., ethyl esters, morpholinoethanol esters) of carboxyl functional groups, N-acyl derivatives (e.g., N-acetyl)N-Mannich bases, Schiff bases and enaminones of amino functional groups, oximes, acetals, ketals and enol esters of ketone and aldehyde functional groups in Compounds Of The Invention, and the like, See Bundegaard, H., Design of Prodrugs, p1-92, Elesevier, New York-Oxford (1985).

[0210] The composition, or pharmaceutically acceptable salts, esters or prodrugs thereof, are administered orally, nasally, transdermally, pulmonary, inhalationally, buccally, sublingually, intraperintoneally, subcutaneously, intramuscularly, intravenously, rectally, intrapleurally, intrathecally and parenterally. In one embodiment, the compound is administered orally. One skilled in the art will recognize the advantages of certain routes of administration.

[0211] The dosage regimen utilizing the compounds is selected in accordance with a variety of factors including type, species, age, weight, sex and medical condition of the patient; the severity of the condition to be treated; the route of administration; the renal and hepatic function of the patient; and the particular compound or salt thereof employed. An ordinarily skilled physician or veterinarian can readily determine and prescribe the effective amount of the drug required to prevent, counter, or arrest the progress of the condition.

[0212] Techniques for formulation and administration of the disclosed Compounds Of The Invention can be found in Remington: the Science and Practice of Pharmacy, 19.sup.th edition, Mack Publishing Co., Easton, Pa. (1995). In an embodiment, the compounds described herein, and the pharmaceutically acceptable salts thereof, are used in pharmaceutical preparations in combination with a pharmaceutically acceptable carrier or diluent. Suitable pharmaceutically acceptable carriers include inert solid fillers or diluents and sterile aqueous or organic solutions. The compounds will be present in such pharmaceutical compositions in amounts sufficient to provide the desired dosage amount in the range described herein.

[0213] All percentages and ratios used herein, unless otherwise indicated, are by weight. Other features and advantages of the present invention are apparent from the different examples. The provided examples illustrate different components and methodology useful in practicing the present invention. The examples do not limit the claimed invention. Based on the present disclosure the skilled artisan can identify and employ other components and methodology

useful for practicing the present invention.

[0214] The present invention provides compositions and methods for treating conditions and diseases the course of which can be influenced by modulating the methylation status of histones or other proteins, wherein said methylation status is mediated at least in part by the activity of SRC. Modulation of the methylation status of histones can in turn influence the level of expression of target genes activated by methylation, and/or target genes suppressed by methylation. The method includes administering to a subject in need of such treatment, a therapeutically effective amount of a composition of the present invention or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, to a subject in need of such treatment.

[0215] Based at least on the fact that abnormal histone methylation has been found to be associated with certain cancers and precancerous conditions, a method for treating cancer or a precancerous condition with a mutant SRC in a subject comprises administering to the subject in need thereof a therapeutically effective amount of a compound that inhibits methylation. In one embodiment a method for treating cancer or a precancerous condition in a subject comprises administering to the subject in need thereof a therapeutically effective amount of a compound that inhibits conversion of unmethylated H3-K27 to monomethylated H3-K27 (H3-K27me1). In one embodiment a method for treating cancer or a precancerous condition in a subject comprises administering to the subject in need thereof a therapeutically effective amount of a compound that inhibits conversion of monomethylated H3-K27 (H3-K27me1) to dimethylated H3-K27 (H3-K27me2). In one embodiment a method for treating cancer or a precancerous condition in a subject comprises administering to the subject in need thereof a therapeutically effective amount of a compound that inhibits conversion of H3-K27me2 to trimethylated H3-K27 (H3-K27me3). In one embodiment a method for treating cancer or a precancerous condition in a subject comprises administering to the subject in need thereof a therapeutically effective amount of a compound that inhibits both conversion of H3-K27me1 to H3-K27me2 and conversion of H3-K27me2 to H3-K27me3. It is important to note that disease-specific increase in methylation can occur at chromatin in key genomic loci in the absence of a global increase in cellular levels of histone or protein methylation. For example, it is possible for aberrant hypermethylation at key disease-relevant genes to occur against a backdrop of global histone or protein hypomethylation.

[0216] Modulators of methylation can be used for modulating cell proliferation, generally. For example, in some cases excessive proliferation may be reduced with agents that decrease

methylation, whereas insufficient proliferation may be stimulated with agents that increase methylation. Accordingly, diseases that may be treated include hyperproliferative diseases, such as benign cell growth and malignant cell growth (cancer).

[0217] The disorder in which SRC-mediated protein methylation plays a part can be cancer, a cell proliferative disorder, or a precancerous condition. The present invention further provides the use of a composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, to a subject in need of such treatment, for the preparation of a medicament useful for the treatment of cancer. Exemplary cancers that may be treated include lymphomas, including non-Hodgkin lymphoma, follicular lymphoma (FL) and diffuse large B-cell lymphoma (DLBCL); melanoma; and leukemia, including CML. Exemplary precancerous condition includes myelodisplastic syndrome (MDS; formerly known as preleukemia).

[0218] In general, compounds that are methylation modulators can be used for modulating cell proliferation, generally. For example, in some cases excessive proliferation may be reduced with agents that decrease methylation, whereas insufficient proliferation may be stimulated with agents that increase methylation. Accordingly, diseases that may be treated by the Compounds Of The Invention include hyperproliferative diseases, such as benign cell growth and malignant cell growth.

[0219] As used herein, a "subject in need thereof" is a subject having a disorder in which SRC-mediated protein plays a part, or a subject having an increased risk of developing such disorder relative to the population at large. A subject in need thereof can have a precancerous condition. Preferably, a subject in need thereof has cancer. A "subject" includes a mammal. The mammal can be e.g., any mammal, e.g., a human, primate, bird, mouse, rat, fowl, dog, cat, cow, horse, goat, camel, sheep or a pig. Preferably, the mammal is a human.

[0220] The subject of the present invention includes any human subject who has been diagnosed with, has symptoms of, or is at risk of developing a cancer or a precancerous condition. The subject of the present invention includes any human subject expressing a mutant SRC. For example, a mutant SRC comprises one or more mutations, wherein the mutation is a substitution, a point mutation, a nonsense mutation, a missense mutation, a deletion, or an insertion or any other SRC mutation described herein.

[0221] A subject in need thereof may have refractory or resistant cancer. "Refractory or resistant cancer" means cancer that does not respond to treatment. The cancer may be resistant at the beginning of treatment or it may become resistant during treatment. In some

embodiments, the subject in need thereof has cancer recurrence following remission on most recent therapy. In some embodiments, the subject in need thereof received and failed all known effective therapies for cancer treatment. In some embodiments, the subject in need thereof received at least one prior therapy. In certain embodiments the prior therapy is monotherapy. In certain embodiments the prior therapy is combination therapy.

[0222] In some embodiments, a subject in need thereof may have a secondary cancer as a result of a previous therapy. "Secondary cancer" means cancer that arises due to or as a result from previous carcinogenic therapies, such as chemotherapy.

[0223] The subject may also exhibit resistance to SRC histone methyltransferase inhibitors or any other therapeutic agent.

[0224] The invention also features a method of selecting a combination therapy for a subject having cancer. The method includes the steps of: detecting one or more SRC mutations described herein in a sample from the subject; and selecting, based on the presence of the one or more SRC mutations, a combination therapy for treating cancer. In one embodiment, the therapy includes administering to the subject a composition of the invention. In one embodiment, the method further includes administrating to the subject a therapeutically effective amount of a composition of the invention. An SRC mutation can be detected using any suitable method known in the art. More methods are described in U.S. patent publication US 20130040906, which is incorporated herein by reference in their entireties.

[0225] The methods and uses described herein may include steps of detecting one or more SRC mutations described herein in a sample from a subject in need thereof prior to and/or after the administration of a composition of the invention (e.g., a composition comprising a compound of Formula (I), (II), (III), or (IV) or pharmaceutically acceptable salts thereof, and one or more therapeutic agents) to the subject. The presence of the one or more SRC mutations described herein in the tested sample indicates the subject is responsive to the combination therapy of the invention.

[0226] The present invention provides personalized medicine, treatment and/or cancer management for a subject by genetic screening of one or more SRC mutations described herein in the subject. For example, the present invention provides methods for treating or alleviating a symptom of cancer or a precancerous condition in a subject in need thereof by determining responsiveness of the subject to a combination therapy and when the subject is responsive to the combination therapy, administering to the subject a composition of the invention. The responsiveness is determined by obtaining a sample from the subject and detecting one or more

SRC mutations described herein, and the presence of such one or more SRC mutations described herein indicates that the subject is responsive to the composition of the invention. Once the responsiveness of a subject is determined, a therapeutically effective amount of a composition, for example, a composition comprising a compound of Formula (I), (II), (III), or (IV) or pharmaceutically acceptable salts thereof, and one or more therapeutic agents, can be administered. The therapeutically effective amount of a composition can be determined by one of ordinary skill in the art.

[0227] As used herein, the term "responsiveness" is interchangeable with terms "responsive", "sensitive", and "sensitivity", and it is meant that a subject is showing therapeutic responses when administered a composition of the invention, e.g., tumor cells or tumor tissues of the subject undergo apoptosis and/or necrosis, and/or display reduced growing, dividing, or proliferation. This term is also meant that a subject will or has a higher probability, relative to the population at large, of showing therapeutic responses when administered a composition of the invention, e.g., tumor cells or tumor tissues of the subject undergo apoptosis and/or necrosis, and/or display reduced growing, dividing, or proliferation.

[0228] By "sample" it means any biological sample derived from the subject, includes but is not limited to, cells, tissues samples, body fluids (including, but not limited to, mucus, blood, plasma, serum, urine, saliva, and semen), tumor cells, and tumor tissues. Preferably, the sample is selected from bone marrow, peripheral blood cells, blood, plasma and serum. Samples can be provided by the subject under treatment or testing. Alternatively samples can be obtained by the physician according to routine practice in the art.

