US 20210401859A1

(19) **United States**

(12) **Patent Application Publication**

Hodgson et al.

(10) **Pub. No.: US 2021/0401859 A1**

(43) **Pub. Date: Dec. 30, 2021**

(54) **METHODS OF TREATING CANCER IN BIOMARKER-IDENTIFIED PATIENTS WITH NON-COVALENT INHIBITORS OF CYCLIN-DEPENDENT KINASE 7 (CDK7)**

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(21) Appl. No.: **17/289,873**

(22) PCT Filed: **Nov. 1, 2019**

(86) PCT No.: **PCT/US19/59535**

§ 371 (c)(1),
(2) Date: **Apr. 29, 2021**

Related U.S. Application Data

(60) Provisional application No. 62/754,398, filed on Nov. 1, 2018, provisional application No. 62/877,189, filed on Jul. 22, 2019, provisional application No. 62/915,983, filed on Oct. 16, 2019, provisional application No. 62/927,469, filed on Oct. 29, 2019.

Publication Classification

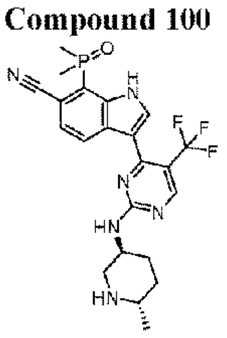
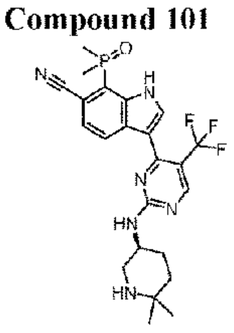
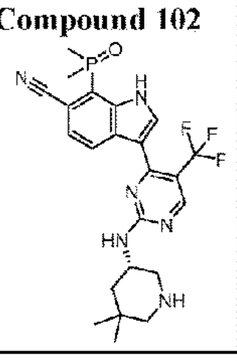
(51) **Int. Cl.**
A61K 31/675 (2006.01)
A61P 35/00 (2006.01)

(52) **U.S. Cl.**
CPC ***A61K 31/675*** (2013.01); ***A61P 35/00*** (2018.01)

(57) **ABSTRACT**

The present invention relates to methods of identifying patients suffering from various types of cancer who are more likely to respond to treatment with a CDK7 inhibitor conforming to structural Formula (I), (Ia), a species thereof, or a specified form thereof (as described herein), either when administered or used alone or in combination with a second therapeutic agent (e.g., another anti-cancer therapy). Patients are identified based on one or more features (e.g., gene copy number or expression level) of certain biomarkers (e.g., RB1 or another member of the E2F pathway). In addition, the present invention relates to methods of treating an identified patient with a compound conforming to structural Formula (I), (Ia), a species thereof, or a specified form thereof, either alone or in combination with a second therapeutic agent. In another aspect, the present invention features kits including instructions for treating a patient identified as described herein.

Specification includes a Sequence Listing.

Compound 100 	CDK7 K_D (nM) = 0.057	
	CDK12 K_i (nM) 230	(K_i /$K_{D\ CDK7}$) *4100*
	CDK2 K_i (nM) 1500	(K_i /$K_{D\ CDK7}$) *26000*
	CDK9 K_i (nM) 800	(K_i /$K_{D\ CDK7}$) *14000*
Compound 101 	CDK7 K_D (nM) =0.065	
	CDK12 K_i (nM) 870	(K_i /$K_{D\ CDK7}$) *13000*
	CDK2 K_i (nM) 2600	(K_i /$K_{D\ CDK7}$) *40000*
	CDK9 K_i (nM) 960	(K_i /$K_{D\ CDK7}$) *15000*
Compound 102 	CDK7 K_D (nM) = 0.059	
	CDK12 K_i (nM) 78	(K_i /$K_{D\ CDK7}$) *1300*
	CDK2 K_i (nM) 390	(K_i /$K_{D\ CDK7}$) *6800*
	CDK9 K_i (nM) 290	(K_i /$K_{D\ CDK7}$) *4900*

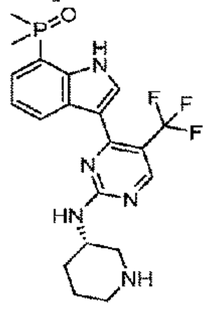
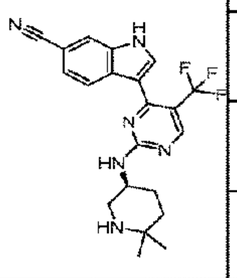
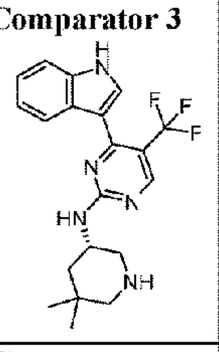
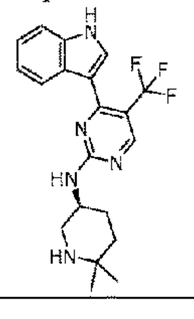
Comparator 1 	CDK7 K_D (nM) = 0.18	
	CDK12 K_i (nM) 51	(K_i /$K_{D\ CDK7}$) *280*
	CDK2 K_i (nM) 210	(K_i /$K_{D\ CDK7}$) *1100*
	CDK9 K_i (nM) 140	(K_i /$K_{D\ CDK7}$) *790*
Comparator 2 	CDK7 K_D (nM) = 0.45	
	CDK12 K_i (nM) 140	(K_i /$K_{D\ CDK7}$) *320*
	CDK2 K_i (nM) 810	(K_i /$K_{D\ CDK7}$) *1800*
	CDK9 K_i (nM) 130	(K_i /$K_{D\ CDK7}$) *280*
Comparator 3 	CDK7 K_D (nM) = 0.21	
	CDK12 K_i (nM) 18	(K_i /$K_{D\ CDK7}$) *89*
	CDK2 K_i (nM) 26	(K_i /$K_{D\ CDK7}$) *130*
	CDK9 K_i (nM) 20	(K_i /$K_{D\ CDK7}$) *99*
Comparator 4 	CDK7 K_D (nM) = 0.34	
	CDK12 K_i (nM) 48	(K_i /$K_{D\ CDK7}$) *140*
	CDK2 K_i (nM) 64	(K_i /$K_{D\ CDK7}$) *190*
	CDK9 K_i (nM) 36	(K_i /$K_{D\ CDK7}$) *100*

FIG. 1

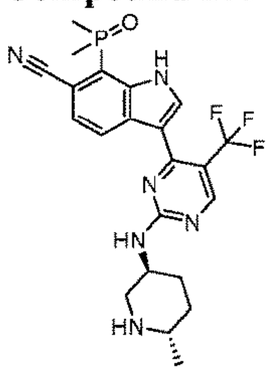
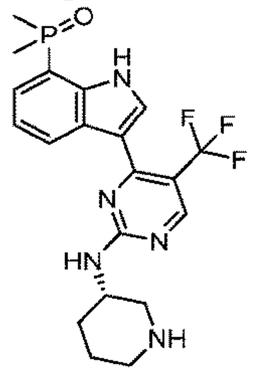
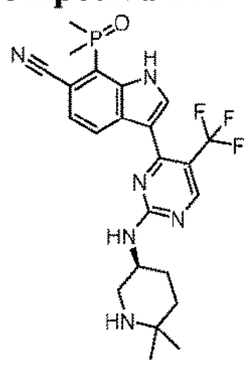
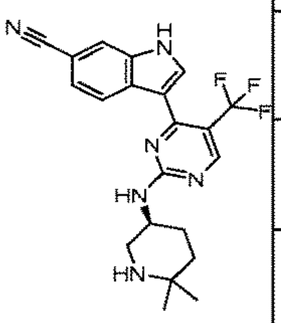
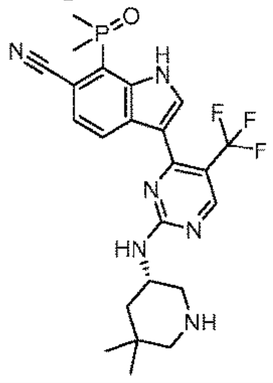
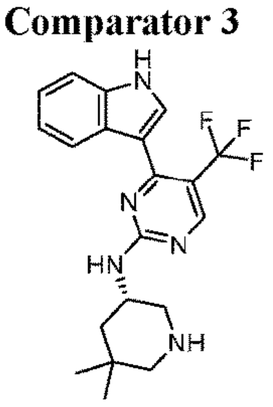
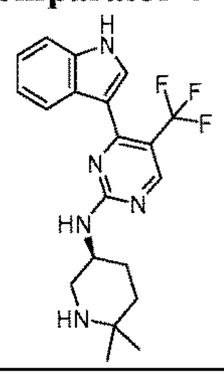
Compound 100 	CDK7 K_D (nM) = 0.057		**Comparator 1** 	CDK7 K_D (nM) = 0.18	
	CDK12 K_i (nM) 230	(K_i /$K_{D\ CDK7}$) *4100*		CDK12 K_i (nM) 51	(K_i /$K_{D\ CDK7}$) *280*
	CDK2 K_i (nM) 1500	(K_i /$K_{D\ CDK7}$) *26000*		CDK2 K_i (nM) 210	(K_i /$K_{D\ CDK7}$) *1100*
	CDK9 K_i (nM) 800	(K_i /$K_{D\ CDK7}$) *14000*		CDK9 K_i (nM) 140	(K_i /$K_{D\ CDK7}$) *790*
Compound 101 	CDK7 K_D (nM) =0.065		**Comparator 2** 	CDK7 K_D (nM) = 0.45	
	CDK12 K_i (nM) 870	(K_i /$K_{D\ CDK7}$) *13000*		CDK12 K_i (nM) 140	(K_i /$K_{D\ CDK7}$) *320*
	CDK2 K_i (nM) 2600	(K_i /$K_{D\ CDK7}$) *40000*		CDK2 K_i (nM) 810	(K_i /$K_{D\ CDK7}$) *1800*
	CDK9 K_i (nM) 960	(K_i /$K_{D\ CDK7}$) *15000*		CDK9 K_i (nM) 130	(K_i /$K_{D\ CDK7}$) *280*
Compound 102 	CDK7 K_D (nM) = 0.059		**Comparator 3** 	CDK7 K_D (nM) = 0.21	
	CDK12 K_i (nM) 78	(K_i /$K_{D\ CDK7}$) *1300*		CDK12 K_i (nM) 18	(K_i /$K_{D\ CDK7}$) *89*
	CDK2 K_i (nM) 390	(K_i /$K_{D\ CDK7}$) *6800*		CDK2 K_i (nM) 26	(K_i /$K_{D\ CDK7}$) *130*
	CDK9 K_i (nM) 290	(K_i /$K_{D\ CDK7}$) *4900*		CDK9 K_i (nM) 20	(K_i /$K_{D\ CDK7}$) *99*
			Comparator 4 	CDK7 K_D (nM) = 0.34	
				CDK12 K_i (nM) 48	(K_i /$K_{D\ CDK7}$) *140*
				CDK2 K_i (nM) 64	(K_i /$K_{D\ CDK7}$) *190*
				CDK9 K_i (nM) 36	(K_i /$K_{D\ CDK7}$) *100*

