CLAIMS

WHAT IS CLAIMED IS:

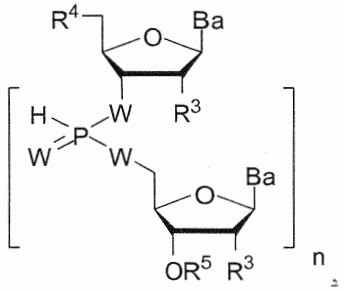
1. A process for the preparation of phosphorothiotriesters of structure IIIa comprising the steps of:

i) reacting an H-phosphonate of structure Ia with ~~an~~ silylating reagent to provide a silyloxyphosphonate; and

ii) reacting the silyloxyphosphonate with a thiosulfonate reagent of structure IIa to provide a phosphorothiotriester of structure IIIa;

wherein,

the H-phosphonate of structure Ia has the following structure:



Structure Ia

wherein~~,~~:

W is ~~independently selected from O, S, NH, or CH_2~~;

R^3 is -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-P(O)(R^e)_2$, $-HP(O)(R^e)$, $-OR^a$ or $-SR^c$;

Y^1 is O, NR^d, S, or Se;

R^a is a blocking group;

R^c is a blocking group;

each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, acyl, substituted silyl, carbamate, $-P(O)(R^e)_2$, or $-HP(O)(R^e)$;

each instance of R^e is independently hydrogen, alkyl, aryl, alkenyl, alkynyl, alkyl-Y^2-, alkenyl-Y^2-, alkynyl-Y^2-, aryl-Y^2-, or heteroaryl-Y^2-, or a cation which is Na^{+1}, Li^{+1}, or K^{+1};

Y^2 is O, NR^d, or S;

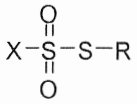
each instance of R^4 is independently hydrogen, -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, -OR^b ,or -SR^c, and R^b is a blocking group;

each instance of Ba is independently a blocked or unblocked adenine, cytosine, guanine, thymine, uracil or modified nucleobase;

R^5 is hydrogen, a blocking group, a linking moiety connected to a solid support or a linking moiety connected to a nucleic acid; and

n is between 1 and about 200; and

the thiosulfonate reagent of structure IIa has the following structure:



Structure IIa

wherein:

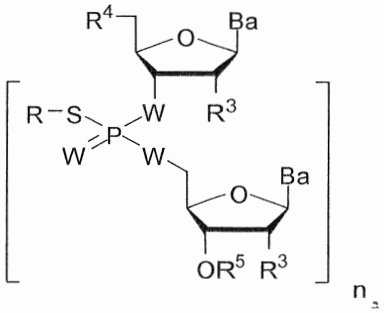
X is alkyl, cycloalkyl, or heteroaryl;

R is alkyl, alkenyl, alkynyl, cycloalkyl, aryl, aralkyl, heteroaryl, heteroaralkyl, or R^1-R^2;

R^1 is selected from -S-alkenylene-, -S-alkylene-, -S-alkylene-aryl-alkylene-, -S-CO-aryl-alkylene-, or -S-CO-alkylene-aryl-alkylene-;

R^2 is selected from heterocyclo-alkylene-S-, heterocyclo-alkenylene-S-,aminoalkyl-S-,or $(alkyl)_4N$-alkylene-S-;

and the phosphorothiotriester of structure IIIa has the following structure:



Structure IIIa

wherein:

W is ~~independently selected from O, S, NH, or CH_2~~;

R is alkyl, alkenyl, alkynyl, cycloalkyl, aryl, aralkyl, heteroaryl, heteroaralkyl, or R^1-R^2;

R^1 is selected from -S-alkenylene-, -S-alkylene-, -S-alkylene-aryl-alkylene-, -S-CO-aryl-alkylene-, or -S-CO-alkylene-aryl-alkylene-;

R^2 is selected from heterocyclo-alkylene-S-, heterocyclo-alkenylene-S-,aminoalkyl-S-,or $(alkyl)_4N$-alkylene-S-;