CONDITIONS, DISORDERS, OR DISEASES THAT CAN BE TREATED OR PREVENTED BY THE COMPOUNDS, COMPOSITIONS, AND COMBINATIONS OF THE INVENTION

[0229] As used herein, the term "cell proliferative disorder" refers to conditions in which unregulated or abnormal growth, or both, of cells can lead to the development of an unwanted condition or disease, which may or may not be cancerous. Exemplary cell proliferative disorders of the invention encompass a variety of conditions wherein cell division is deregulated. Exemplary cell proliferative disorder include, but are not limited to, neoplasms, benign tumors, malignant tumors, pre-cancerous conditions, in situ tumors, encapsulated tumors, metastatic tumors, liquid tumors, solid tumors, immunological tumors, hematological tumors, cancers, carcinomas, leukemias, lymphomas, sarcomas, and rapidly dividing cells. The

term "rapidly dividing cell" as used herein is defined as any cell that divides at a rate that exceeds or is greater than what is expected or observed among neighboring or juxtaposed cells within the same tissue. A cell proliferative disorder includes a precancer or a precancerous condition. A cell proliferative disorder includes cancer. Preferably, the methods provided herein are used to treat or alleviate a symptom of cancer. The term "cancer" includes solid tumors, as well as, hematologic tumors and/or malignancies. A "precancer cell" or "precancerous cell" is a cell manifesting a cell proliferative disorder that is a precancer or a precancerous condition. A "cancer cell" or "cancerous cell" is a cell manifesting a cell proliferative disorder that is a cancer. Any reproducible means of measurement may be used to identify cancer cells or precancerous cells. Cancer cells or precancerous cells can be identified by histological typing or grading of a tissue sample (e.g., a biopsy sample). Cancer cells or precancerous cells can be identified through the use of appropriate molecular markers.

[0230] Exemplary non-cancerous conditions or disorders include, but are not limited to, rheumatoid arthritis; inflammation; autoimmune disease; lymphoproliferative conditions; acromegaly; rheumatoid spondylitis; osteoarthritis; gout, other arthritic conditions; sepsis; septic shock; endotoxic shock; gram-negative sepsis; toxic shock syndrome; asthma; adult respiratory distress syndrome; chronic obstructive pulmonary disease; chronic pulmonary inflammation; inflammatory bowel disease; Crohn's disease; psoriasis; eczema; ulcerative colitis; pancreatic fibrosis; hepatic fibrosis; acute and chronic renal disease; irritable bowel syndrome; pyresis; restenosis; cerebral malaria; stroke and ischemic injury; neural trauma; Alzheimer's disease; Huntington's disease; Parkinson's disease; acute and chronic pain; allergic rhinitis; allergic conjunctivitis; chronic heart failure; acute coronary syndrome; cachexia; malaria; leprosy; leishmaniasis; Lyme disease; Reiter's syndrome; acute synovitis; muscle degeneration, bursitis; tendonitis; tenosynovitis; herniated, ruptures, or prolapsed intervertebral disk syndrome; osteopetrosis; thrombosis; restenosis; silicosis; pulmonary sarcosis; bone resorption diseases, such as osteoporosis; graft-versus-host reaction; Multiple Sclerosis; lupus; fibromyalgia; AIDS and other viral diseases such as Herpes Zoster, Herpes Simplex I or II, influenza virus and cytomegalovirus; and diabetes mellitus.

[0231] Exemplary cancers include, but are not limited to, adrenocortical carcinoma, AIDS-related cancers, AIDS-related lymphoma, anal cancer, anorectal cancer, cancer of the anal canal, appendix cancer, childhood cerebellar astrocytoma, childhood cerebral astrocytoma, basal cell carcinoma, skin cancer (non-melanoma), biliary cancer, extrahepatic bile duct cancer, intrahepatic bile duct cancer, bladder cancer, uringary bladder cancer, bone and joint cancer,

osteosarcoma and malignant fibrous histiocytoma, brain cancer, brain tumor, brain stem glioma, cerebellar astrocytoma, cerebral astrocytoma/malignant glioma, ependymoma, medulloblastoma, supratentorial primitive neuroectodeimal tumors, visual pathway and hypothalamic glioma, breast cancer, bronchial adenomas/carcinoids, carcinoid tumor, gastrointestinal, nervous system cancer, nervous system lymphoma, central nervous system cancer, central nervous system lymphoma, cervical cancer, childhood cancers, chronic lymphocytic leukemia, chronic myelogenous leukemia, chronic myeloproliferative disorders, colon cancer, colorectal cancer, cutaneous T-cell lymphoma, lymphoid neoplasm, mycosis fungoides, Seziary Syndrome, endometrial cancer, esophageal cancer, extracranial germ cell tumor, extragonadal germ cell tumor, extrahepatic bile duct cancer, eye cancer, intraocular melanoma, retinoblastoma, gallbladder cancer, gastric (stomach) cancer, gastrointestinal carcinoid tumor, gastrointestinal stromal tumor (GIST), germ cell tumor, ovarian germ cell tumor, gestational trophoblastic tumor glioma, head and neck cancer, hepatocellular (liver) cancer, Hodgkin lymphoma, hypopharyngeal cancer, intraocular melanoma, ocular cancer, islet cell tumors (endocrine pancreas), Kaposi Sarcoma, kidney cancer, renal cancer, kidney cancer, laryngeal cancer, acute lymphoblastic leukemia, acute myeloid leukemia, chronic lymphocytic leukemia, chronic myelogenous leukemia, hairy cell leukemia, lip and oral cavity cancer, liver cancer, lung cancer, non-small cell lung cancer, small cell lung cancer, AIDS-related lymphoma, non-Hodgkin lymphoma, primary central nervous system lymphoma, Waldenstram macroglobulinemia, medulloblastoma, melanoma, intraocular (eye) melanoma, merkel cell carcinoma, mesothelioma malignant, mesothelioma, metastatic squamous neck cancer, mouth cancer, cancer of the tongue, multiple endocrine neoplasia syndrome, mycosis fungoides, myelodysplastic syndromes, myelodysplastic/myeloproliferative diseases, chronic myelogenous leukemia, acute myeloid leukemia, multiple myeloma, chronic myeloproliferative disorders, nasopharyngeal cancer, neuroblastoma, oral cancer, oral cavity cancer, oropharyngeal cancer, ovarian cancer, ovarian epithelial cancer, ovarian low malignant potential tumor, pancreatic cancer, islet cell pancreatic cancer, paranasal sinus and nasal cavity cancer, parathyroid cancer, penile cancer, pharyngeal cancer, pheochromocytoma, pineoblastoma and supratentorial primitive neuroectodermal tumors, pituitary tumor, plasma cell neoplasm/multiple myeloma, pleuropulmonary blastoma, prostate cancer, rectal cancer, renal pelvis and ureter, transitional cell cancer, retinoblastoma, rhabdomyosarcoma, salivary gland cancer, ewing family of sarcoma tumors, Kaposi Sarcoma, soft tissue sarcoma, uterine cancer, uterine sarcoma, skin cancer (non-melanoma), skin cancer (melanoma), merkel cell

skin carcinoma, small intestine cancer, soft tissue sarcoma, squamous cell carcinoma, stomach (gastric) cancer, supratentorial primitive neuroectodermal tumors, testicular cancer, throat cancer, thymoma, thymoma and thymic carcinoma, thyroid cancer, transitional cell cancer of the renal pelvis and ureter and other urinary organs, gestational trophoblastic tumor, urethral cancer, endometrial uterine cancer, uterine sarcoma, uterine corpus cancer, vaginal cancer, vulvar cancer, and Wilm's Tumor.

[0232] A "cell proliferative disorder of the hematologic system" is a cell proliferative disorder involving cells of the hematologic system. A cell proliferative disorder of the hematologic system can include lymphoma, leukemia, myeloid neoplasms, mast cell neoplasms, myelodysplasia, benign monoclonal gammopathy, lymphomatoid granulomatosis, lymphomatoid papulosis, polycythemia vera, chronic myelocytic leukemia, agnogenic myeloid metaplasia, and essential thrombocythemia. A cell proliferative disorder of the hematologic system can include hyperplasia, dysplasia, and metaplasia of cells of the hematologic system. Preferably, compositions of the present invention may be used to treat a cancer selected from the group consisting of a hematologic cancer of the present invention or a hematologic cell proliferative disorder of the present invention. A hematologic cancer of the present invention can include multiple myeloma, lymphoma (including Hodgkin's lymphoma, non-Hodgkin's lymphoma, childhood lymphomas, and lymphomas of lymphocytic and cutaneous origin), leukemia (including childhood leukemia, hairy-cell leukemia, acute lymphocytic leukemia, acute myelocytic leukemia, chronic lymphocytic leukemia, chronic myelocytic leukemia, chronic myelogenous leukemia, and mast cell leukemia), myeloid neoplasms and mast cell neoplasms.

[0233] A "cell proliferative disorder of the lung" is a cell proliferative disorder involving cells of the lung. Cell proliferative disorders of the lung can include all forms of cell proliferative disorders affecting lung cells. Cell proliferative disorders of the lung can include lung cancer, a precancer or precancerous condition of the lung, benign growths or lesions of the lung, and malignant growths or lesions of the lung, and metastatic lesions in tissue and organs in the body other than the lung. Preferably, compositions of the present invention may be used to treat lung cancer or cell proliferative disorders of the lung. Lung cancer can include all forms of cancer of the lung. Lung cancer can include malignant lung neoplasms, carcinoma in situ, typical carcinoid tumors, and atypical carcinoid tumors. Lung cancer can include small cell lung cancer ("SCLC"), non-small cell lung cancer ("NSCLC"), squamous cell carcinoma, adenocarcinoma, small cell carcinoma, large cell carcinoma, adenosquamous cell carcinoma, and mesothelioma.

Lung cancer can include "scar carcinoma," bronchioalveolar carcinoma, giant cell carcinoma, spindle cell carcinoma, and large cell neuroendocrine carcinoma. Lung cancer can include lung neoplasms having histologic and ultrastructual heterogeneity (e.g., mixed cell types).