FIG. 2

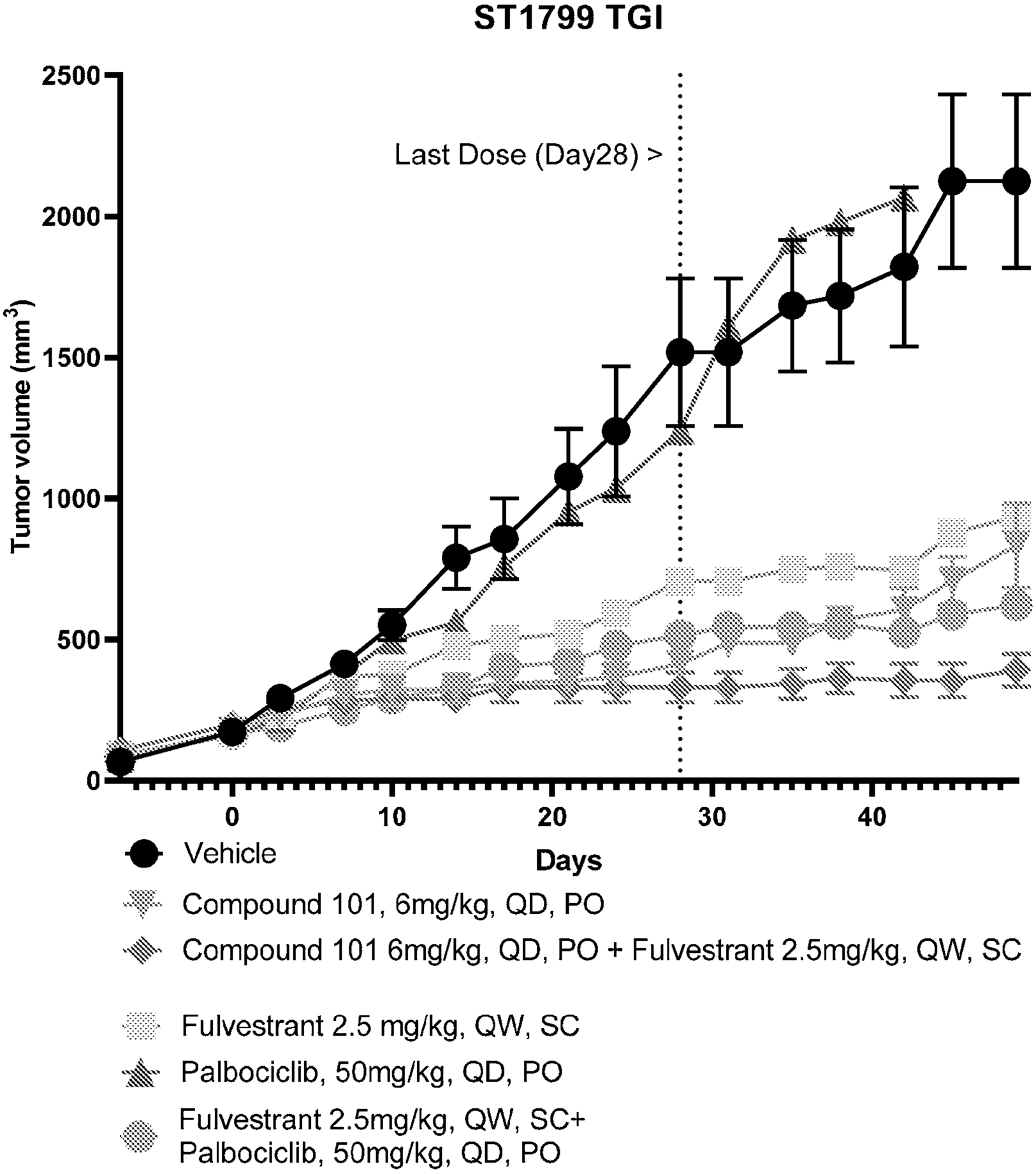

ST1799 TGI
Last Dose (Day28) >
Tumor volume (mm3)
2500
2000
1500
1000
500
0
0
10
20
30
40
Days
Vehicle
Compound 101, 6mg/kg, QD, PO
Compound 101 6mg/kg, QD, PO + Fulvestrant 2.5mg/kg, QW, SC
Fulvestrant 2.5 mg/kg, QW, SC
Palbociclib, 50mg/kg, QD, PO
Fulvestrant 2.5mg/kg, QW, SC+
Palbociclib, 50mg/kg, QD, PO

FIG. 3

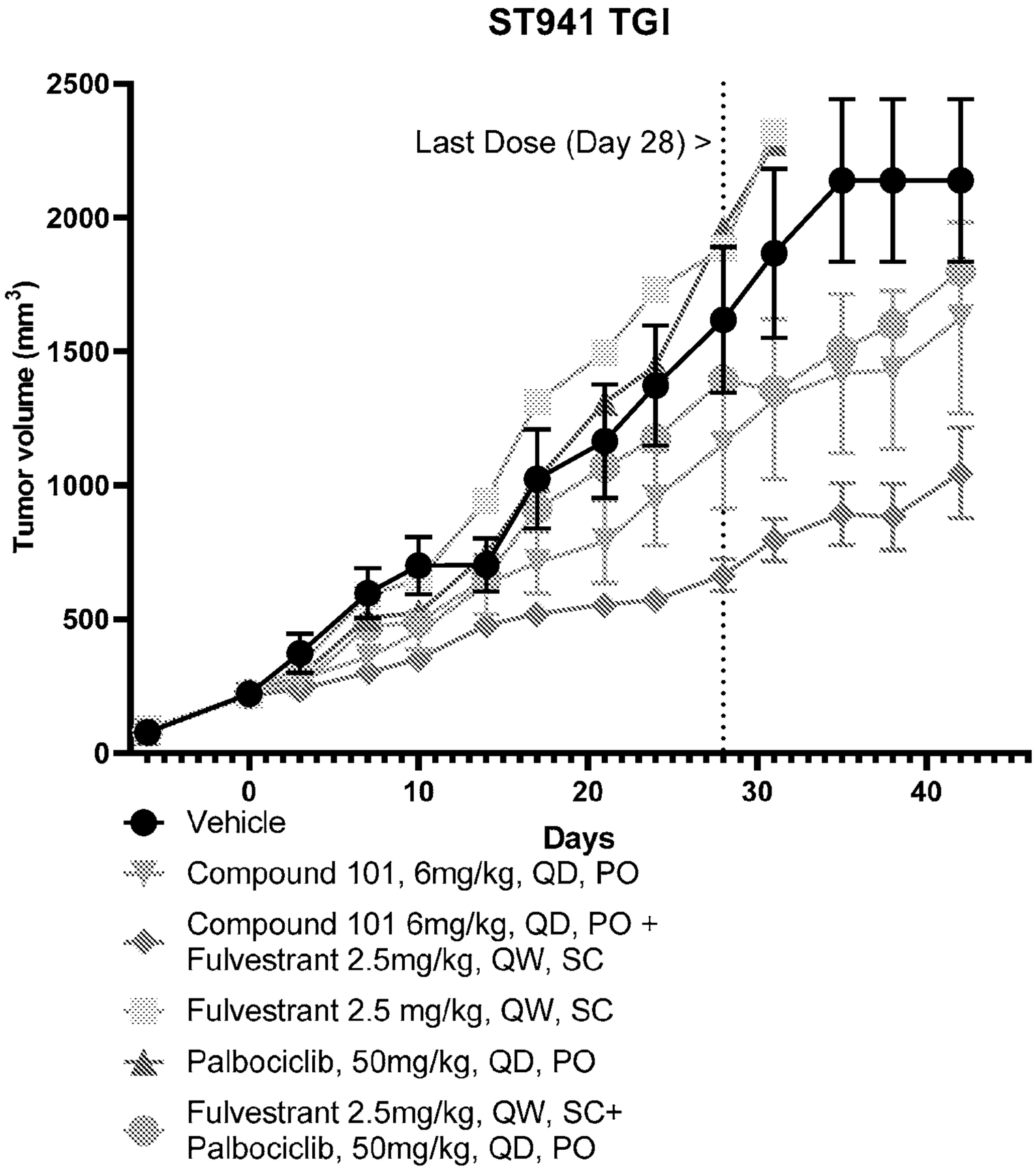

ST941 TGI
Last Dose (Day 28) >
Tumor volume (mm3)
2500
2000
1500
1000
500
0
0
10
20
30
40
Days
Vehicle
Compound 101, 6mg/kg, QD, PO
Compound 101 6mg/kg, QD, PO +
Fulvestrant 2.5mg/kg, QW, SC
Fulvestrant 2.5 mg/kg, QW, SC
Palbociclib, 50mg/kg, QD, PO
Fulvestrant 2.5mg/kg, QW, SC+
Palbociclib, 50mg/kg, QD, PO