R^3 is -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-P(O)(R^e)_2$, $-HP(O)(R^e)$, $-OR^a$ or $-SR^c$;

Y^1 is O, NR^d, S, or Se;

R^a is a blocking group;

R^c is a blocking group;

each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, acyl, substituted silyl, carbamate, $-P(O)(R^e)_2$, or $-HP(O)(R^e)$;

each instance of R^e is independently hydrogen, alkyl, aryl, alkenyl, alkynyl, alkyl-Y^2-, alkenyl-Y^2-, alkynyl-Y^2-, aryl-Y^2-, or heteroaryl-Y^2-, or a cation which is Na^{+1}, Li^{+1}, or K^{+1};

Y^2 is O, NR^d, or S;

each instance of R^4 is independently hydrogen, -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-OR^b$,or $-SR^c$, and R^b is a blocking group;

each instance of Ba is independently a blocked or unblocked adenine, cytosine, guanine, thymine, uracil or modified nucleobase;

R^5 is hydrogen, a blocking group, a linking moiety connected to a solid support or a linking moiety connected to a nucleic acid; and

n is between 1 and about 200.

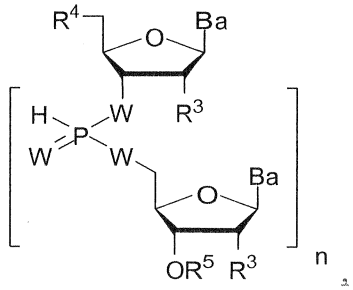
2. A process for the preparation of phosphorothiotriesters comprising non-stereorandom phosphorous linkages of structure IIIb comprising the steps of:

i) reacting an H-phosphonate comprising non-stereorandom phosphorous linkages of structure Ib with ~~an~~ a silylating reagent to provide a silyloxyphosphonate; and

ii) reacting the silyloxyphosphonate with a thiosulfonate reagent of structure IIb to provide a phosphorothiotriester comprising non-stereorandom phosphorous linkages of structure IIIb;

wherein,

the H-phosphonate comprising non-stereorandom phosphorous linkages of structure Ib has the following structure:



Structure Ib

wherein:

W is ~~independently selected from O, NH, or CH_2~~;

R^3 is -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-P(O)(R^e)_2$, $-HP(O)(R^e)$, $-OR^a$ or $-SR^c$;

Y^1 is O, NR^d, S, or Se;

R^a is a blocking group;

R^c is a blocking group;

each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, acyl, substituted silyl, carbamate, $-P(O)(R^e)_2$, or $-HP(O)(R^e)$;

each instance of R^e is independently hydrogen, alkyl, aryl, alkenyl, alkynyl, alkyl-Y^2-, alkenyl-Y^2-, alkynyl-Y^2-, aryl-Y^2-, or heteroaryl-Y^2-, or a cation which is Na^{+1}, Li^{+1}, or K^{+1};

Y^2 is O, NR^d, or S;

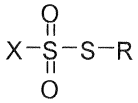
each instance of R^4 is independently hydrogen, -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-OR^b$,or $-SR^c$, and R^b is a blocking group;

each instance of Ba is independently a blocked or unblocked adenine, cytosine, guanine, thymine, uracil or modified nucleobase;

R^5 is hydrogen, a blocking group, a linking moiety connected to a solid support or a linking moiety connected to a nucleic acid; and

n is between 1 and about 200; and

the thiosulfonate reagent of structure IIb has the following structure:



Structure IIb

wherein:

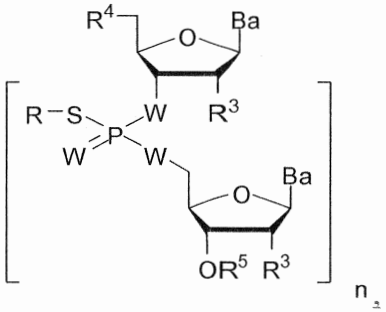
X is alkyl, cycloalkyl, aryl, or heteroaryl;