[0234] Cell proliferative disorders of the lung can include all forms of cell proliferative disorders affecting lung cells. Cell proliferative disorders of the lung can include lung cancer, precancerous conditions of the lung. Cell proliferative disorders of the lung can include hyperplasia, metaplasia, and dysplasia of the lung. Cell proliferative disorders of the lung can include asbestos-induced hyperplasia, squamous metaplasia, and benign reactive mesothelial metaplasia. Cell proliferative disorders of the lung can include replacement of columnar epithelium with stratified squamous epithelium, and mucosal dysplasia. Individuals exposed to inhaled injurious environmental agents such as cigarette smoke and asbestos may be at increased risk for developing cell proliferative disorders of the lung. Prior lung diseases that may predispose individuals to development of cell proliferative disorders of the lung can include chronic interstitial lung disease, necrotizing pulmonary disease, scleroderma, rheumatoid disease, sarcoidosis, interstitial pneumonitis, tuberculosis, repeated pneumonias, idiopathic pulmonary fibrosis, granulomata, asbestosis, fibrosing alveolitis, and Hodgkin's disease.

[0235] A "cell proliferative disorder of the colon" is a cell proliferative disorder involving cells of the colon. Preferably, the cell proliferative disorder of the colon is colon cancer. Preferably, compositions of the present invention may be used to treat colon cancer or cell proliferative disorders of the colon. Colon cancer can include all forms of cancer of the colon. Colon cancer can include sporadic and hereditary colon cancers. Colon cancer can include malignant colon neoplasms, carcinoma in situ, typical carcinoid tumors, and atypical carcinoid tumors. Colon cancer can include adenocarcinoma, squamous cell carcinoma, and adenosquamous cell carcinoma. Colon cancer can be associated with a hereditary syndrome selected from the group consisting of hereditary nonpolyposis colorectal cancer, familial adenomatous polyposis, Gardner's syndrome, Peutz-Jeghers syndrome, Turcot's syndrome and juvenile polyposis. Colon cancer can be caused by a hereditary syndrome selected from the group consisting of hereditary nonpolyposis colorectal cancer, familial adenomatous polyposis, Gardner's syndrome, Peutz-Jeghers syndrome, Turcot's syndrome and juvenile polyposis.

[0236] Cell proliferative disorders of the colon can include all forms of cell proliferative disorders affecting colon cells. Cell proliferative disorders of the colon can include colon cancer, precancerous conditions of the colon, adenomatous polyps of the colon and

metachronous lesions of the colon. A cell proliferative disorder of the colon can include adenoma. Cell proliferative disorders of the colon can be characterized by hyperplasia, metaplasia, and dysplasia of the colon. Prior colon diseases that may predispose individuals to development of cell proliferative disorders of the colon can include prior colon cancer. Current disease that may predispose individuals to development of cell proliferative disorders of the colon can include Crohn's disease and ulcerative colitis. A cell proliferative disorder of the colon can be associated with a mutation in a gene selected from the group consisting of p53, ras, FAP and DCC. An individual can have an elevated risk of developing a cell proliferative disorder of the colon due to the presence of a mutation in a gene selected from the group consisting of p53, ras, FAP and DCC.

[0237] A "cell proliferative disorder of the pancreas" is a cell proliferative disorder involving cells of the pancreas. Cell proliferative disorders of the pancreas can include all forms of cell proliferative disorders affecting pancreatic cells. Cell proliferative disorders of the pancreas can include pancreas cancer, a precancer or precancerous condition of the pancreas, hyperplasia of the pancreas, and dysaplasia of the pancreas, benign growths or lesions of the pancreas, and malignant growths or lesions of the pancreas, and metastatic lesions in tissue and organs in the body other than the pancreas. Pancreatic cancer includes all forms of cancer of the pancreas. Pancreatic cancer can include ductal adenocarcinoma, adenosquamous carcinoma, pleomorphic giant cell carcinoma, mucinous adenocarcinoma, osteoclast-like giant cell carcinoma, mucinous cystadenocarcinoma, acinar carcinoma, unclassified large cell carcinoma, small cell carcinoma, pancreatoblastoma, papillary neoplasm, mucinous cystadenoma, papillary cystic neoplasm, and serous cystadenoma. Pancreatic cancer can also include pancreatic neoplasms having histologic and ultrastructual heterogeneity (e.g., mixed cell types).

[0238] A "cell proliferative disorder of the prostate" is a cell proliferative disorder involving cells of the prostate. Cell proliferative disorders of the prostate can include all forms of cell proliferative disorders affecting prostate cells. Cell proliferative disorders of the prostate can include prostate cancer, a precancer or precancerous condition of the prostate, benign growths or lesions of the prostate, and malignant growths or lesions of the prostate, and metastatic lesions in tissue and organs in the body other than the prostate. Cell proliferative disorders of the prostate can include hyperplasia, metaplasia, and dysplasia of the prostate.

[0239] A "cell proliferative disorder of the skin" is a cell proliferative disorder involving cells of the skin. Cell proliferative disorders of the skin can include all forms of cell proliferative

disorders affecting skin cells. Cell proliferative disorders of the skin can include a precancer or precancerous condition of the skin, benign growths or lesions of the skin, melanoma, malignant melanoma and other malignant growths or lesions of the skin, and metastatic lesions in tissue and organs in the body other than the skin. Cell proliferative disorders of the skin can include hyperplasia, metaplasia, and dysplasia of the skin.

[0240] A "cell proliferative disorder of the ovary" is a cell proliferative disorder involving cells of the ovary. Cell proliferative disorders of the ovary can include all forms of cell proliferative disorders affecting cells of the ovary. Cell proliferative disorders of the ovary can include a precancer or precancerous condition of the ovary, benign growths or lesions of the ovary, ovarian cancer, malignant growths or lesions of the ovary, and metastatic lesions in tissue and organs in the body other than the ovary. Cell proliferative disorders of the skin can include hyperplasia, metaplasia, and dysplasia of cells of the ovary.

[0241] A "cell proliferative disorder of the breast" is a cell proliferative disorder involving cells of the breast. Cell proliferative disorders of the breast can include all forms of cell proliferative disorders affecting breast cells. Cell proliferative disorders of the breast can include breast cancer, a pre-cancer or precancerous condition of the breast, benign growths or lesions of the breast, and malignant growths or lesions of the breast, and metastatic lesions in tissue and organs in the body other than the breast. Cell proliferative disorders of the breast can include hyperplasia, metaplasia, and dysplasia of the breast.

[0242] A cell proliferative disorder of the breast can be a precancerous condition of the breast. Compositions of the present invention may be used to treat a precancerous condition of the breast. A precancerous condition of the breast can include atypical hyperplasia of the breast, ductal carcinoma in situ (DCIS), intraductal carcinoma, lobular carcinoma in situ (LCIS), lobular neoplasia, and stage 0 or grade 0 growth or lesion of the breast (e.g., stage 0 or grade 0 breast cancer, or carcinoma in situ). A precancerous condition of the breast can be staged according to the TNM classification scheme as accepted by the American Joint Committee on Cancer (AJCC), where the primary tumor (T) has been assigned a stage of TO or Tis; and where the regional lymph nodes (N) have been assigned a stage of NO; and where distant metastasis (M) has been assigned a stage of MO.

[0243] The cell proliferative disorder of the breast can be breast cancer. Preferably, compositions of the present invention may be used to treat breast cancer. Breast cancer includes all forms of cancer of the breast. Breast cancer can include primary epithelial breast cancers. Breast cancer can include cancers in which the breast is involved by other tumors such as

lymphoma, sarcoma or melanoma. Breast cancer can include carcinoma of the breast, ductal carcinoma of the breast, lobular carcinoma of the breast, undifferentiated carcinoma of the breast, cystosarcoma phyllodes of the breast, angiosarcoma of the breast, and primary lymphoma of the breast. Breast cancer can include Stage I, II, II, IIIB, IIIC and IV breast cancer. Ductal carcinoma of the breast can include invasive carcinoma, invasive carcinoma in situ with predominant intraductal component, inflammatory breast cancer, and a ductal carcinoma of the breast with a histologic type selected from the group consisting of comedo, mucinous (colloid), medullary, medullary with lymphcytic infiltrate, papillary, scirrhous, and tubular. Lobular carcinoma of the breast can include invasive lobular carcinoma with predominant in situ component, invasive lobular carcinoma, and infiltrating lobular carcinoma. Breast cancer can include Paget's disease, Paget's disease with intraductal carcinoma, and Paget's disease with invasive ductal carcinoma. Breast cancer can include breast neoplasms having histologic and ultrastructual heterogeneity (e.g., mixed cell types).

[0244] Preferably, compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph, or solvate thereof, may be used to treat breast cancer. A breast cancer that is to be treated can include familial breast cancer. A breast cancer that is to be treated can include sporadic breast cancer. A breast cancer that is to be treated can arise in a male subject. A breast cancer that is to be treated can arise in a female subject. A breast cancer that is to be treated can arise in a premenopausal female subject or a postmenopausal female subject. A breast cancer that is to be treated can arise in a subject equal to or older than 30 years old, or a subject younger than 30 years old. A breast cancer that is to be treated has arisen in a subject equal to or older than 50 years old, or a subject younger than 50 years old. A breast cancer that is to be treated can arise in a subject equal to or older than 70 years old, or a subject younger than 70 years old.

[0245] A breast cancer that is to be treated can be typed to identify a familial or spontaneous mutation in BRCA1, BRCA2, or p53. A breast cancer that is to be treated can be typed as having a HER2/neu gene amplification, as overexpressing HER2/neu, or as having a low, intermediate or high level of HER2/neu expression. A breast cancer that is to be treated can be typed for a marker selected from the group consisting of estrogen receptor (ER), progesterone receptor (PR), human epidermal growth factor receptor-2, Ki-67, CA15-3, CA 27-29, and c-Met. A breast cancer that is to be treated can be typed as ER-unknown, ER-rich or ER-poor. A breast cancer that is to be treated can be typed as ER-negative or ER-positive. ER-typing of a breast cancer may be performed by any reproducible means. ER-typing of a breast cancer may

be performed as set forth in Onkologie 27: 175-179 (2004). A breast cancer that is to be treated can be typed as PR-unknown, PR-rich, or PR-poor. A breast cancer that is to be treated can be typed as PR-negative or PR-positive. A breast cancer that is to be treated can be typed as receptor positive or receptor negative. A breast cancer that is to be treated can be typed as being associated with elevated blood levels of CA 15-3, or CA 27-29, or both.