FIG. 4

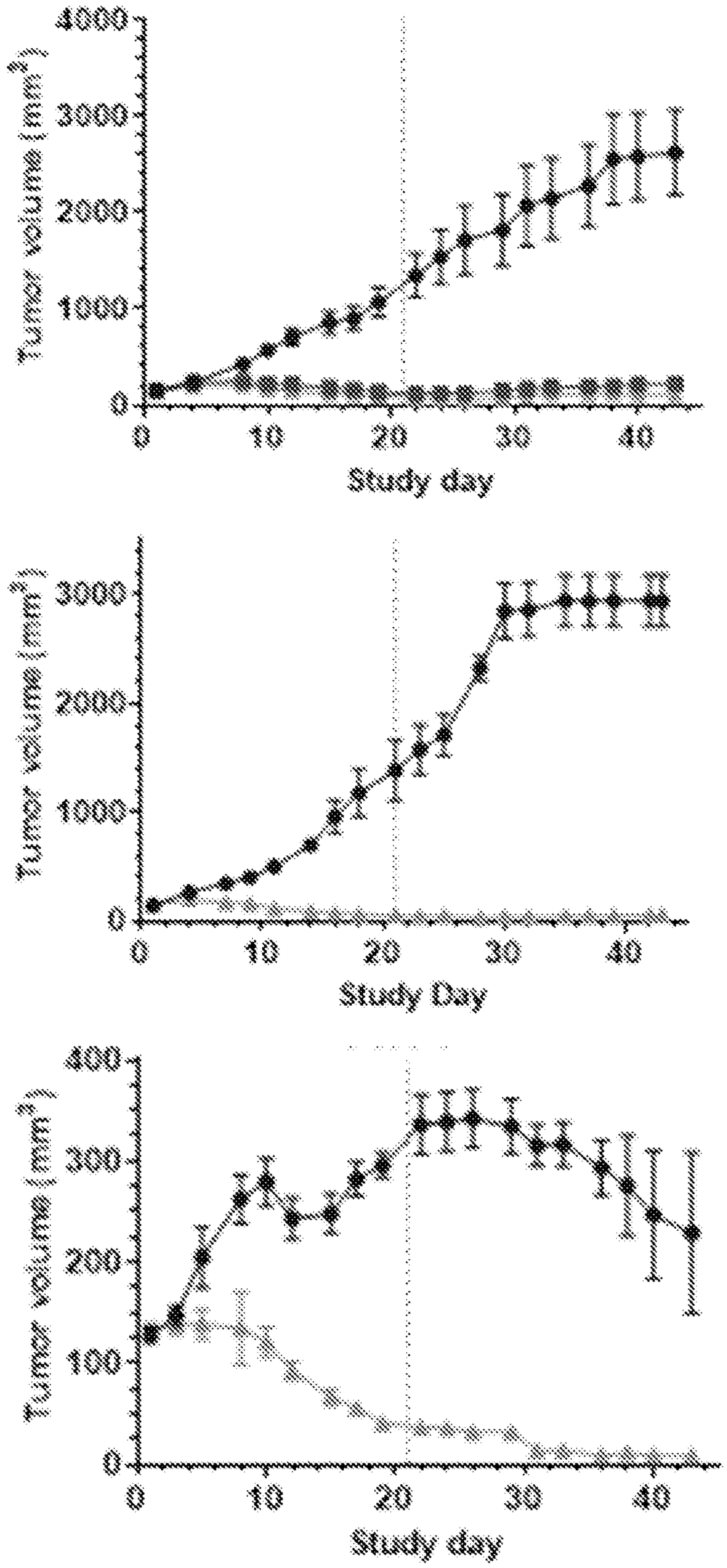

tumor volume (mm³)
4000
3000
2000
1000
0
0
10
20
30
40
Study day
tumor volume (mm³)
3000
2000
1000
0
0
10
20
30
40
Study Day
tumor volume (mm³)
400
300
200
100
0
0
10
20
30
40
Study day

FIG. 5

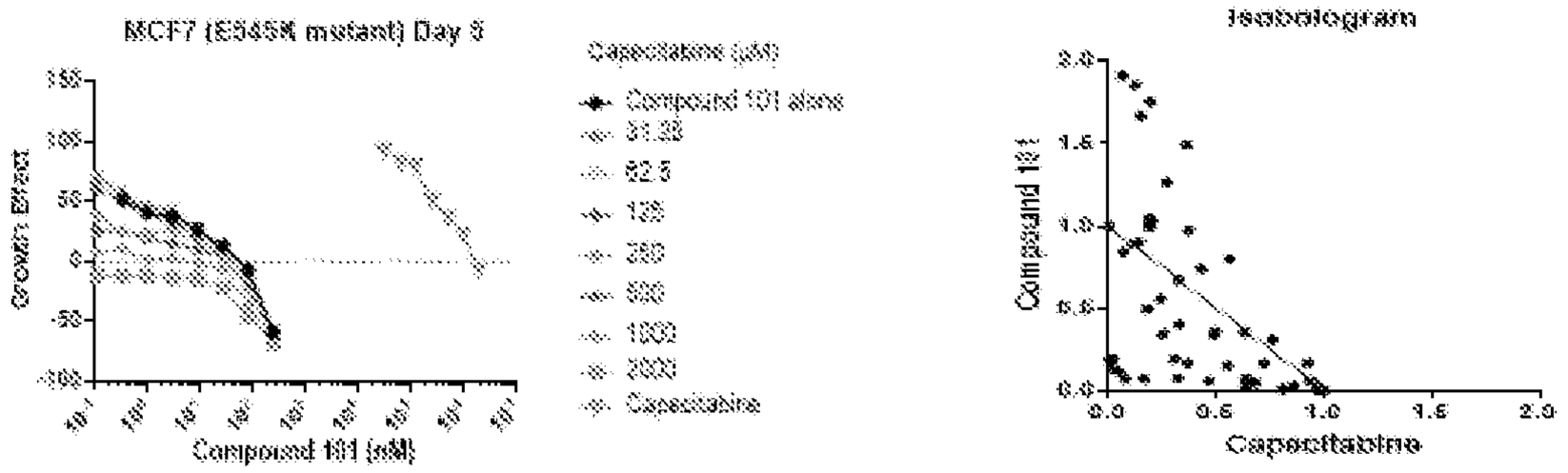

MCF7 (E545K mutant) Day 5
Growth Effect
Compound 101 (nM)
Capecitabine (μM)
Compound 101 alone
31.25
62.5
125
250
500
1000
2000
Capecitabine
Isobologram
Compound 101
Capecitabine
T47D (H1047R mutant) Day5
Growth Effect
Compound 101 (nM)
Alpelisib (μM)
Compound 101 alone
0.034294
0.102881
0.308642
0.925926
2.777778
8.333333
25
Alpelisib
Isobologram
Compound 101
Alpelisib
MCF7 (E545K mutant) Day 5
Growth Effect
Compound 101 (nM)
Alpelisib (μM)
Compound 101 alone
0.0343
0.1029
0.3086
0.9259
2.7778
8.3333
25
Alpelisib
Isobologram
Compound 101
Alpelisib
RKO (V600E mutant) Day5
Growth Effect
Compound 101 (nM)
Oxaliplatin (μM)
Compound 101 alone
0.013717
0.041152
0.123457
0.37037
1.1111
3.3333
10
Oxaliplatin
Isobologram
Compound 101
Oxaliplatin

FIG. 5 (cont.)