R is alkyl, alkenyl, alkynyl, cycloalkyl, aryl, aralkyl, heteroaryl, heteroaralkyl, or R^1-R^2;

R^1 is selected from -S-alkenylene-, -S-alkylene-, -S-alkylene-aryl-alkylene-, -S-CO-aryl-alkylene-, or -S-CO-alkylene-aryl-alkylene-;

R^2 is selected from heterocyclo-alkylene-S-, heterocyclo-alkenylene-S-,aminoalkyl-S-,or $(alkyl)_4N$-alkylene-S-;

and the chiral phosphorothiotriester comprising non-stereorandom phosphorous linkages of structure IIIb has the following structure:



Structure IIIb

wherein:

W is ~~independently selected from O, NH, or CH_2~~;

R is alkyl, alkenyl, alkynyl, cycloalkyl, aryl, aralkyl, heteroaryl, heteroaralkyl, or R^1-R^2;

R^1 is selected from -S-alkenylene-, -S-alkylene-, -S-alkylene-aryl-alkylene-, -S-CO-aryl-alkylene-, or -S-CO-alkylene-aryl-alkylene-;

R^2 is selected from heterocyclo-alkylene-S-, heterocyclo-alkenylene-S-,aminoalkyl-S-,or $(alkyl)_4N$-alkylene-S-;

R^3 is -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-P(O)(R^e)_2$, $-HP(O)(R^e)$, $-OR^a$ or $-SR^c$;

Y^1 is O, NR^d, S, or Se;

R^a is a blocking group;

R^c is a blocking group;

each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, acyl, substituted silyl, carbamate, $-P(O)(R^e)_2$, or $-HP(O)(R^e)$;

each instance of R^e is independently hydrogen, alkyl, aryl, alkenyl, alkynyl, alkyl-Y^2-, alkenyl-Y^2-, alkynyl-Y^2-, aryl-Y^2-, or heteroaryl-Y^2-, or a cation which is Na^{+1}, Li^{+1}, or K^{+1};

Y^2 is O, NR^d, or S;

each instance of R^4 is independently hydrogen, -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-OR^b$,or $-SR^c$, and R^b is a blocking group;

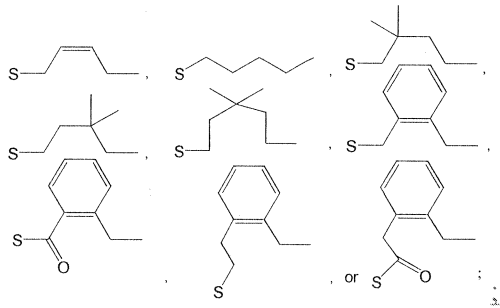
each instance of Ba is independently a blocked or unblocked adenine, cytosine, guanine, thymine, uracil or modified nucleobase;

R^5 is hydrogen, a blocking group, a linking moiety connected to a solid support or a linking moiety connected to a nucleic acid; and

n is between 1 and about 200.

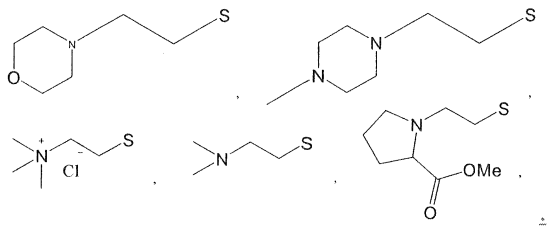
~~3. The process of claim 1 or 2, wherein W is O.~~

43. The process of claim 1 or 2, wherein R^1 is selected from:



; and

R^2 is selected from:



54. The process of claim 1 or 2, wherein the silylating reagent is selected from

1,1,3,3-tetramethyl-1,3-diphenyldisilazane;

1,3-dimethyl-1,1,3,3-tetraphenyldisilazane;

1-(trimethylsilyl)imidazole;