[0246] A breast cancer that is to be treated can include a localized tumor of the breast. A breast cancer that is to be treated can include a tumor of the breast that is associated with a negative sentinel lymph node (SLN) biopsy. A breast cancer that is to be treated can include a tumor of the breast that is associated with a positive sentinel lymph node (SLN) biopsy. A breast cancer that is to be treated can include a tumor of the breast that is associated with one or more positive axillary lymph nodes, where the axillary lymph nodes have been staged by any applicable method. A breast cancer that is to be treated can include a tumor of the breast that has been typed as having nodal negative status (e.g., node-negative) or nodal positive status (e.g., node-positive). A breast cancer that is to be treated can include a tumor of the breast that has metastasized to other locations in the body. A breast cancer that is to be treated can be classified as having metastasized to a location selected from the group consisting of bone, lung, liver, or brain. A breast cancer that is to be treated can be classified according to a characteristic selected from the group consisting of metastatic, localized, regional, local-regional, locally advanced, distant, multicentric, bilateral, ipsilateral, contralateral, newly diagnosed, recurrent, and inoperable.

[0247] A compound or composition of the present invention, or a pharmaceutically acceptable salt, ester, prodrug, metabolite, polymorph or solvate thereof, may be used to treat or prevent a cell proliferative disorder of the breast, or to treat or prevent breast cancer, in a subject having an increased risk of developing breast cancer relative to the population at large. A subject with an increased risk of developing breast cancer relative to the population at large is a female subject with a family history or personal history of breast cancer. A subject with an increased risk of developing breast cancer relative to the population at large is a female subject having a germ-line or spontaneous mutation in BRCA1 or BRCA2, or both. A subject with an increased risk of developing breast cancer relative to the population at large is a female subject with a family history of breast cancer and a germ-line or spontaneous mutation in BRCA1 or BRCA2, or both. A subject with an increased risk of developing breast cancer relative to the population at large is a female who is greater than 30 years old, greater than 40 years old, greater than 50 years old, greater than 60 years old, greater than 70 years old, greater than 80 years old, or

greater than 90 years old. A subject with an increased risk of developing breast cancer relative to the population at large is a subject with atypical hyperplasia of the breast, ductal carcinoma in situ (DCIS), intraductal carcinoma, lobular carcinoma in situ (LCIS), lobular neoplasia, or a stage 0 growth or lesion of the breast (e.g., stage 0 or grade 0 breast cancer, or carcinoma in situ).

[0248] A breast cancer that is to be treated can histologically graded according to the Scarff-Bloom-Richardson system, wherein a breast tumor has been assigned a mitosis count score of 1, 2, or 3; a nuclear pleiomorphism score of 1, 2, or 3; a tubule formation score of 1, 2, or 3; and a total Scarff-Bloom-Richardson score of between 3 and 9. A breast cancer that is to be treated can be assigned a tumor grade according to the International Consensus Panel on the Treatment of Breast Cancer selected from the group consisting of grade 1, grade 1-2, grade 2, grade 2-3, or grade 3.

[0249] A cancer that is to be treated can be staged according to the American Joint Committee on Cancer (AJCC) TNM classification system, where the tumor (T) has been assigned a stage of TX, T1, T1mic, T1a, T1b, T1c, T2, T3, T4, T4a, T4b, T4c, or T4d; and where the regional lymph nodes (N) have been assigned a stage of NX, N0, N1, N2, N2a, N2b, N3, N3a, N3b, or N3c; and where distant metastasis (M) can be assigned a stage of MX, M0, or Ml. A cancer that is to be treated can be staged according to an American Joint Committee on Cancer (AJCC) classification as Stage I, Stage I, Stage IIB, Stage II, Stage IIIB, Stage IIIC, or Stage IV. A cancer that is to be treated can be assigned a grade according to an AJCC classification as Grade GX (e.g., grade cannot be assessed), Grade 1, Grade 2, Grade 3 or Grade 4. A cancer that is to be treated can be staged according to an AJCC pathologic classification (pN) of pNX, pN0, PN0 (I-), PN0 (I+), PN0 (mol-), PN0 (mol+), PN1, PN1(mi), PN1a, PN1b, PN1c, pN2, pN2a, pN2b, pN3, pN3a, pN3b, or pN3c.

[0250] A cancer that is to be treated can include a tumor that has been determined to be less than or equal to about 2 centimeters in diameter. A cancer that is to be treated can include a tumor that has been determined to be from about 2 to about 5 centimeters in diameter. A cancer that is to be treated can include a tumor that has been determined to be greater than or equal to about 3 centimeters in diameter. A cancer that is to be treated can include a tumor that has been determined to be greater than 5 centimeters in diameter. A cancer that is to be treated can be classified by microscopic appearance as well differentiated, moderately differentiated, poorly differentiated, or undifferentiated. A cancer that is to be treated can be classified by microscopic appearance with respect to mitosis count (e.g., amount of cell division) or nuclear

pleiomorphism (e.g., change in cells). A cancer that is to be treated can be classified by microscopic appearance as being associated with areas of necrosis (e.g., areas of dying or degenerating cells). A cancer that is to be treated can be classified as having an abnormal karyotype, having an abnormal number of chromosomes, or having one or more chromosomes that are abnormal in appearance. A cancer that is to be treated can be classified as being aneuploid, triploid, tetraploid, or as having an altered ploidy. A cancer that is to be treated can be classified as having a chromosomal translocation, or a deletion or duplication of an entire chromosome, or a region of deletion, duplication or amplification of a portion of a chromosome.

[0251] A cancer that is to be treated can be evaluated by DNA cytometry, flow cytometry, or image cytometry. A cancer that is to be treated can be typed as having 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, or 90% of cells in the synthesis stage of cell division (e.g., in S phase of cell division). A cancer that is to be treated can be typed as having a low S-phase fraction or a high S-phase fraction.

[0252] As used herein, a "normal cell" is a cell that cannot be classified as part of a "cell proliferative disorder". A normal cell lacks unregulated or abnormal growth, or both, that can lead to the development of an unwanted condition or disease. Preferably, a normal cell possesses normally functioning cell cycle checkpoint control mechanisms.

[0253] As used herein, "contacting a cell" refers to a condition in which a compound or other composition of matter is in direct contact with a cell, or is close enough to induce a desired biological effect in a cell.

[0254] As used herein, "candidate compound" or “compound of the invention” refers to a compound of Formula (I), (II), (III), or (IV) or a pharmaceutically acceptable salt, ester, prodrug, metabolite, polymorph or solvate thereof, that has been or will be tested in one or more in vitro or in vivo biological assays, in order to determine if that compound is likely to elicit a desired biological or medical response in a cell, tissue, system, animal or human that is being sought by a researcher or clinician. A candidate compound is a compound of the present invention, or a pharmaceutically acceptable salt, ester, prodrug, metabolite, polymorph or solvate thereof. The biological or medical response can be the treatment of cancer. The biological or medical response can be treatment or prevention of a cell proliferative disorder. In vitro or in vivo biological assays can include, but are not limited to, enzymatic activity assays, electrophoretic mobility shift assays, reporter gene assays, in vitro cell viability assays, and the assays described herein.

[0255] As used herein, "treating" or "treat" describes the management and care of a patient for the purpose of combating a disease, condition, or disorder and includes the administration of a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, to alleviate the symptoms or complications of a disease, condition or disorder, or to eliminate the disease, condition or disorder.

[0256] A composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, can also be used to prevent a disease, condition or disorder. As used herein, "preventing" or "prevent" describes reducing or eliminating the onset of the symptoms or complications of the disease, condition or disorder.

[0257] As used herein, the term "alleviate" is meant to describe a process by which the severity of a sign or symptom of a disorder is decreased. Importantly, a sign or symptom can be alleviated without being eliminated. In a preferred embodiment, the administration of pharmaceutical compositions of the invention leads to the elimination of a sign or symptom, however, elimination is not required. Effective dosages are expected to decrease the severity of a sign or symptom. For instance, a sign or symptom of a disorder such as cancer, which can occur in multiple locations, is alleviated if the severity of the cancer is decreased within at least one of multiple locations.

[0258] As used herein, the term "severity" is meant to describe the potential of cancer to transform from a precancerous, or benign, state into a malignant state. Alternatively, or in addition, severity is meant to describe a cancer stage, for example, according to the TNM system (accepted by the International Union Against Cancer (UICC) and the American Joint Committee on Cancer (AJCC)) or by other art-recognized methods. Cancer stage refers to the extent or severity of the cancer, based on factors such as the location of the primary tumor, tumor size, number of tumors, and lymph node involvement (spread of cancer into lymph nodes). Alternatively, or in addition, severity is meant to describe the tumor grade by art-recognized methods (see, National Cancer Institute, www.cancer.gov). Tumor grade is a system used to classify cancer cells in terms of how abnormal they look under a microscope and how quickly the tumor is likely to grow and spread. Many factors are considered when determining tumor grade, including the structure and growth pattern of the cells. The specific factors used to determine tumor grade vary with each type of cancer. Severity also describes a histologic grade, also called differentiation, which refers to how much the tumor cells resemble normal cells of the same tissue type (see, National Cancer Institute, www.cancer.gov). Furthermore, severity describes a nuclear grade, which refers to the size and shape of the

nucleus in tumor cells and the percentage of tumor cells that are dividing (see, National Cancer Institute, www.cancer.gov).

[0259] In another aspect of the invention, severity describes the degree to which a tumor has secreted growth factors, degraded the extracellular matrix, become vascularized, lost adhesion to juxtaposed tissues, or metastasized. Moreover, severity describes the number of locations to which a primary tumor has metastasized. Finally, severity includes the difficulty of treating tumors of varying types and locations. For example, inoperable tumors, those cancers which have greater access to multiple body systems (hematological and immunological tumors), and those which are the most resistant to traditional treatments are considered most severe. In these situations, prolonging the life expectancy of the subject and/or reducing pain, decreasing the proportion of cancerous cells or restricting cells to one system, and improving cancer stage/tumor grade/histological grade/nuclear grade are considered alleviating a sign or symptom of the cancer.