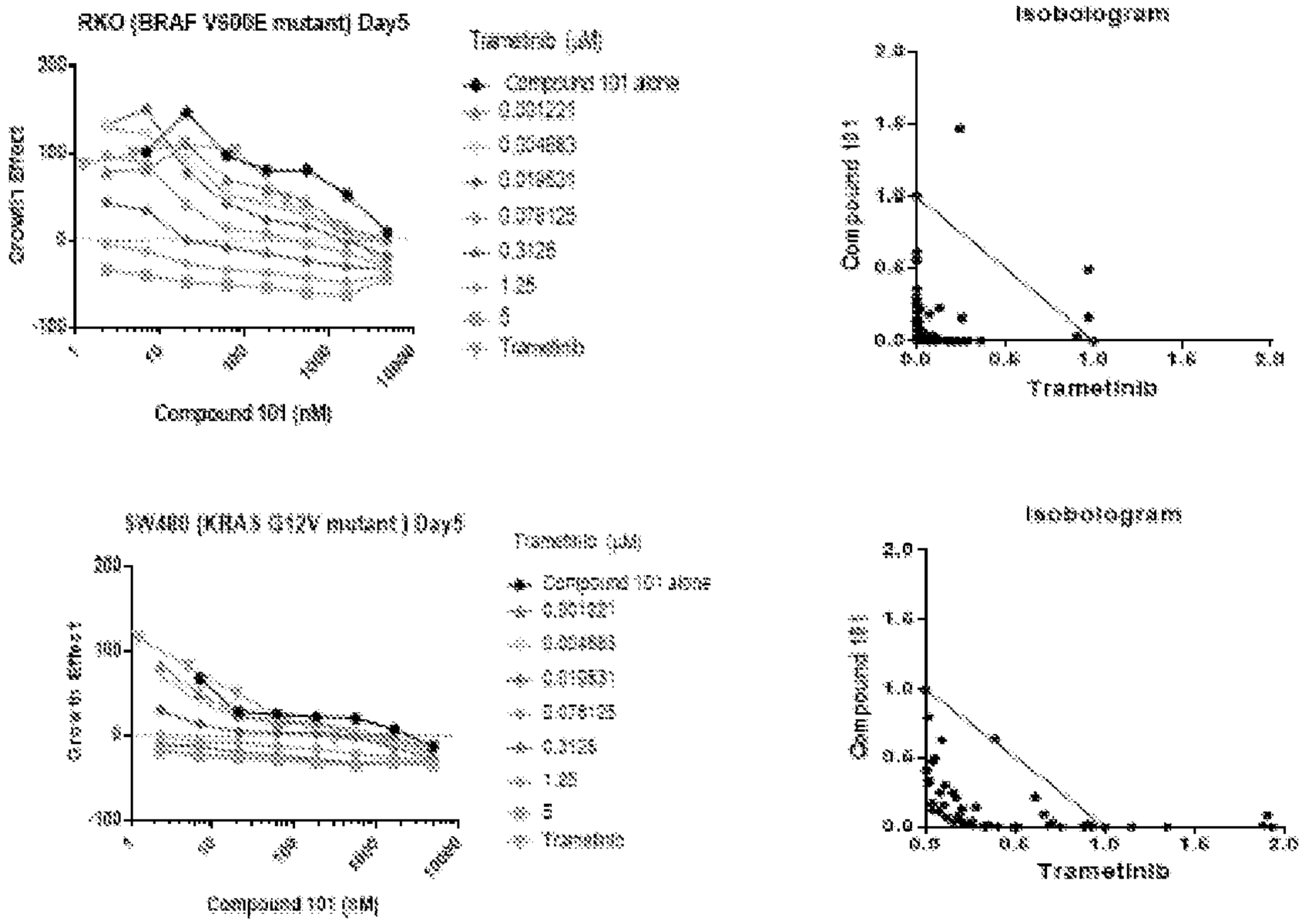

RKO (BRAF V600E mutant) Day5
Growth Effect
Trametinib (μM)
Compound 101 alone
0.391221
0.048883
0.019531
0.078125
0.3125
1.25
5
Trametinib
Compound 101 (nM)
Isobologram
Compound 101
Trametinib
SW480 (KRAS G12V mutant) Day5
Growth Effect
Trametinib (μM)
Compound 101 alone
0.391221
0.048883
0.019531
0.078125
0.3125
1.25
5
Trametinib
Compound 101 (nM)
Isobologram
Compound 101
Trametinib
NCI-H1648 (NS mutant/deletion) Day5
Growth Effect
Gemcitabine (μM)
Compound 101 alone
0.25
6.1E-05
0.000244
0.000977
0.003906
0.015625
0.0625
Gemcitabine
Compound 101 (nM)
Isobologram
Compound 101
Gemcitabine
NCI-H1648 (RB mutant/deletion) Day5
Growth Effect
Carboplatin (μM)
Compound 101 alone
100
1.5625
3.125
6.25
12.5
25
50
Carboplatin
Compound 101 (nM)
Isobologram
Compound 101
Carboplatin

FIG. 6

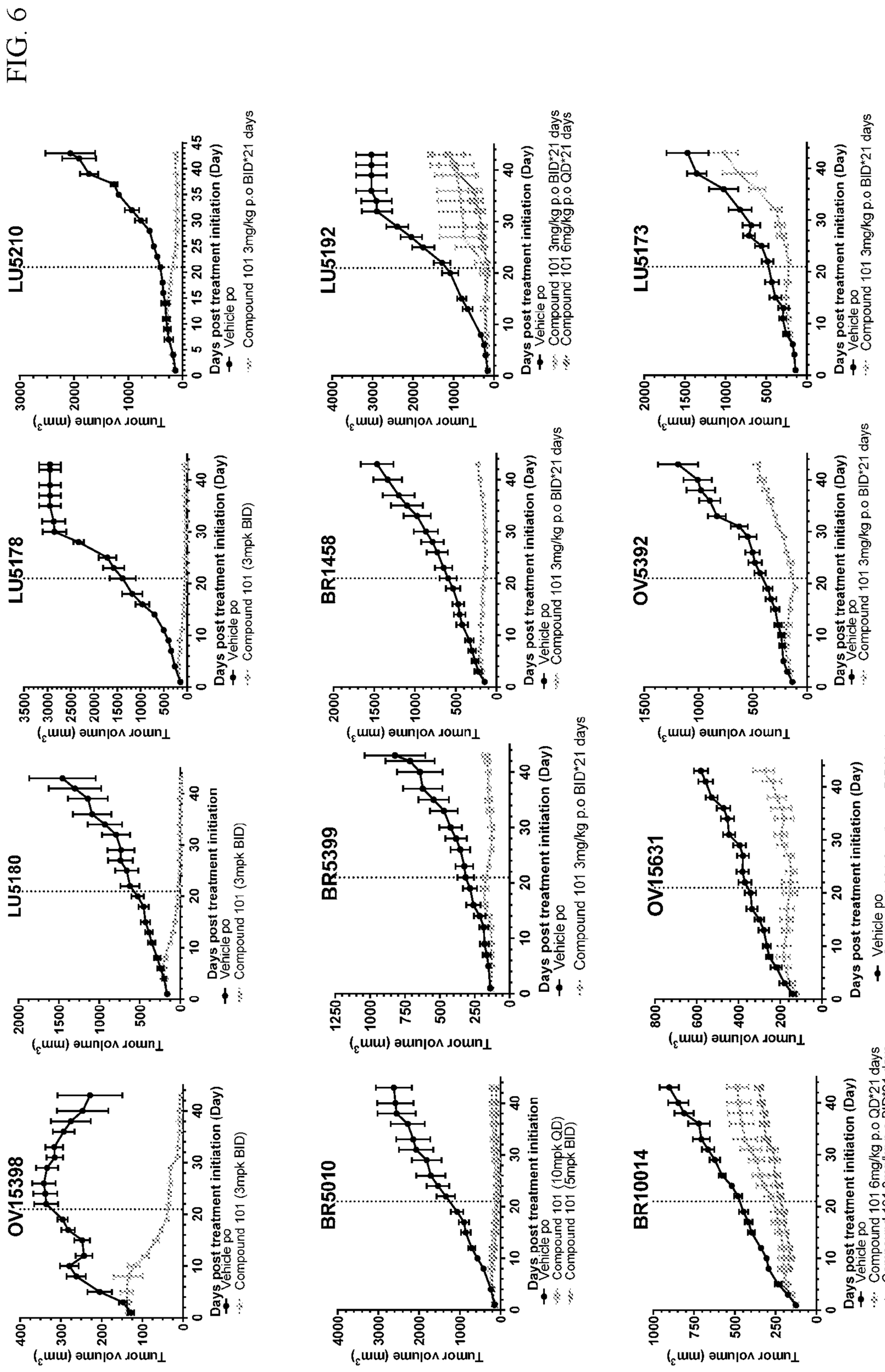

LU5210
Tumor volume (mm3)
Days post treatment initiation (Day)
Vehicle po
Compound 101 3mg/kg p.o BID*21 days
LU5178
Compound 101 (3mpk BID)
LU5180
Compound 101 (3mpk BID)
LU5192
Compound 101 6mg/kg p.o QD*21 days
BR1458
BR5399
Compound 101 3mg/kg p.o BID*21 days
LU5173
OV5392
OV15631
OV15398
BR5010
Compound 101 (10mpk QD)
Compound 101 (5mpk BID)
BR10014
Compound 101 6mg/kg p.o QD*21 days

FIG. 7

PDX model	End of Treatment Day 21		Post-treatment Day 42		RB pathway genetics
	% TGI	% Regression	% TGI	% Regression	
OV15398	>100	80	>100	98	RB1 (CN=1, p.Q637*)
LU5180	>100	72	>100	93	CDKN2A (CN=1)
LU5178	>100	64	>100	73	RB1 (CN=0)
BR5010	>100	55	>100	27	CCNE1 (CN=21)
LU5210	82	0	>100	8	RB1 (p.L343SfsTer3)
BR1458	90	0	95	0	CDKN2A (CN=0) RB1 (CN=1, p.Y155C)
BR5399	82	0	95	0	RB1 (CN=1)
LU5192	94	0	69	0	None
BR10014	91	0	67	0	None
OV5392	92	0	64	0	None
OV15631	70	0	71	0	None
LU5173	74	0	33	0	None

METHODS OF TREATING CANCER IN BIOMARKER-IDENTIFIED PATIENTS WITH NON-COVALENT INHIBITORS OF CYCLIN-DEPENDENT KINASE 7 (CDK7)

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of the filing date of U.S. provisional application No. 62/754,398, filed Nov. 1, 2018; U.S. provisional application No. 62/877,189, filed Jul. 22, 2019; U.S. provisional application No. 62/915,983, filed Oct. 16, 2019, and U.S. provisional application No. 62/927,469, filed Oct. 29, 2019. The content of each of these prior applications is hereby incorporated by reference herein in its entirety.

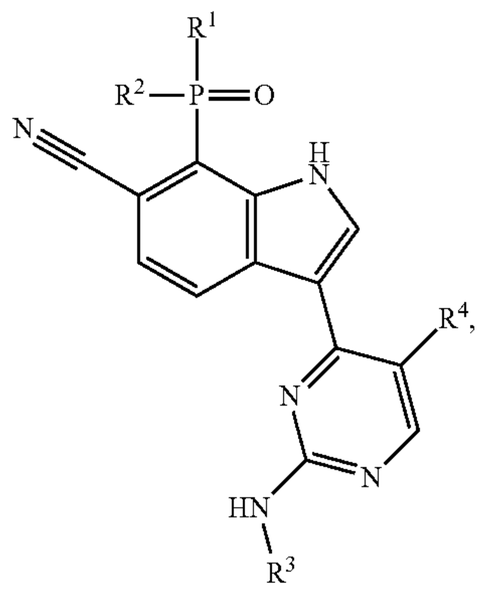
BACKGROUND OF THE INVENTION

[0002] The long evolution of healthcare has reached a point in time where the promise of biomarker analysis is beginning to be realized. When physicians can stratify patients, even those who share many similar physiological traits and exhibit common symptoms of a given disease, into more specific groups, they can better tailor treatment and optimize the outcome for each patient. However, it is challenging to develop molecular diagnostics and few are commercially available.