N-trimethylsilyl-N-methyl trifluoroacetamide;

bis(dimethylamino)dimethylsilane;

bromotrimethylsilane;

chlorodimethyl(pentafluorophenyl)silane;

chlorotriethylsilane;

chlorotriisopropylsilane;

chlorotrimethylsilane;
dichlorodimethylsilane;
hexamethyldisilazane;
N,N′-bis(trimethylsilyl)urea;
N,N-bis(trimethylsilyl)methylamine;
N,N-dimethyltrimethylsilylamine;
N,O-bis(trimethylsilyl)acetamide;
N,O-bis(trimethylsilyl)carbamate;
N,O-bis(trimethylsilyl)trifluoroacetamide;
N-methyl-N-(trimethylsilyl)trifluoroacetamide;
N-methyl-N-trimethylsilylacetamide;
N-methyl-N-trimethylsilylheptafluorobutyramide;
N-tert-butyldimethylsilyl-N-methyltrifluoroacetamide;
N-methyl-N-trimethylsilylheptafluorobutyramide;
trimethylsilyltriflate;
triethylsilyltriflate;
triisopropylsilyltriflate; ~~or~~ and
tert-butyldimethylsilyltriflate.

~~6~~5. The process of claim ~~5~~4, wherein the silylating reagent is selected from N,O-bis(trimethylsilyl)trifluoroacetamide, trimethylsilyltriflate, chlorotrimethylsilane, ~~or~~ and 1-(trimethylsilyl)imidazole.

~~7~~6. The process of claim ~~6~~5, wherein the silylating reagent is ~~selected from~~ N,O-bis(trimethylsilyl)trifluoroacetamide.

~~8~~7. The process of claim 1 or 2, wherein the H-phosphonate is covalently linked to a solid phase.

~~9~~8. A process for the preparation of phosphorothiotriesters of structure IIIc comprising the steps of:

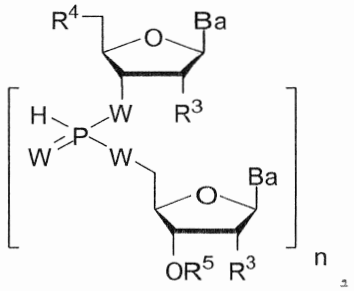
i) reacting an H-phosphonate of structure Ic with ~~a~~an silylating reagent to provide a silyloxyphosphonate;

ii) reacting the silyloxyphosphonate with a bis(thiosulfonate) reagent of structure IVc to provide a phosphorothiotriester comprising a thiosulfonate group ~~of structure Vc~~;

iii) reacting the phosphorothiotriester comprising a thiosulfonate group ~~of structure Vc~~ with a nucleophile of structure VIc to provide the phosphorothiotriesters of structure IIIc;

wherein:

the H-phosphonate of structure Ic has the following structure:



Structure Ic

wherein:

W is ~~independently selected from O, S, NH, or CH_2~~;

R^3 is -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $P(O)(R^e)_2$, $-HP(O)(R^e)$, $-OR^a$ or $-SR^c$;

Y^1 is O, NR^d, S, or Se;

R^a is a blocking group;

R^c is a blocking group;

each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, acyl, substituted silyl, carbamate, $-P(O)(R^e)_2$, or $-HP(O)(R^e)$;

each instance of R^e is independently hydrogen, alkyl, aryl, alkenyl, alkynyl, alkyl-Y^2-, alkenyl-Y^2-, alkynyl-Y^2-, aryl-Y^2-, or heteroaryl-Y^2-, or a cation which is Na^{+1}, Li^{+1}, or K^{+1};

Y^2 is O, NR^d, or S;

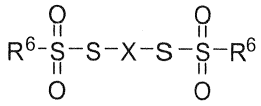
each instance of R^4 is independently hydrogen, -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-OR^b$,or $-SR^c$, and R^b is a blocking group;

each instance of Ba is independently a blocked or unblocked adenine, cytosine, guanine, thymine, uracil or modified nucleobase;

R^5 is hydrogen, a blocking group, a linking moiety connected to a solid support or a linking moiety connected to a nucleic acid; and

n is between 1 and about 200; and

the bis(thiosulfonate) reagent of structure IVc has the following structure:



Structure IVc

wherein:

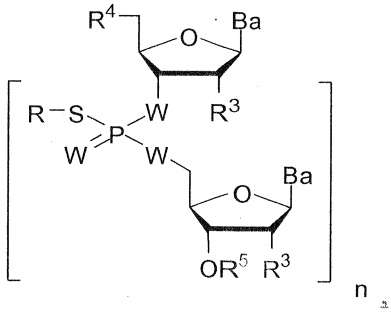
X is alkylene, alkenylene, arylene, or heteroarylene;

each R^6 is independently alkyl, cycloalkyl, aryl, or heteroaryl;

the nucleophile of structure VIc has the following structure:

R^7-SH, wherein R^7 is selected from alkyl, alkenyl, aryl, heterocyclo, aminoalkyl, or (heterocyclo)alkyl;

and phosphorothiotriesters of structure IIIc has the following structure:



Structure IIIc

wherein:

W is ~~independently selected from O, S, NH, or CH_2~~;

R is R^7-S-S-X-

R^7 is alkyl, alkenyl, aryl, heterocyclo, aminoalkyl, or (heterocyclo)alkyl;

X is alkylene, alkenylene, arylene, or heteroarylene;

R^3 is -OH, -SH, -NR^dR^d, -N_3, halogen, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, -$P(O)(R^e)_2$, -$HP(O)(R^e)$, -OR^a or -SR^c;

Y^1 is O, NR^d, S, or Se;

R^a is a blocking group;

R^c is a blocking group;

each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, acyl, substituted silyl, carbamate, -$P(O)(R^e)_2$, or -$HP(O)(R^e)$;

each instance of R^e is independently hydrogen, alkyl, aryl, alkenyl, alkynyl, alkyl-Y^2-, alkenyl-Y^2-, alkynyl-Y^2-, aryl-Y^2-, or heteroaryl-Y^2-, or a cation which is Na^{+1}, Li^{+1}, or K^{+1};

Y^2 is O, NR^d, or S;

each instance of R^4 is independently hydrogen, -OH, -SH, $-NR^dR^d$, $-N_3$, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1-, alkenyl-Y^1-, alkynyl-Y^1-, aryl-Y^1-, heteroaryl-Y^1-, $-OR^b$,or $-SR^c$, and R^b is a blocking group;

each instance of Ba is independently a blocked or unblocked adenine, cytosine, guanine, thymine, uracil or modified nucleobase;

R^5 is hydrogen, a blocking group, a linking moiety connected to a solid support or a linking moiety connected to a nucleic acid;

n is between 1 and about 200; and

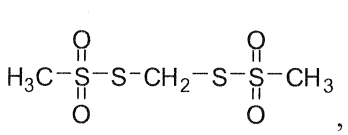
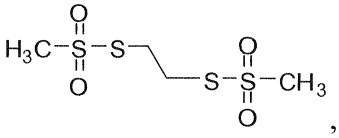
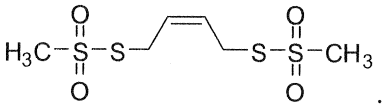
wherein the phosphorous linkages of the H-phosphonate of structure Ic, the phosphorothiotriester comprising a thiosulfonate group ~~of structure Ve~~, and the phosphorothiotriesters of structure IIIc may optionally comprise non-stereorandom phosphorous linkages.

~~109~~. The process of claim ~~8~~7, wherein the phosphorothiotriesters of structure IIIb comprise non-stereorandom phosphorous linkages and the H-phosphonate of structure ~~Ic~~ Ia or Ib comprise non-stereorandom phosphorous linkages~~; and W is independently selected from O, NH, or CH_2~~.