[0260] As used herein the term "symptom" is defined as an indication of disease, illness, injury, or that something is not right in the body. Symptoms are felt or noticed by the individual experiencing the symptom, but may not easily be noticed by others. Others are defined as non-health-care professionals.

[0261] As used herein the term "sign" is also defined as an indication that something is not right in the body. But signs are defined as things that can be seen by a doctor, nurse, or other health care professional.

[0262] Cancer is a group of diseases that may cause almost any sign or symptom. The signs and symptoms will depend on where the cancer is, the size of the cancer, and how much it affects the nearby organs or structures. If a cancer spreads (metastasizes), then symptoms may appear in different parts of the body.

[0263] The disorder in which SRC-mediated protein methylation plays a part can be a neurological disease. The compound of this invention can thus also be used for treating neurologic diseases such as epilepsy, schizophrenia, bipolar disorder or other psychological and/or psychiatric disorders, neuropathies, skeletal muscle atrophy, and neurodegenerative diseases, e.g., a neurodegenerative disease. Exemplary neurodegenerative diseases include: Alzheimer's, Amyotrophic Lateral Sclerosis (ALS), and Parkinson's disease. Another class of neurodegenerative diseases includes diseases caused at least in part by aggregation of poly-glutamine. Diseases of this class include: Huntington's Diseases, Spinalbulbar Muscular Atrophy (SBMA or Kennedy's Disease) Dentatorubropallidoluysian Atrophy (DRPLA),

Spinocerebellar Ataxia 1 (SCA1), Spinocerebellar Ataxia 2 (SCA2), Machado-Joseph Disease m (MJD; SCA3), Spinocerebellar Ataxia 6 (SCA6), Spinocerebellar Ataxia 7 (SCAT), and Spinocerebellar Ataxia 12 (SCA12).

[0264] Any other disease in which epigenetic methylation, which is mediated by SRC, plays a role may be treatable or preventable using compositions and methods described herein.

[0265] Treating cancer can result in a reduction in size of a tumor. A reduction in size of a tumor may also be referred to as "tumor regression". Preferably, after treatment, tumor size is reduced by 5% or greater relative to its size prior to treatment; more preferably, tumor size is reduced by 10% or greater; more preferably, reduced by 20% or greater; more preferably, reduced by 30% or greater; more preferably, reduced by 40% or greater; even more preferably, reduced by 50% or greater; and most preferably, reduced by greater than 75% or greater. Size of a tumor may be measured by any reproducible means of measurement. The size of a tumor may be measured as a diameter of the tumor.

[0266] Treating cancer can result in a reduction in tumor volume. Preferably, after treatment, tumor volume is reduced by 5% or greater relative to its size prior to treatment; more preferably, tumor volume is reduced by 10% or greater; more preferably, reduced by 20% or greater; more preferably, reduced by 30% or greater; more preferably, reduced by 40% or greater; even more preferably, reduced by 50% or greater; and most preferably, reduced by greater than 75% or greater. Tumor volume may be measured by any reproducible means of measurement.

[0267] Treating cancer results in a decrease in number of tumors. Preferably, after treatment, tumor number is reduced by 5% or greater relative to number prior to treatment; more preferably, tumor number is reduced by 10% or greater; more preferably, reduced by 20% or greater; more preferably, reduced by 30% or greater; more preferably, reduced by 40% or greater; even more preferably, reduced by 50% or greater; and most preferably, reduced by greater than 75%. Number of tumors may be measured by any reproducible means of measurement. The number of tumors may be measured by counting tumors visible to the naked eye or at a specified magnification. Preferably, the specified magnification is 2.times., 3.times., 4.times., 5.times., 10.times., or 50.times..

[0268] Treating cancer can result in a decrease in number of metastatic lesions in other tissues or organs distant from the primary tumor site. Preferably, after treatment, the number of metastatic lesions is reduced by 5% or greater relative to number prior to treatment; more preferably, the number of metastatic lesions is reduced by 10% or greater; more preferably,

reduced by 20% or greater; more preferably, reduced by 30% or greater; more preferably, reduced by 40% or greater; even more preferably, reduced by 50% or greater; and most preferably, reduced by greater than 75%. The number of metastatic lesions may be measured by any reproducible means of measurement. The number of metastatic lesions may be measured by counting metastatic lesions visible to the naked eye or at a specified magnification. Preferably, the specified magnification is 2x, 3x, 4x, 5x, 10x, or 50x.

[0269] Treating cancer can result in an increase in average survival time of a population of treated subjects in comparison to a population receiving carrier alone. Preferably, the average survival time is increased by more than 30 days; more preferably, by more than 60 days; more preferably, by more than 90 days; and most preferably, by more than 120 days. An increase in average survival time of a population may be measured by any reproducible means. An increase in average survival time of a population may be measured, for example, by calculating for a population the average length of survival following initiation of treatment with an active compound. An increase in average survival time of a population may also be measured, for example, by calculating for a population the average length of survival following completion of a first round of treatment with an active compound.

[0270] Treating cancer can result in an increase in average survival time of a population of treated subjects in comparison to a population of untreated subjects. Preferably, the average survival time is increased by more than 30 days; more preferably, by more than 60 days; more preferably, by more than 90 days; and most preferably, by more than 120 days. An increase in average survival time of a population may be measured by any reproducible means. An increase in average survival time of a population may be measured, for example, by calculating for a population the average length of survival following initiation of treatment with an active compound. An increase in average survival time of a population may also be measured, for example, by calculating for a population the average length of survival following completion of a first round of treatment with an active compound.

[0271] Treating cancer can result in increase in average survival time of a population of treated subjects in comparison to a population receiving monotherapy with a drug that is not a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, analog or derivative thereof. Preferably, the average survival time is increased by more than 30 days; more preferably, by more than 60 days; more preferably, by more than 90 days; and most preferably, by more than 120 days. An increase in average survival time of a population may be measured by any reproducible means. An increase in average survival time of a population

may be measured, for example, by calculating for a population the average length of survival following initiation of treatment with an active compound. An increase in average survival time of a population may also be measured, for example, by calculating for a population the average length of survival following completion of a first round of treatment with an active compound.

[0272] Treating cancer can result in a decrease in the mortality rate of a population of treated subjects in comparison to a population receiving carrier alone. Treating cancer can result in a decrease in the mortality rate of a population of treated subjects in comparison to an untreated population. Treating cancer can result in a decrease in the mortality rate of a population of treated subjects in comparison to a population receiving monotherapy with a drug that is not a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, analog or derivative thereof. Preferably, the mortality rate is decreased by more than 2%; more preferably, by more than 5%; more preferably, by more than 10%; and most preferably, by more than 25%. A decrease in the mortality rate of a population of treated subjects may be measured by any reproducible means. A decrease in the mortality rate of a population may be measured, for example, by calculating for a population the average number of disease-related deaths per unit time following initiation of treatment with an active compound. A decrease in the mortality rate of a population may also be measured, for example, by calculating for a population the average number of disease-related deaths per unit time following completion of a first round of treatment with an active compound.

[0273] Treating cancer can result in a decrease in tumor growth rate. Preferably, after treatment, tumor growth rate is reduced by at least 5% relative to number prior to treatment; more preferably, tumor growth rate is reduced by at least 10%; more preferably, reduced by at least 20%; more preferably, reduced by at least 30%; more preferably, reduced by at least 40%; more preferably, reduced by at least 50%; even more preferably, reduced by at least 50%; and most preferably, reduced by at least 75%. Tumor growth rate may be measured by any reproducible means of measurement. Tumor growth rate can be measured according to a change in tumor diameter per unit time.

[0274] Treating cancer can result in a decrease in tumor regrowth. Preferably, after treatment, tumor regrowth is less than 5%; more preferably, tumor regrowth is less than 10%; more preferably, less than 20%; more preferably, less than 30%; more preferably, less than 40%; more preferably, less than 50%; even more preferably, less than 50%; and most preferably, less than 75%. Tumor regrowth may be measured by any reproducible means of measurement.

Tumor regrowth is measured, for example, by measuring an increase in the diameter of a tumor after a prior tumor shrinkage that followed treatment. A decrease in tumor regrowth is indicated by failure of tumors to reoccur after treatment has stopped.

[0275] Treating or preventing a cell proliferative disorder can result in a reduction in the rate of cellular proliferation. Preferably, after treatment, the rate of cellular proliferation is reduced by at least 5%; more preferably, by at least 10%; more preferably, by at least 20%; more preferably, by at least 30%; more preferably, by at least 40%; more preferably, by at least 50%; even more preferably, by at least 50%; and most preferably, by at least 75%. The rate of cellular proliferation may be measured by any reproducible means of measurement. The rate of cellular proliferation is measured, for example, by measuring the number of dividing cells in a tissue sample per unit time.

[0276] Treating or preventing a cell proliferative disorder can result in a reduction in the proportion of proliferating cells. Preferably, after treatment, the proportion of proliferating cells is reduced by at least 5%; more preferably, by at least 10%; more preferably, by at least 20%; more preferably, by at least 30%; more preferably, by at least 40%; more preferably, by at least 50%; even more preferably, by at least 50%; and most preferably, by at least 75%. The proportion of proliferating cells may be measured by any reproducible means of measurement. Preferably, the proportion of proliferating cells is measured, for example, by quantifying the number of dividing cells relative to the number of nondividing cells in a tissue sample. The proportion of proliferating cells can be equivalent to the mitotic index.

[0277] Treating or preventing a cell proliferative disorder can result in a decrease in size of an area or zone of cellular proliferation. Preferably, after treatment, size of an area or zone of cellular proliferation is reduced by at least 5% relative to its size prior to treatment; more preferably, reduced by at least 10%; more preferably, reduced by at least 20%; more preferably, reduced by at least 30%; more preferably, reduced by at least 40%; more preferably, reduced by at least 50%; even more preferably, reduced by at least 50%; and most preferably, reduced by at least 75%. Size of an area or zone of cellular proliferation may be measured by any reproducible means of measurement. The size of an area or zone of cellular proliferation may be measured as a diameter or width of an area or zone of cellular proliferation.