SUMMARY OF THE INVENTION

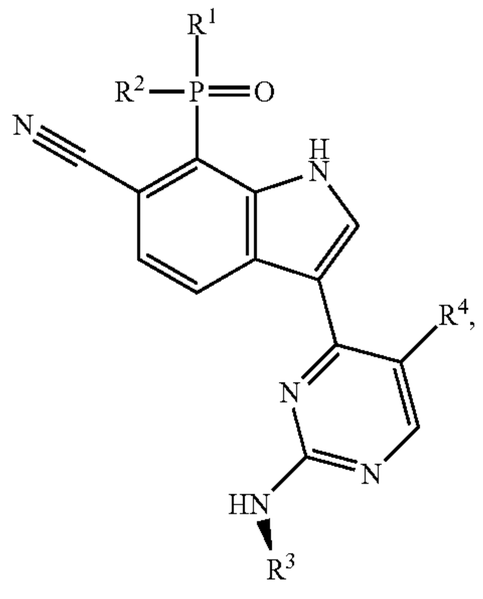
[0003] The present invention features, inter alia, diagnostic methods for identifying cancer patients for treatment with a non-covalent CDK7 inhibitor described herein (i.e., diagnostic methods for selecting a patient for treatment) and methods for treating identified patients with such an inhibitor, either alone or in combination with one or more additional therapeutic agents (e.g., a second anti-cancer agent), as described further below. The diagnostic methods include a step of identifying a patient suffering from a cancer that is likely to respond well to treatment with a non-covalent CDK7 inhibitor represented by structural Formula (I), (Ia), a species thereof, or a specified form thereof, as shown and described further below. The treatment methods include a step of administering such a non-covalent CDK7 inhibitor to an identified patient, whose response can be, for example, significant tumor growth inhibition (TGI; e.g., more than about 80-90% TGI and/or continued tumor suppression for a period of time after cessation of treatment). Thus, the present invention encompasses methods in which a patient is only diagnosed as being a good candidate for treatment (i.e., identified for treatment), methods in which a patient who has been determined to be a good candidate for treatment (e.g., previously identified) is treated, and methods requiring that a patient be both diagnosed and treated as described herein. The diagnostic methods that identify a patient for treatment include a step of analyzing one or more of the biomarkers described herein in a biological sample obtained from the patient by determining, having determined, or receiving information concerning the state of the biomarker. In various embodiments, the biomarker is analyzed to determine: whether it is present and/or in what amount (e.g., analyzed for a genetic deletion or amplification (e.g., copy number variation (CNV)); its location (e.g., chromosomal translocation); its sequence (i.e., the analysis can include determining whether the gene is present in wild type form or includes a mutation); whether it includes an epigenetic modification (e.g., histone and/or DNA methylation or histone acetylation); whether it is associated with a super-enhancer (SE) or a SE of a certain strength; its level of expression (as evidenced by, for example, the level of transcribed RNA (e.g., primary RNA or mRNA)); and/or whether a protein encoded by the biomarker gene has an aberrant level of expression or activity (in case of doubt, a protein encoded by a biomarker gene described herein can also serve as the biomarker). The state of a biomarker can be assessed by examining any one or more of the features just listed, and when we refer to "analyzing a/the biomarker," we mean analyzing one or more of these features (i.e., sequence, copy number, association with a SE, a level of RNA expression, and so forth, as provided above). For example, when we refer to analyzing the biomarker RB1, we mean analyzing or determining whether an RB1 gene is, for example, absent in a biological sample, contains a mutation (e.g., a mutation predisposing a patient to cancer), is translocated, has a CNV (copy number alteration (CNA)), bears an epigenetic modification, is associated with a super-enhancer (SE), is overexpressed or under-expressed (as evidenced by, for example, its level of RNA (e.g., primary RNA or mRNA), and/or encodes a protein with a level of expression or activity that is above or below a predetermined threshold level. As this implies, each feature analyzed can be determined to be equal to or above a pre-determined threshold level or equal to or below a pre-determined threshold level, as described further below. More specifically, in the methods of the present invention, one can analyze a biomarker selected from the genes BRAF, c-myc (also known as MYC), CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2; see also the Table below), or the proteins encoded thereby, by determining, having determined, and/or receiving information that the state of such a biomarker, as evidenced by a feature just described (e.g., RNA level) is equal to or above (e.g., above) a pre-determined threshold level. Alternatively, or in addition, one can analyze a biomarker selected from the genes BCL2-like 1, CDK7, CDK9, CDKN2A, and RB (also known as RB1 or another E2F pathway member, such as RBL1, RBL2, CDKN2A, CDKN2B, CDKN2C, CDKN2D, CDKN1A, CDKN1B, CDKN1C, and FBXW7), or the proteins encoded thereby, by determining, having determined, and/or receiving information that the state of such biomarker is equal to or below (e.g., below) a pre-determined threshold level. The proteins encoded by the genes just listed as useful biomarkers in the present methods are known in the art. For example, BRAF encodes B-Raf; c-myc encodes MYC, CCNE1 encodes cyclin E1 (see Koff et al., *Cell* 66:1217-1228, 1991); FGFR1 encodes FGFR1, a cell surface membrane receptor with tyrosine kinase activity; RB encodes pRB, which binds to the activator domain of activator E2F; BCL2-like 1 encodes BCL-xL, a transmembrane protein in mitochondria; CDK7 encodes CDK7; CDK9 encodes CDK9; PIK3CA encodes the p110α protein (a catalytic subunit of the class I PI3-kinases), and CDKN2A encodes p16 and p14arf. Aliases, chromosomal locations, splice variants, and homologs of the genes and proteins described herein as biomarkers, in *Homo sapiens* and species other than *Homo sapiens*, are also known.

[0004] The treatment methods of the invention and corresponding "uses" include administering, or the use of, a compound of Formula (I), any of which may be included in a pharmaceutically acceptable composition and administered, e.g., by a route and regimen described herein, to a patient identified as described herein. Compounds useful in the present methods have structural Formula (I):



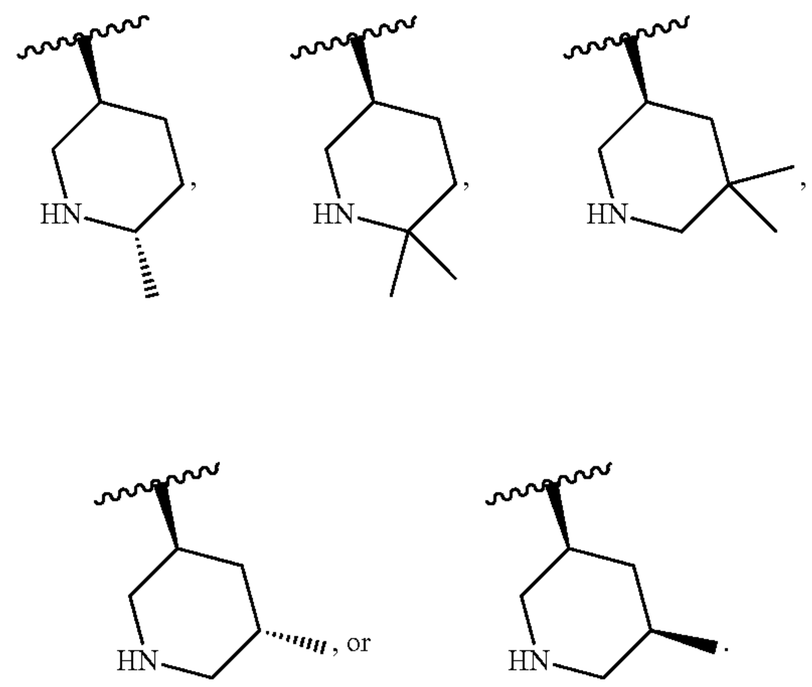
(I)

or a pharmaceutically acceptable salt, solvate, stereoisomer or mixture of stereoisomers, tautomer, or isotopic form thereof, wherein R^1 is methyl or ethyl; R^2 is methyl or ethyl; R^3 is 5-methylpiperidin-3-yl, 5,5-dimethylpiperidin-3-yl, 6-methylpiperdin-3-yl, or 6,6-dimethylpiperidin-3-yl, wherein one or more hydrogen atoms in R^3 is optionally replaced by deuterium; and R^4 is $—CF_3$ or chloro. More specifically, in a compound of Formula (I) or in the pharmaceutically acceptable salt, solvate, stereoisomer or mixture of stereoisomers, tautomer, isotopic form, or other specified form thereof (i) IV is methyl and R^2 is methyl or (ii) R^1 is methyl and R^2 is ethyl. In other embodiments, R^1 is ethyl and R^2 is ethyl. In some aspects of any one of these embodiments, R^4 is $—CF_3$. In other aspects of any one of these embodiments, R^4 is chloro. In various aspects of any of the preceding embodiments, R^3 is 5-methylpiperidin-3-yl, R^3 is 5,5-dimethylpiperidin-3-yl, R^3 is 6-methyl-piperdin-3-yl, or R^3 is 6,6-dimethylpiperidin-3-yl, wherein one or more hydrogen atoms in R^3 is optionally replaced by deuterium. A compound of Formula (I) can have structural Formula (Ia):

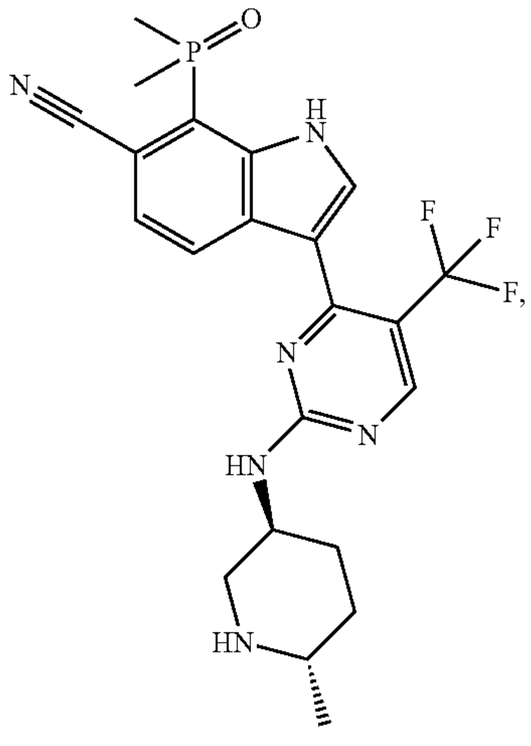


(Ia)

and the invention encompasses pharmaceutically acceptable salts, solvates (e.g., hydrates), tautomers, isotopic forms, or other specified forms of a compound of Formula (Ia), wherein R^3 is