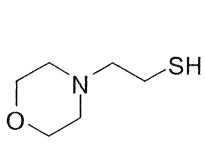
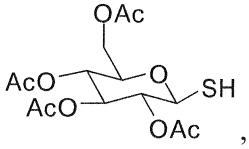
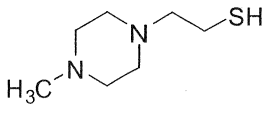
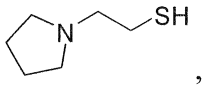
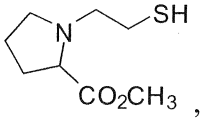
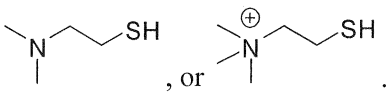
~~11. The process of claim 9 or 10, wherein W is O.~~

~~12~~10. The process of claim ~~9~~8, wherein R^6 is methyl.

~~13~~11. The process of claim ~~9~~8, wherein bis(thiosulfonate) reagent of structure IVc is selected from:


, 
, ~~or~~ and 
.

~~14~~12. The process of claim ~~9~~8, wherein the nucleophile of structure VIc has the following structure:


, 
, 
, 
, 
, 
.

~~15~~13. The process of claim ~~9~~8, wherein the silylating reagent is selected from 1,1,3,3-tetramethyl-1,3-diphenyldisilazane;

1,3-dimethyl-1,1,3,3-tetraphenyldisilazane;
1-(trimethylsilyl)imidazole;
N-trimethylsilyl-N-methyl trifluoroacetamide;
bis(dimethylamino)dimethylsilane;
bromotrimethylsilane;
chlorodimethyl(pentafluorophenyl)silane;
chlorotriethylsilane;
chlorotriisopropylsilane;
chlorotrimethylsilane;
dichlorodimethylsilane;
hexamethyldisilazane;
N,N′-bis(trimethylsilyl)urea;
N,N-bis(trimethylsilyl)methylamine;
N,N-dimethyltrimethylsilylamine;
N,O-bis(trimethylsilyl)acetamide;
N,O-bis(trimethylsilyl)carbamate;
N,O-bis(trimethylsilyl)trifluoroacetamide;
N-methyl-N-(trimethylsilyl)trifluoroacetamide;
N-methyl-N-trimethylsilylacetamide;
N-methyl-N-trimethylsilylheptafluorobutyramide;
N-tert-butyldimethylsilyl-N-methyltrifluoroacetamide;
N-methyl-N-trimethylsilylheptafluorobutyramide;
trimethylsilyltriflate;
triethylsilyltriflate;
triisopropylsilyltriflate; ~~or~~ and
tert-butyldimethylsilyltriflate.

~~16~~14. The process of claim ~~15~~13, wherein the silylating reagent is selected from N,O-bis(trimethylsilyl)trifluoroacetamide, trimethylsilyltriflate, chlorotrimethylsilane, ~~or~~ and 1-(trimethylsilyl)imidazole.

~~17~~15. The process of claim ~~16~~14, wherein the silylating reagent is ~~selected from~~ N,O-bis(trimethylsilyl)trifluoroacetamide.

~~18~~16. The process of claim ~~17~~15, wherein the H-phosphonate is covalently linked to a solid phase.

17. The process of any one of the preceding claims, wherein Y^1 is O.

18. The process of any one of the preceding claims, wherein:

each instance of R^3 is independently hydrogen, –OH, halogen, alkyl, alkenyl, alkynyl, alkyl-Y^1–, alkenyl-Y^1–, alkynyl-Y^1–, aryl-Y^1–, heteroaryl-Y^1–, or $-OR^a$;

each instance of R^4 is independently –OH, hydrogen, alkyl, alkenyl, alkynyl, alkyl-Y^1–, alkenyl-Y^1–, alkynyl-Y^1–, aryl-Y^1–, heteroaryl-Y^1–, or $-OR^b$; and

R^5 is hydrogen or a blocking group.

19. The process of any one of the preceding claims, wherein each instance of R^d is independently hydrogen, alkyl, alkenyl, alkynyl, aryl, or acyl.

20. The process of any one of the preceding claims, wherein Y^2 is O or S.