[0278] Treating or preventing a cell proliferative disorder can result in a decrease in the number or proportion of cells having an abnormal appearance or morphology. Preferably, after treatment, the number of cells having an abnormal morphology is reduced by at least 5% relative to its size prior to treatment; more preferably, reduced by at least 10%; more preferably,

reduced by at least 20%; more preferably, reduced by at least 30%; more preferably, reduced by at least 40%; more preferably, reduced by at least 50%; even more preferably, reduced by at least 50%; and most preferably, reduced by at least 75%. An abnormal cellular appearance or morphology may be measured by any reproducible means of measurement. An abnormal cellular morphology can be measured by microscopy, e.g., using an inverted tissue culture microscope. An abnormal cellular morphology can take the form of nuclear pleiomorphism.

[0279] As used herein, the term "selectively" means tending to occur at a higher frequency in one population than in another population. The compared populations can be cell populations. Preferably, a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, acts selectively on a cancer or precancerous cell but not on a normal cell. Preferably, a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, acts selectively to modulate one molecular target (e.g., a target protein methyltransferase) but does not significantly modulate another molecular target (e.g., a non-target protein methyltransferase). The invention also provides a method for selectively inhibiting the activity of an enzyme, such as a protein methyltransferase. Preferably, an event occurs selectively in population A relative to population B if it occurs greater than two times more frequently in population A as compared to population B. An event occurs selectively if it occurs greater than five times more frequently in population A. An event occurs selectively if it occurs greater than ten times more frequently in population A; more preferably, greater than fifty times; even more preferably, greater than 100 times; and most preferably, greater than 1000 times more frequently in population A as compared to population B. For example, cell death would be said to occur selectively in cancer cells if it occurred greater than twice as frequently in cancer cells as compared to normal cells.

[0280] In certain embodiments, a composition of the present invention (*e.g.*, a composition comprising any compound of Formula (I), (II), (III), or (IV) or pharmaceutically acceptable salt thereof), and one or more other therapeutic agents, such as prednisone, can modulate the activity of a molecular target (e.g., a target protein methyltransferase). Modulating refers to stimulating or inhibiting an activity of a molecular target. Preferably, a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, modulates the activity of a molecular target if it stimulates or inhibits the activity of the molecular target by at least 2-fold relative to the activity of the molecular target under the same conditions but lacking only the presence of said compound. More preferably, a

compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, modulates the activity of a molecular target if it stimulates or inhibits the activity of the molecular target by at least 5-fold, at least 10-fold, at least 20-fold, at least 50-fold, at least 100-fold relative to the activity of the molecular target under the same conditions but lacking only the presence of said compound. The activity of a molecular target may be measured by any reproducible means. The activity of a molecular target may be measured in vitro or in vivo. For example, the activity of a molecular target may be measured in vitro by an enzymatic activity assay or a DNA binding assay, or the activity of a molecular target may be measured in vivo by assaying for expression of a reporter gene.

[0281] A composition of the present invention does not significantly modulate the activity of a molecular target if the addition of the compound does not stimulate or inhibit the activity of the molecular target by greater than 10% relative to the activity of the molecular target under the same conditions but lacking only the presence of said compound.

[0282] As used herein, the term "isozyme selective" means preferential inhibition or stimulation of a first isoform of an enzyme in comparison to a second isoform of an enzyme (e.g., preferential inhibition or stimulation of a protein methyltransferase isozyme alpha in comparison to a protein methyltransferase isozyme beta). Preferably, a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, demonstrates a minimum of a fourfold differential, preferably a tenfold differential, more preferably a fifty fold differential, in the dosage required to achieve a biological effect. Preferably, a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, demonstrates this differential across the range of inhibition, and the differential is exemplified at the IC5o, i.e., a 50% inhibition, for a molecular target of interest.

[0283] Administering a composition of the present invention to a cell or a subject in need thereof can result in modulation (i.e., stimulation or inhibition) of an activity of a protein methyltransferase of interest.

[0284] Administering a Compound Of The Invention, e.g., a composition comprising any compound of Formula (I), (II), (III), or (IV) or pharmaceutically acceptable salt thereof, and one or more other therapeutic agents, such as prednisone, to a cell or a subject in need thereof results in modulation (i.e., stimulation or inhibition) of an activity of an intracellular target (e.g., substrate). Several intracellular targets can be modulated with the compounds of the present invention, including, but not limited to, protein methyltrasferase.

[0285] Activating refers to placing a composition of matter (e.g., protein or nucleic acid) in a state suitable for carrying out a desired biological function. A composition of matter capable of being activated also has an unactivated state. An activated composition of matter may have an inhibitory or stimulatory biological function, or both.

[0286] Elevation refers to an increase in a desired biological activity of a composition of matter (e.g., a protein or a nucleic acid). Elevation may occur through an increase in concentration of a composition of matter.

[0287] As used herein, "a cell cycle checkpoint pathway" refers to a biochemical pathway that is involved in modulation of a cell cycle checkpoint. A cell cycle checkpoint pathway may have stimulatory or inhibitory effects, or both, on one or more functions comprising a cell cycle checkpoint. A cell cycle checkpoint pathway is comprised of at least two compositions of matter, preferably proteins, both of which contribute to modulation of a cell cycle checkpoint. A cell cycle checkpoint pathway may be activated through an activation of one or more members of the cell cycle checkpoint pathway. Preferably, a cell cycle checkpoint pathway is a biochemical signaling pathway.

[0288] As used herein, "cell cycle checkpoint regulator" refers to a composition of matter that can function, at least in part, in modulation of a cell cycle checkpoint. A cell cycle checkpoint regulator may have stimulatory or inhibitory effects, or both, on one or more functions comprising a cell cycle checkpoint. A cell cycle checkpoint regulator can be a protein or not a protein.

[0289] Treating cancer or a cell proliferative disorder can result in cell death, and preferably, cell death results in a decrease of at least 10% in number of cells in a population. More preferably, cell death means a decrease of at least 20%; more preferably, a decrease of at least 30%; more preferably, a decrease of at least 40%; more preferably, a decrease of at least 50%; most preferably, a decrease of at least 75%. Number of cells in a population may be measured by any reproducible means. A number of cells in a population can be measured by fluorescence activated cell sorting (FACS), immunofluorescence microscopy and light microscopy. Methods of measuring cell death are as shown in Li et al., Proc Natl Acad Sci U.S.A. 100(5): 2674-8, 2003. In an aspect, cell death occurs by apoptosis.

[0290] Preferably, an effective amount of a composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, is not significantly cytotoxic to normal cells. A therapeutically effective amount of a compound is not significantly cytotoxic to normal cells if administration of the compound in a

therapeutically effective amount does not induce cell death in greater than 10% of normal cells. A therapeutically effective amount of a compound does not significantly affect the viability of normal cells if administration of the compound in a therapeutically effective amount does not induce cell death in greater than 10% of normal cells. In an aspect, cell death occurs by apoptosis.

[0291] Contacting a cell with a composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, can induce or activate cell death selectively in cancer cells. Administering to a subject in need thereof a compound of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, can induce or activate cell death selectively in cancer cells. Contacting a cell with a composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, can induce cell death selectively in one or more cells affected by a cell proliferative disorder. Preferably, administering to a subject in need thereof a composition of the present invention, or a pharmaceutically acceptable salt, prodrug, metabolite, polymorph or solvate thereof, induces cell death selectively in one or more cells affected by a cell proliferative disorder.

[0292] The following examples are provided to illustrate certain embodiments of the invention. They are not intended to limit the invention in anyway. Such modifications are intended to fall within the scope of the appended claims.

EXPERIMENTATION AND RESULTS:

[0293] 1. Process for Isolation and Purification of the active constituent from *Vernonia cinerea* (Sahadevi):

[0294] Plant material and Sample Preparation

[0295] Freshly harvested whole plant of Sahadevi was chopped into pieces and thoroughly washed in water followed by grinding into a mixer grinder to make an aqueous suspension. The suspension was filtered through muslin cloth followed by high-speed centrifugation (15,000 x g for 30 minutes) to remove any debris. The aqueous extract was subjected to chloroform treatment (1:1 v/v, 3X) to separate chlorophyll and other organic components. Trace amount of chloroform was removed from the aqueous part by rotary evaporation. The aqueous part was then subjected to ethanol precipitation by treating it with 100% ice-chilled ethanol to precipitate proteins and nucleic acids. The trace amount of ethanol was further removed by rotary evaporation followed by freeze drying the aqueous part in a freeze drier.

The crude powder was stored in an airtight container, at room temperature. 50 g of crude powder of *Vernonia cinerea*. was added to 500 ml of Milli Q water and kept overnight on gentle stirring. The supernatant was decanted and dried in freeze dryer. The water soluble dried powder was used in bioassay to check its activity before proceeding for further downstream purification. The dried power of water extract was further re-suspended in 500ml of Methanol and kept on stirring for two hours at ambient temperature. The methanol extract was further dried. The dried methanol extract was further analysed for the activity using biological assay

[0296] Process Flow Chart

[0297] Purification process flow along with process parameters and role of each step is shown as below.