[0005] More specifically, in a compound of Formula (Ia) or a pharmaceutically acceptable salt, solvate, tautomer, isotopic form, or other specified form thereof (i) R^1 is methyl and R^2 is methyl or (ii) R^1 is methyl and R^2 is ethyl. In other embodiments, R^1 is ethyl and R^2 is ethyl. In some embodiments, in a compound of Formula (Ia) or a specified form thereof, R^4 is $—CF_3$. In other embodiments, in a compound of Formula (Ia) or a specified form thereof, R^4 is chloro. In some embodiments, a compound of Formula (I) or (Ia) is:



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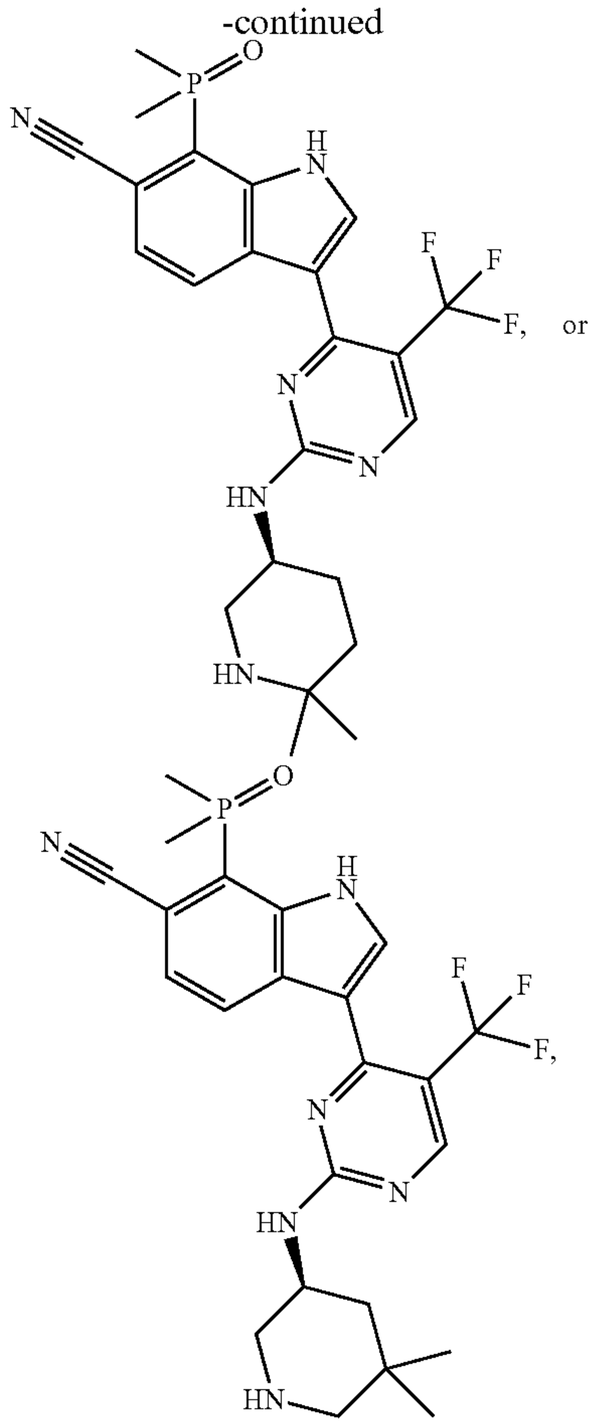
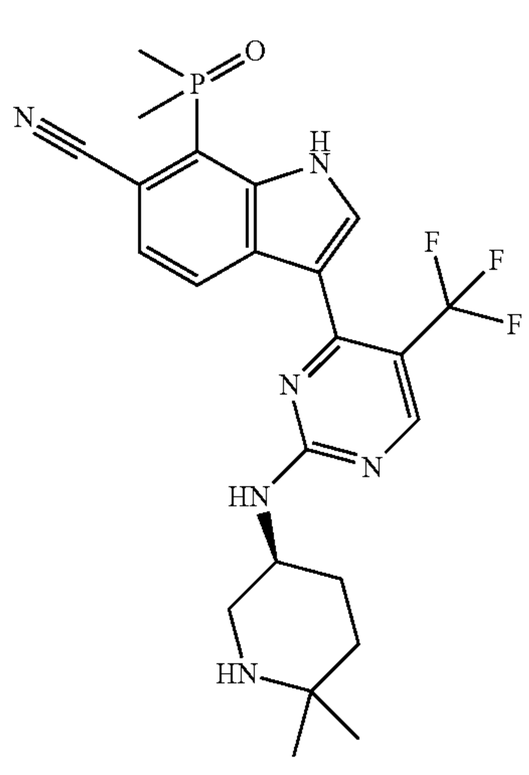


and the invention encompasses methods and the use of pharmaceutically acceptable salts, solvates (e.g., hydrates), tautomers, isotopic forms or other specified forms of any one of the three foregoing compounds. In one embodiment, the compound is



or a pharmaceutically acceptable salt thereof. The invention also encompasses solvates (e.g., hydrates), tautomers, isotopic forms or other specified forms of the foregoing compound. In isotopic forms, one or more hydrogen atoms in R^3 is replaced with deuterium. In other embodiments, none of the hydrogen atoms of a compound (e.g., none of the hydrogen atoms in R^3) are replaced with deuterium. Any compound of Formula (I), (Ia), or a species thereof can be of a "specified form," by which we mean a salt, solvate (e.g., hydrate), stereoisomer (or mixture thereof), tautomer, or isotopic form of a compound of Formula (I), (Ia), or a species thereof. Also within the meaning of "specified form" are forms of a compound that manifest a combination of the attributes, features, or properties of a salt, solvate, stereoisomer, tautomer, or isotopic form. For example, the methods and uses of the invention can be carried out with a salt that has been solvated (e.g., a hydrated) or a salt of a stereoisomer, tautomer, or isotopic form of a compound of Formula I, I(a), or a species thereof; with a solvate (e.g., hydrate) containing a salt, stereoisomer, tautomer, or isotopic form of a compound of Formula I, I(a), or a species thereof; with a stereoisomer of a compound of Formula I, I(a), or a species thereof that is in the form of a salt or solvate (e.g., hydrate) or is a tautomer or isotopic form of a compound of Formula I, I(a), or a species thereof, with a tautomer of a compound of Formula I, I(a), or a species thereof that is in the form of a salt or solvate (e.g., hydrate) or that is a stereoisomer or isotopic form of a compound of Formula I, I(a), or a species thereof; or with an isotopic form of a compound of Formula I, I(a), or a species thereof that is a salt, solvate (e.g., hydrate), stereoisomer, or tautomer of a compound of Formula I, I(a), or a species thereof. Any of these specified forms can be pharmaceutically acceptable and/or contained within a pharmaceutically acceptable composition (e.g., formulated for oral administration) for use in a method described herein.

[0006] Accordingly, the invention features treatment methods including a step of administering a compound of structural Formula (I), or a pharmaceutically acceptable salt, solvate, stereoisomer or mixture of stereoisomers, tautomer, or isotopic form thereof, optionally within a pharmaceutical composition, wherein R', R^2, R^3, and R^4 are as defined herein, in treating cancer in a selected patient, wherein the patient has been determined to have a cancer in which: (a) a gene selected from RB1, RBL1, RBL2, CDKN2A, CDKN2B, CDKN2C, CDKN2D, CDKN1A, CDKN1B, CDKN1C, and FBWX7 is mutated, is genetically deleted, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard; (b) a gene selected from E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CDK1, CDK2, CDK4, CDK6, CCNA1, CCNB1, CCND1, CCND2, CCND3, CCNE1, CCNE2, and BRAF is mutated, is genetically gained or amplified, contains an epigenetic alteration, is translocated, transcribed at a level equal to or above a pre-determined threshold, or encodes a protein that is translated at a level equal to or above a pre-determined threshold or has increased activity relative to a reference standard; or (c) the gene Bcl2-like 1 is mutated, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a BCL-xL protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard. In any embodiment of this method, the cancer is a blood cancer,