Process Parameter	Process Step	In-process Tests
• **Resin**: Capto adhere ImpRes • **Bed height**: 15 cm ± 10% • **Column diameter**: 32 mm • **Bed volume**: ~120 mL • **Residence time**: ~ 4 min • **Operational flow rate**: ~30 mL/min • **Equilibration Buffer**: 50 mM Sodium acetete buffer, pH 5.5 • **Elution buffer**50 mM Sodium acetete buffer, 300 mM NaCl pH 5.5 • **Regeneration and sanitization solution**: 1M NaOH, 2M NaCl solution • **Column storage solution**: 20 % Ethanol or 10 % NaOH. • **Elution:** Linear gradient with 0 to 50% of elution buffer • Elution collection: For 2 mm path length of UV flow cell collect elute fraction of five column volumes of 120ml column after post load wash	**Capture Step (Multimodal Chromatography)**	RP-HPLC of the elute fractions
⇩		
• **Resin**: Puritas C18, 100A° • **Bed height**: 25 cm • **Column diameter**: 50mm	**Intermediate Step (Prep-RPHPLC)**	RP-HPLC of the elute fractions and Bioassay

• **Bed volume**: ~480 mL • **Residence time**: ~ 10 min • **Operational flow rate**: ~48 mL/min • **Equilibration Buffer**: 0.1% Acetic acid • **Elution buffer:** 0.1% acetic acid in 85% Acetonitrile • **Regeneration and sanitization solution**: 90% ACN • **Column storage solution**: 60% ACN • **Elution:** Linear gradient with 0 to 100% of elution buffer for 100min • Elution collection: Fractions of 1min each collected after 34min		after drying
⇩		
• **Resin**: Capto adhere ImpRes • **Bed height**: ~5 cm ± 10% • **Column diameter**: 11mm • **Bed volume**: ~5 mL • **Residence time**: ~4 min • **Operational flow rate**: 1.25 mL/min • **Equilibration Buffer**: 50 mM Sodium acetate, pH 5.5 • **Elution buffer**50 mM Sodium acetete buffer, 300 mM NaCl pH 5.5 • **Regeneration and sanitization solution**: 1M NaOH, 2M NaCl solution • **Column storage solution**: 20 % Ethanol or 10 % NaOH. • **Elution:** Linear gradient with 0 to 33 % of elution buffer for 10 CV followed by 33% to 50% of elution buffer for 3CV. • Elution collection: For 2 mm path length of UV flow cell collect elute fraction of 1ml each is collected from 20 mAu ascending to 20 mAu decending	**Polishing Step (Multimodal Chromatography)**	
⇩		

• **Resin**: PharmPrep® P 100 RP-18e (10µm) • **Bed height**: 15 cm • **Column diameter**: 4.6mm • **Bed volume**: ~4.2 mL • **Residence time**: ~ 8 min • **Operational flow rate**: ~0.5 mL/min • **Equilibration Buffer**: 0.1% Acetic acid • **Elution buffer:** 0.1% acetic acid in 85% Acetonitrile • **Regeneration and sanitization solution**: 90% ACN • **Column storage solution**: 60% ACN • **Elution:** Linear gradient with 0 to 15% of elution buffer for 30min followed by 15% to 20% of Elution buffer for 40min Elution collection: Fractions of 0.5 min each collected after 20 min post gradient is strated	**Desalting Step (RP-HPLC)**	RP-HPLC of the elute fractions and Bioassay of pool after drying

[0298] Purification Process Description

[0299] Chromatography 1 (Capture Step)

[0300] The dried methanol extract was subjected to purification with Capto adhere ImpRes resin (120 ml column volume) (Make, GE Healthcare) that has multimodal functionality at the mentioned process parameters. The resin was equilibrated with 50mM Sodium acetate pH 5.5 and the methanol extract was re-suspended in the same equilibration buffer and loaded onto the column at 4 min residence time. The column was washed further to remove the unbound fractions and then eluted with a linear gradient using 50mM Sodium acetate containing 300mM Sodium chloride, pH 5.5. The fractions were collected and analysed with the RP-HPLC and Mass Spectroscopy. The fractions were also analysed for activity with biological assay method.

[0301] Chromatography 2 (Intermediate Step)

[0302] The fractions that contain the bioactivity were further pooled and subjected to purification with RP- HPLC Chromatography. The Puritas C18 (PP18-05-100-250C) Prep Column from Chromachemie was used for Chromatography 2 step purification. The column was equilibrated with 0.1% Acetic acid and the dried powder of Chrom 1 elute pool was re-suspended in the same equilibration buffer and loaded onto the column at 10 min residence time. The column was washed further to remove the unbound fractions and then eluted with a

linear gradient using 0.1% acetic acid in 85% Acetonitrile. The fractions were collected and analysed by the RP-HPLC, Mass Spectroscopy and by using biological assay method.

[0303] Chromatography 3 (Polishing Step)

[0304] Fractions that contain activity were further pooled and subjected to polishing step purification again with Capto adhere ImpRes (5ml column volume) with the same buffer conditions and the linear gradient as mentioned above (Chrom 1 step). The elution fractions were collected and pooled considering the highest purity according to analytical RP-HPLC and then dried.

[0305] Chromatography 4 (Desalting Step)

[0306] The dried elute pool of Chrom 3 was resuspened and subjected to desalting step using the PharmPrep® P 100 RP-18e (10µm) column from Merck. The column was equilibrated with 0.1% Acetic acid and the dried powder of Chrom 3 elute pool was re-suspended in the same equilibration buffer and loaded onto the column at approx. 8 min residence time. The column was washed further to remove the salt fractions and then eluted with a linear gradient using 0.1% acetic acid in 85% Acetonitrile. The fractions were collected and analysed by the RP-HPLC, Mass Spectroscopy and biological assay method.

[0307] Characterization of the active fractions:

[0308] For structural elucidation, the active compound was analysed by elemental analysis, ^{1}H-NMR, ^{13}C- NMR, Distortionless Enhancement by Polarization Transfer (DEPT), Infrared Spectroscopy (IR), LC-MS and X Ray Crystallography.

[0309] Elemental analysis confirmed the elements C, H and O only (C:57.62%, H:4.72%, rest O).The purified compound (E05) was analysed by ^{1}H NMR. The analysis revealed the presence of a benzene ring structure along with 2 OH groups, one COOH group and one methylene group (**Figure 5**).

[0310] The purified compound (E05) was analysed by ^{13}C NMR. The analysis confirmed the presence of 9 different carbon atoms (**Figure 6**).

[0311] DEPT (Distortionless Enhancement by Polarization Transfer) showed the presence of 3 carbon atoms with odd number of hydrogens and 2 carbon atoms with even number of hydrogens (**Figure 7**)

[0312] The pure fraction (E05) was subjected to LC-MS analysis. MS analysis revealed the major peak of 182 Dalton (**Figure 9**).

[0313] X Ray Crystallography:

[0314] A single crystal suitable for single crystal X-ray diffraction analysis was selected using

Leica microscope. A specimen of compound E-05, approximate dimensions 0.238 mm x 0.214 mm x 0.087 mm, was used for the X-ray crystallographic analysis. The X-ray intensity data were measured on a Bruker D8 VENTURE Kappa Duo PHOTON II CPAD diffractometer equipped with Incoatech multilayer mirrors optics. The intensity measurements were carried out with Mo microfocus sealed tube diffraction source (Mo-Kα = 0.71073 Å) at 100(2) K temperature. The X-ray generator was operated at 50 kV and 1.4 mA. A preliminary set of cell constants and an orientation matrix were calculated from three sets of 12 frames. Data were collected with ω and φ scan width of 0.5° at different settings of φ and ω with a frame time of 20 secs keeping the sample-to-detector distance fixed at 4.00 cm. The X-ray data collection was monitored by APEX3 program (Bruker, 2016).1 The total exposure time was 5 hours. The frames were integrated with the Bruker SAINT Software package using a narrow-frame algorithm. All the data were corrected for Lorentzian polarization and absorption effects using SAINT and SADABS programs. ShelX-97 was used for structure solution and full matrix least-squares refinement on F2.2 All the hydrogen atoms were placed in a geometrically idealized positions and constrained to ride on its parent atoms. An ORTEP III3 (**Figure 10**) view of compound was drawn with 50% probability displacement ellipsoids and H atoms are shown as small spheres of arbitrary radii. The single crystal analysis revealed that the unknown compound is 3-(3,4-dihydroxy phenyl) propanoic acid.

Table 1: Sample and crystal data for the purified compound

Identification code	**E-05**	
Chemical formula	$C_9H_{10}O_4$	
Formula weight	182.17 g/mol	
Temperature	100(2) K	
Wavelength	0.71073 Å	
Crystal size	0.087 x 0.214 x 0.238 mm	
Crystal system	monoclinic	
Space group	$P2_1/c$	
Unit cell dimensions	a = 11.3269(9)Å	α = 90°
	b = 5.5745(4) Å	β = 109.283(3)°
	c = 13.8497(9) Å	γ = 90°
Volume	825.43(10) $Å^3$	
Z	4	
Density (calculated)	1.466 g/cm^3	
Absorption coefficient	0.116 mm^{-1}	
F(000)	384	

[0315] 3. *in vitro* biological assay using water soluble extract of Sahadevi, different fractions during purification, pure and synthetic compound, 3-(3,4-dihydroxy phenyl) propanoic acid

[0316] The F-36E cell line (Riken, BRC, RCB0776) which was derived from a patient with erythro-leukaemia, shows complete growth dependency on EPO (Erythropoietin). For the present study, F-36E cells were used for measuring the *in-vitro* activity in the water soluble plant extract, different fractions collected by analytical techniques, purified samples (E05) and with the synthetic compound [3-(3,4-dihydroxy phenyl)propanoic acid]. The cells were grown and maintained in RPMI-1640 complete medium supplemented with 1IU/ml EPO. For assay, F-36E cells were plated at 10,000 cells/well in a 96-well plate. Cells were cultured overnight in RPMI 1640 containing 5% FBS and EPO. Cells were then treated with either water-soluble extract (200ug/well) or with different fractions or purified compound or with synthetic compound for 24-48 h. The cells treated with either EPO (1IU/ml) [EPO control] or without any growth factors (cell control) were used as control. The cellular viability was measured using the alamar blue cell viability reagent. The 96-well plate was read at an excitation

wavelength of 530 nm and emission wavelength of 590 nm. The relative fluorescence units obtained are directly proportional to the number of live cells.

[0317] The anti-proliferative activity of the synthetic compound [3-(3,4-dihydroxy phenyl)propanoic acid] and Bosutinib was also tested using F-36E bioassay. Values obtained from different fractions were normalized to cell control value.

[0318] The anti-proliferative activity of Sahadevi extract was specific to leukaemia cell lines, F-36E and TF-1 (erythro-leukaemia cell line). EPO or GM-CSF treated cells were used as control to normalize the value of E05 treated cells. Chinese hamster ovary cells (CHO), epidermoid carcinoma cells (A431) and the rat medullary thyroid carcinoma cells (6-23) did not show any effect upon extract treatment (**Figure 11**). A dose dependent inhibition of growth of colon cancer cell line HCT116 and breast cancer cell line BT-474 could also be observed upon treatment with E05 (**Figure 12 and Figure 13**).

[0319] The IC_{50} values are comparable between the two molecules (**Figure 14**).

[0320] Kinase inhibitor screening assay

[0321] The kinase selectivity profiling systems from Promega was used to check the inhibitory activity of the purified compound against the broad panel of Tyrosine kinases. The kinase selectivity assays were assembled in a 384-well plate using 1µl of the purified compound (1µM final concentration), 2µl of each kinase working stock and 2µl of the corresponding ATP/Substrate working stock as per the manufacturer protocol and kinase activity was quantified using the ADP-Glo™ kinase assay from Promega. The reaction assembled with kinase and corresponding ATP/Substrate without the purified compound was used as a positive control. Out of 16 tyrosine kinases, SRC kinase activity was found to be inhibited by the purified compound isolated from Sahadevi extract.

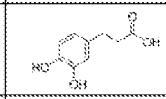
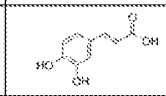
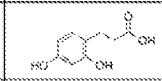
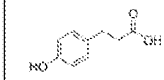
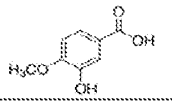
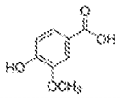
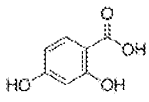
[0322] *in vitro* bioassay using triple negative breast cancer cell line

[0323] MDA-MB-468 cells, isolated from a pleural effusion of a female patient with metastatic adenocarcinoma of the breast, is a triple negative breast cancer (TNBC) cell line. The IC 50 value of E05 or lapatinib (Sigma) or the structural analogues of E05 were tested in this cell line.

[0324] Briefly, MDA-MB-468 cells (ATCC# HTB-32) were seeded with a density of 5,000 cells per well in a 96 well white plate (Costar Cat#3917) in DMEM media supplemented with 10% FBS (assay medium). The plates were incubated at 37°C, 5%CO2 for 24hr. E05 and analogues were diluted in the assay medium and added to the respective wells of the assay plate. The plates were further incubated at 37°C, 5%CO2 for 24hrs. After the incubation, the

cell viability was assessed using CellTiter-Glo® and the plates were read for luminescence using Cytation 5 (Biotek). The relative luminescence unit (RLU) obtained was plotted against the concentration and EC50 values were estimated using Graphpad Prism 5 software (**Figure 15 and Table 2**).

Table 2: Analysis of different structural analogues of E05 in MDA-MB-468 cells. The activity of the E05 and the structural analogues were tested in the TNBC cell line. E05 and E05_A and E05_E showed the cell killing activity in the TNBC cell line

Compound	Structure	Sample-ID	Activity (EC50)
3-(3,4-Dihydroxyphenyl) propanoic acid		E05	86.13 µM
3-(3,4-Dihydroxyphenyl)-2-propionic acid (Caffeic acid)		E05_A	122.1 µM
3-(2,4-Hydroxyphenyl)propionic acid		E05_B	Inactive
3-(3-Hydroxyphenyl)propionic acid		E05_C	Inactive
3-(4-Hydroxyphenyl)propionic acid (Phloretic acid)		E05_D	Inactive
1,2-Dihydroxybenzene (Catechol)		E05_E	102.7 µM
3-Hydroxy-4-methoxybenzoic acid (Isovanillic acid)		E05_F	Inactive
4-Hydroxy-3-methoxybenzoic acid (Vanillic acid)		E05_G	Inactive
2,4-Dihydroxybenzoic acid		E05_H	Inactive

[0325] Anti-proliferative activity of synthetic E05 was compared with that of lapatinib in triple-negative breast cancer cell line, MDA-MB-468. MDA-MB-468 cells (ATCC# HTB-32) were seeded with a density of 5,000 cells per well in a 96 well white plate (Costar Cat#3917) in DMEM media supplemented with 10% FBS (assay medium). The plates were incubated at

37°C, 5% CO_2 for 24hr. E05 and Lapatinib was diluted in the assay medium and added to the respective wells of the assay plates. The plates were further incubated at 37°C, 5%CO2 for 8hrs. After the incubation, the cell viability was assessed using CellTiter-Glo® and the plates were read for luminescence using Cytation 5 (Biotek). The relative luminescence unit (RLU) obtained was plotted against the concentration and EC50 values were estimated using Graphpad Prism 5 software.

[0326] E05 showed stronger inhibition in compare to lapatinib within 8 hrs of incubation (**Figure 16**).

[0327] Xenograft study of E05 in SCID mice

[0328] Efficacy study of E05 and positive control (5-Fluorouracil) were carried out in a murine xenograft model of Triple-Negative Breast Cancer (MDA-MB-468). Dosing was done as per the Table. 3.

Table 3: Dosing pattern in different groups during xenograft study

Groups	Group ID	Treatment*	No of mice /group
1	Vehicle Control	Vehicle, oral, twice daily, for 6 weeks	10
2	Positive control (5-Fluorouracil)	i.p., 10 mg/kg, q2days, for 6 weeks	10
3	E05	Oral, twice daily (q12h), for 6 weeks (75 mg/kg)	10
4	E05	Oral, twice daily(q12h), for 6 weeks (200 mg/kg)	10
5	E05	Oral, twice daily (q12h), for 6 weeks (500 mg/kg)	10

[0329] Female SCID (Severe Combined Immunodeficient) mice were used for this study.

[0330] Approximately 5×10^6 cells in 0.2 mL FBS free medium containing 50% of Matrigel was injected into sub-cutaneous tissue on right flank of each mouse. Treatment with E05 or with positive control was initiated when average tumor volume reaches ~ 100 mm3. Tumor volume was measured periodically. Dose dependent decrease in tumor volumes were observed for the test compound. The E05 showed statistically significant efficacy in the murine xenograft model of triple-negative breast cancer (TNBC) (**Figure 17**).

[0331] Animal toxicological study.

[0332] Repeated dose 7-day dose range finding study (DRF) of E05 through intravenous bolus injection was carried out in Wistar rats. E05 did not produce any systemic toxicity up to the dose level of 500 mg/kg b. wt. when administered daily by intravenous bolus injection for 7

consecutive days.

TABLE 4: Mortality and Morbidity Record of the study Repeated Dose 7-Day Dose Range Finding Study of E05 through Intravenous Bolus Injection in Wistar Rats

Sex and Group N°	Total N° of Rats	Morbidity		Mortality	
		N°	%	N°	%
Male					
G1	5	0	0	0	0
G2	5	0	0	0	0
G3	5	0	0	0	0
G4	5	0	0	0	0
Female					
G1	5	0	0	0	0
G2	5	0	0	0	0
G3	5	0	0	0	0
G4	5	0	0	0	0

Dose: G1 - 0; G2 - 125; G3 - 250; G4 - 500 mg/kg b. wt

[0333] Repeated dose 7-day dose range finding study (DRF) of E05 through oral gavage was carried out in Wistar rats. In this DRF study, no mortality, morbidity or clinical sign was observed during the treatment period. No treatment-related changes were observed in food consumption, organ weight and relative organ weight of male and female rats from treatment groups. Based on the results, it is concluded that E05 did not produce any toxicity up to the dose level of 1000 mg/kg b. wt. when administered orally, through gavage, for 7 consecutive days in Wistar rats.

TABLE 5: Mortality and Morbidity Record of the study Repeated Dose 7-Day Dose Range Finding Study of E05 through oral gavage in Wistar Rats

Sex and Group N°	Total N° of Rats	Morbidity		Mortality	
		N°	%	N°	%
Male					
G1	5	0	0	0	0
G2	5	0	0	0	0
G3	5	0	0	0	0
G4	5	0	0	0	0

Female					
G1	5	0	0	0	0
G2	5	0	0	0	0
G3	5	0	0	0	0
G4	5	0	0	0	0

Dose: G1 - 0; G2 - 250; G3 - 500; G4 - 1000 mg/kg b. wt.

[0334] A maximum tolerated dose (MTD) study was carried out with E05. In MTD study, group of 3 male and 3 female rats were dosed at 175 (group 1), 550 (group 2), 1750 (group 3) and 2000 (group 4) mg/kg b. wt. In MTD, no mortality, morbidity, or clinical sign was observed during the 72-h observation period after single administration of 175, 550, 1750, and 2000 mg/kg b. wt. of the test item, E05.

TABLE 6: Mortality and Morbidity Record of the of Maximum Tolerated Dose Study of E05 through oral gavage in Wistar Rats

Sex and Group N°	**Dose (mg/kg b. wt.)**	**Number of Rat**	**Mortality on Experimental Day**				**Morbidity on Experimental Day**			
			1	**2**	**3**	**4**	**1**	**2**	**3**	**4**
Male										
G1	175	3	0	0	0	0	0	0	0	0
G2	550	3	0	0	0	0	0	0	0	0
G3	1750	3	0	0	0	0	0	0	0	0
G4	2000	3	0	0	0	0	0	0	0	0
Female										
G1	175	3	0	0	0	0	0	0	0	0
G2	550	3	0	0	0	0	0	0	0	0
G3	1750	3	0	0	0	0	0	0	0	0
G4	2000	3	0	0	0	0	0	0	0	0

[0335] Pharmacokinetic study of E05 in male Sprague Dawley rats

[0336] A study to determine the pharmacokinetics of E05 after single oral (10 mg/kg) administration in male Sprague Dawley rats was carried out. Following single oral gavage administration of E05 dose formulation to male Sprague Dawley rats (Dose: 10 mg/kg), mean time to reach peak plasma concentration (Tmax) was found to be 0.25 h, suggesting rapid rate of absorption. The exposure (Cmax and AUClast) was found to be 306 ng/mL and 151 ng.h/mL, respectively. The absolute oral bioavailability of Small molecule was 80% (Figure 18).

[0337] The patent and scientific literature referred to herein establishes the knowledge that is available to those with skill in the art. All United States patents and published or unpublished United States patent applications cited herein are incorporated by reference. All published foreign patents and patent applications cited herein are hereby incorporated by reference. All other published references, documents, manuscripts and scientific literature cited herein are hereby incorporated by reference.

[0338] While this invention has been particularly shown and described with references to preferred embodiments thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the scope of the invention encompassed by the appended claims.