preferably an acute myeloid leukemia (AML), a breast cancer, preferably a triple negative breast cancer (TNBC) or a hormone receptor positive (HR+) breast cancer, an osteosarcoma or Ewing's sarcoma, fallopian tube cancer, a GI tract cancer, preferably colorectal cancer, a glioma, a lung cancer, preferably small cell or non-small cell lung cancer, melanoma, an ovarian cancer, preferably a high grade serous ovarian cancer, epithelial ovarian cancer, or clear cell ovarian cancer, a pancreatic cancer, a primary peritoneal cancer, prostate cancer, retinoblastoma, or a squamous cell cancer of the head or neck. For example, the patient may have such a cancer and can be treated as described herein when it has been determined that, in a biological sample obtained from the patient, Bcl2-like 1 is mutated, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a BCL-xL protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard, preferably wherein a level of Bcl2-like 1 mRNA in the cancer is equal to or below the pre-determined threshold level. Further, such a patient can be one who has undergone, is presently undergoing, or is prescribed treatment with a Bcl-2 inhibitor, as known in the art and/or described herein. In some embodiments, the Bcl-2 inhibitor is venetoclax and the patient has a breast cancer (e.g., TNBC); a blood cancer (e.g., AML); an ovarian cancer (e.g., HGSOC); or a lung cancer (e.g., SCLC or NSCLC). In other embodiments, the patient may have such a cancer and can be treated as described herein when it has been determined that, in a biological sample obtained from the patient: (a) RB1 or CDKN2A is mutated, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard, preferably wherein RB1 or CDKN2A mRNA, preferably RB1 mRNA, is equal to or below the pre-determined threshold; and/or (b) CDK6, CCND2, or CCNE1 is mutated, has a copy number alteration, contains an epigenetic alteration, is translocated, transcribed at a level equal to or above a pre-determined threshold, or encodes a protein that is translated at a level equal to or above a pre-determined threshold or has increased activity relative to a reference standard, preferably wherein CDK6, CCND2, or CCNE1 mRNA, preferably CCNE1 mRNA, is equal to or above a pre-determined threshold level. Such a patient can be one who has undergone, is presently undergoing, or is prescribed treatment with a selective estrogen receptor modulator (SERM; e.g., tamoxifen, raloxifene, or toremifene), a selective estrogen receptor degrader (SERD; e.g., fulvestrant), a PARP inhibitor (e.g., olaparib or niraparib); or a platinum-based therapeutic agent (e.g., cisplatin, oxaliplatin, nedaplatin, carboplatin, phenanthriplatin, picoplatin, satraplatin (JM216). More specifically, the patient treated with a SERM or SERD may have an HR+ breast cancer; the patient treated with a PARP inhibitor may have a TNBC or a $Her2^{+}/ER^{-}/PR^{-}$ breast cancer, fallopian tube cancer, glioma, ovarian cancer (e.g., an epithelial ovarian cancer), or primary peritoneal cancer; and the patient treated with a platinum-based therapeutic agent may have an ovarian cancer.

[0007] In any of the present methods where a compound of Formula (I), (Ia), a species thereof or a specified form thereof is used or administered, optionally within a pharmaceutical composition, the patient can be one who has undergone, is presently undergoing, or is prescribed treatment with a BET inhibitor such as ABBV-075, BAY-299, BAY-1238097, BMS-986158, CPI-0610, CPI-203, FT-1101, GS-5829, GSK-2820151, GSK-525762, I-BET151, I-BET762, INCB054329, JQ1, MS436, OTX015, PFI-1, PLX51107, RVX2135, TEN-010, ZEN-3694, or a compound disclosed in U.S. application Ser. No. 12/810,564; with a CDK4/6 inhibitor such as BPI-1178, G1T38, palbociclib, ribociclib, ON 123300, trilaciclib, or abemaciclib, preferably palbociclib; with a FLT3 inhibitor such as CDX-301, CG'806, CT053PTSA, crenolanib (e.g., crenolanib besylate), ENMD-2076, FF-10101-01, FLYSYN, gilteritinib (ASP2215), HM43239, lestautinib, ponatinib, NMS-088, sorafenib, sunitinib, pacritinib, pexidartinib/PLX3397, quizartinib, midostaurin, SEL24, SKI-G-801, or SKLB1028, preferably crenolanib, gilteritinib, or midostaurin; or with a MEK inhibitor such as trametinib, cobimetinib, or binimetinib. More specifically, a patient who has undergone, is presently undergoing, or is prescribed treatment: with a CDK4/6 inhibitor has a breast cancer, preferably a TNBC or an estrogen receptor-positive (ER^{+}) breast cancer, a pancreatic cancer, or a squamous cell cancer of the head or neck; with a FLT3 inhibitor has a blood cancer, preferably AML; with a BET inhibitor has a breast cancer, preferably TNBC, a blood cancer, preferably AML, Ewing's sarcoma, or an osteosarcoma.

[0008] In any of the present methods where compound of Formula (I), (Ia), a species thereof or a specified form thereof is used or administered, optionally within a pharmaceutical composition, the patient can be one who has undergone, is presently undergoing, or is prescribed treatment with a Bcl-2 inhibitor such as APG-1252, APG-2575, BP1002 (prexigebersen), the antisense oligonucleotide known as oblimersen (G3139), S55746/BCL201, or venetoclax; a CDK9 inhibitor such as alvocidib/DSP-2033/flavopiridol, AT7519, AZD5576, BAY1251152, BAY1143572, CYC065, nanoflavopiridol, NVP2, seliciclib (CYC202), TG02, TP-1287, VS2-370 or voruciclib (formerly P1446A-05); a hormone receptor (e.g., estrogen receptor) degradation agent, such as fulvestrant; a Flt3 (FMS-like tyrosine kinase 3) inhibitor such as CDX-301, CG'806, CT053PTSA, crenolanib (e.g., crenolanib besylate), ENMD-2076, FF-10101-01, FLYSYN, gilteritinib (ASP2215), HM43239, lestautinib, ponatinib, NMS-088, sorafenib, sunitinib, pacritinib, pexidartinib/PLX3397, quizartinib, midostaurin, SEL24, SKI-G-801, or SKLB1028; a PARP inhibitor such as olaparib, rucaparib, talazoparib, veliparib (ABT-888), or niraparib; a BET inhibitor such as ABBV-075, BAY-299, BAY-1238097, BMS-986158, CPI-0610, CPI-203, FT-1101, GS-5829, GSK-2820151, GSK-525762, I-BET151, I-BET762, INCB054329, JQ1, MS436, OTX015, PFI-1, PLX51107, RVX2135, TEN-010, ZEN-3694, or a compound disclosed in U.S. application Ser. No. 12/810,564 (now U.S. Pat. No. 8,476,260); a platinum-based therapeutic agent such as cisplatin, oxaliplatin, nedaplatin, carboplatin, phenanthriplatin, picoplatin, satraplatin (JM216), or triplatin tetranitrate; a CDK4/6 inhibitor such as BPI-1178, G1T38, palbociclib, ribociclib, ON 123300, trilaciclib, or abemaciclib; a MEK inhibitor such as trametinib; or a phosphoinositide 3-kinase (PI3 kinase) inhibitor, optionally of Class I (e.g., Class IA) and/or optionally directed against a specific PI3K isoform, such as idelalisib, copanlisib, duvelisib, or alpelisib; or capecitabine. More specifically, the second

agent is selected from a Bcl-2 inhibitor such as venetoclax, a PARP inhibitor such as olaparib or niraparib, a platinum-based anti-cancer agent such as carboplatin or oxaliplatin, a taxane such as paclitaxel, a CDK4/6 inhibitor such as palbociclib, ribociclib, abemaciclib, or trilaciclib, a selective estrogen receptor modulator such as tamoxifen, and a selective estrogen receptor degrader such as fulvestrant.

[0009] The invention also features kits that include a compound of Formula I, I(a), a species thereof, or a specified form thereof, and instructional material (e.g., a product insert) that describes a suitable/identified patient, methods of identifying such a patient for treatment (e.g., by any one of the diagnostic stratification methods described herein for analyzing a biomarker), and/or instructions for administering the compound of Formula I, I(a), a species thereof, or a specified form thereof, alone or in combination with at least one other therapeutic agent (e.g., an additional/second anti-cancer therapeutic including any one or more of the second agents described herein). The kits of the invention can also include the second agent (e.g., an anti-cancer agent), including any one or more of the second agents described herein and instructions for use in a population of patients identified as described.

BRIEF DESCRIPTION OF THE DRAWINGS

[0010] FIG. **1** is a table depicting the inhibitory and dissociation constants and selectivity of the indicated compounds (three compounds of the invention and four comparators) against CDK2, CDK7, CDK9, and CDK12.

[0011] FIG. **2** is a line graph depicting changes in tumor volume (mm^3) over time (days) in a palbociclib-resistant HR+BC PDX model (ST1799), as described in the Examples below.

[0012] FIG. **3** is a line graph depicting tumor volume (mm^3) over time (days) in the palbociclib- and fulvestrant-resistant HR+BC PDX model ST941, as described in the Examples below.

[0013] FIG. **4** is a panel showing three line graphs that depict changes in tumor volume (mm^3) over time (days) in PDX models of TNBC (BR5010; top), small cell lung cancer (LU5178; middle), and ovarian cancer (OV15398; bottom). The animals were treated with Compound 101 as described in Example 10. Data obtained from vehicle-treated (control) animals is represented by filled circles (upper traces in each graph). Data from animals modeling TNBC and given 10 mg/kg Compound 101 QD are represented in the top graph by filled squares; the dose of 5 mg/kg BID is represented by triangles. Triangles also represent data obtained from the animal models of SCLC and ovarian cancer treated with Compound 101 in the middle and bottom graphs.

[0014] FIG. **5** is a panel of line graphs showing tumor growth in the PDX models indicated and corresponding isobolograms, each generated as described in Example 11. Compound 101 was applied to cells in combination with the indicated second agents at the concentrations shown.

[0015] FIG. **6** is a panel of graphs generated from data collected in the Compound 101-treated PDX models described in Example 12. Black lines with squares represent vehicle-treated animals. Gray lines represent Compound 101-treated animals. Error bars are SEM. BID=twice daily; CNV=copy number variation; MPK=mg per kg body weight; PO=oral; QD=once daily; RB=retinoblastoma; SCLC=small cell lung cancer; TNBC=triple negative breast cancer. Vertical dotted lines mark the last day of treatment.

[0016] FIG. **7** is a Table summarizing the TGI values and genetic status of the 12 PDX models studied as described in Example 12. Models in the table are sorted based on highest to lowest response at end of study. BID, CNV, RB, SCLC, and TNBC are as defined for FIG. **6** and elsewhere herein. In case of doubt, CCNE1=the gene encoding cyclin E1; CDKN2A=cyclin-dependent kinase inhibitor 2A, EoS=end of study, EoT=end of treatment, HGSOC=high-grade serous ovarian cancer, OVA=ovarian cancer, and TGI=tumor growth inhibition. For the LU5210 model, tissue was not available for confirmation of RB pathway genetics.

DETAILED DESCRIPTION

[0017] Despite the efficacy of compounds of Formula (I), we believe that such efficacy will be higher in patients that have certain genetic signatures (i.e., biomarkers in a particular state, which can be analyzed as described herein). Moreover, we believe the efficacy of compounds of Formula (I) may be enhanced when combined with other anti-cancer therapies in newly diagnosed and refractory cancer patients identified as described herein.

[0018] The following definitions apply to the compositions, methods, and uses described herein unless the context clearly indicates otherwise, and it is to be understood that the claims may be amended to include language within a definition as needed or desired. Moreover, the definitions apply to linguistic and grammatical variants of the defined terms (e.g., the singular and plural forms of a term), and some linguistic variants are particularly mentioned below (e.g., "administration" and "administering"). The chemical elements are identified in accordance with the Periodic Table of the Elements, CAS version, Handbook of Chemistry and Physics, 75th Ed. Additionally, general principles of organic chemistry are well established and one of ordinary skill in the art can consult, if desired, *Organic Chemistry* by Thomas Sorrell, University Science Books, Sausalito, 1999; Smith and March, *March's Advanced Organic Chemistry,* 5^{th} Edition, John Wiley & Sons, Inc., New York, 2001; Larock, *Comprehensive Organic Transformations*, VCH Publishers, Inc., New York, 1989; and Carruthers, *Some Modern Methods of Organic Synthesis,* 3^{rd} Edition, Cambridge University Press, Cambridge, 1987.

[0019] The term "about," when used in reference to a value, signifies any value or range of values that is plus-or-minus 10% of the stated value (e.g., within plus-or-minus 1%, 2%, 3%, 4%, 5%, 6%, 7%, 8%, 9% or 10% of the stated value). For example, a dose of about 10 mg means any dose as low as 10% less than 10 mg (9 mg), any dose as high as 10% more than 10 mg (11 mg), and any dose or dosage range therebetween (e.g., 9-11 mg; 9.1-10.9 mg; 9.2-10.8 mg; and so on). As another example, a prevalence rank in a population of about 80% means a prevalence rank of 72-88% (e.g., 79.2-80.8%). In case of doubt, "about X" can be "X" (e.g., about 80% can be 80%). Where a stated value cannot be exceeded (e.g., 100%), "about" signifies any value or range of values that is up to and including 10% less than the stated value (e.g., a purity of about 100% means 90%-100% pure (e.g., 95%-100% pure, 96%-100% pure, 97%-100% pure etc. . . .)). In the event an instrument or technique measuring a value has a margin of error greater than 10%, a given value will be about the same as a stated value when they are both within the margin of error for that instrument or technique.

[0020] The term "administration" and variants thereof, such as "administering," refer to the administration of a

compound described herein (e.g., a compound of Formula (I), (Ia), a species thereof or a specified form thereof (e.g., a pharmaceutically acceptable salt of a compound of Formula (I), (Ia), or a species thereof), or an additional/second agent), or a composition containing the compound to a subject (e.g., a human patient) or system (e.g., a cell- or tissue-based system that is maintained ex vivo); as a result of the administration, the compound or composition containing the compound (e.g., a pharmaceutical composition) is introduced to the subject or system. In addition to compositions of the invention and second agents useful in combination therapies, items used as positive controls, negative controls, and placebos, any of which can also be a compound, can also be "administered." One of ordinary skill in the art will be aware of a variety of routes that can, in appropriate circumstances, be utilized for administration to a subject or system. For example, the route of administration can be oral (i.e., by swallowing a pharmaceutical composition) or may be parenteral. More specifically, the route of administration can be bronchial (e.g., by bronchial instillation), by mouth (i.e., oral), dermal (which may be or comprise topical application to the dermis or intradermal, interdermal, or transdermal administration), intragastric or enteral (i.e., directly to the stomach or intestine, respectively), intramedullary, intramuscular, intranasal, intraperitoneal, intrathecal, intratumoral, intravenous (or intra-arterial), intraventricular, by application to or injection into a specific organ (e.g., intrahepatic), mucosal (e.g., buccal, rectal, sublingual, or vaginal), subcutaneous, tracheal (e.g., by intratracheal instillation), or ocular (e.g., topical, subconjunctival, or intravitreal). Administration can involve intermittent dosing (i.e., doses separated by various times) and/or periodic dosing (i.e., doses separated by a common period of time (e.g., every so many hours, daily (e.g., once daily oral dosing), weekly, twice per week, etc.)). In other embodiments, administration may involve continuous dosing (e.g., perfusion) for a selected time (e.g., about 1-2 hours).

[0021] Two events, two entities, or an event and an entity are "associated" with one another if one or more features of the first (e.g., its presence, level and/or form) are correlated with a feature of the second. For example, a first entity (e.g., an enzyme (e.g., CDK7)), gene expression profile, genetic signature (i.e., a single or combined group of genes in a cell with a uniquely characteristic pattern of gene expression), metabolite, or event (e.g., myeloid infiltration)) is associated with an event (e.g., the onset or progression of a particular disease), if its presence, level and/or form correlates with the incidence of, severity of, and/or susceptibility to the disease (e.g., a cancer disclosed herein). The biomarkers described herein are associated with an identified patient in the manner described herein (e.g., by virtue of their level of expression) and, depending on their status, can also be associated with a clinical outcome (e.g., a better prognosis based on an increased likelihood that a treatment regimen described herein will be more successful as evidenced by, e.g., TGI, preferably beyond the cessation of treatment). Associations are typically assessed across a relevant population. Two or more entities are physically "associated" with one another if they interact, directly or indirectly, so that they are and/or remain in physical proximity with one another in a given circumstance (e.g., within a cell maintained under physiological conditions (e.g., within cell culture) or within a pharmaceutical composition). Entities that are physically associated with one another can be covalently linked to one another or non-covalently associated by, for example, hydrogen bonds, van der Waals forces, hydrophobic interactions, magnetism, or combinations thereof. A compound of Formula (I), (Ia), a species thereof, or a specified form thereof (e.g., a pharmaceutically acceptable salt) can non-covalently associate with CDK7.

[0022] The term "biological sample" refers to a sample obtained or derived from a biological source of interest (e.g., a tissue or organism (e.g., an animal or human patient) or cell culture). For example, a biological sample can be a sample obtained from an individual (e.g., a patient or an animal model) suffering from a disease (or, in the case of an animal model, a simulation of that disease in a human patient) to be diagnosed and/or treated by the methods of this invention or from an individual serving in the capacity of a reference or control (or whose sample contributes to a reference standard or control population). The biological sample can contain a biological cell, tissue or fluid or any combination thereof. For example, a biological sample can be or can include ascites; blood; blood cells; a bodily fluid, any of which may include or exclude cells (e.g., tumor cells (e.g., circulating tumor cells (CTCs) found in at least blood or lymph vessels)) or circulating tumor DNA (ctDNA); bone marrow or a component thereof (e.g., hematopoietic cells, marrow adipose tissue, or stromal cells); cerebrospinal fluid (CSF); feces; flexural fluid; free-floating nucleic acids (e.g., circulating tumor DNA); gynecological fluids; hair; immune infiltrates; lymph; peritoneal fluid; plasma; saliva; skin or a component part thereof (e.g., a hair follicle); sputum; surgically-obtained specimens; tissue scraped or swabbed from the skin or a mucus membrane (e.g., in the nose, mouth, or vagina); tissue or fine needle biopsy samples; urine; washings or lavages such as a ductal lavage or broncheoalveolar lavage; or other body fluids, tissues, secretions, and/or excretions. Samples of, or samples obtained from, a bodily fluid (e.g., blood, CSF, lymph, plasma, or urine) may include tumor cells (e.g., CTCs) and/or free-floating or cell-free nucleic acids of the tumor. Cells (e.g., cancer cells) within the sample may have been obtained from an individual patient for whom a treatment is intended. Samples used in the form in which they were obtained may be referred to as "primary" samples, and samples that have been further manipulated (e.g., by removing one or more components of the sample) may be referred to as "secondary" or "processed" samples. Such processed samples may contain or be enriched for a particular cell type (e.g., a CDK7-expressing cell, which may be a tumor cell), cellular component (e.g., a membrane fraction), or cellular material (e.g., one or more cellular proteins, including CDK7, DNA, or RNA (e.g., mRNA), which may encode CDK7 and may be subjected to amplification). As used herein, the term "biomarker" refers to an entity whose state correlates with a particular biological event so that it is considered to be a "marker" for that event (e.g., the presence of a particular cancer and its susceptibility to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof). A biomarker can be analyzed at the nucleic acid or protein level; at the nucleic acid level, one can analyze the presence (e.g., copy number alterations (CNAs)), absence, or chromosomal location of a gene in wild type or mutant form, epigenetic alterations (in, e.g., methylation), its association with a super-enhancer, and/or its level of expression (as evidenced, for example, by primary RNA transcript or mRNA levels). At the protein level, one can analyze the level of expression and/or activity

of a protein encoded by a biomarker gene. A biomarker may indicate a therapeutic outcome or likelihood (e.g., increased likelihood) thereof. Thus, a biomarker can be predictive or prognostic and is therefore useful in methods of identifying or treating a patient as described herein.

[0023] The term "cancer" refers to a disease in which biological cells exhibit an aberrant growth phenotype characterized by loss of control of cell proliferation to an extent that will be detrimental to a patient having the disease. A cancer can be classified by the type of tissue in which it originated (histological type) and/or by the primary site in the body in which the cancer first developed. Based on histological type, cancers are generally grouped into six major categories: carcinomas; sarcomas; myelomas; leukemias; lymphomas; and mixed types. A cancer treated as described herein may be of any one of these types and may comprise cells that are precancerous (e.g., benign), malignant, pre-metastatic, metastatic, and/or non-metastatic. A patient who has a malignancy or malignant lesion has a cancer. The present disclosure specifically identifies certain cancers to which its teachings may be particularly relevant, and one or more of these cancers may be characterized by a solid tumor or by a hematologic tumor, which may also be known as a blood cancer (e.g., of a type described herein). Although not all cancers manifest as solid tumors, we may use the terms "cancer cell" and "tumor cell" interchangeably to refer to any malignant cell.

[0024] The term "combination therapy" refers to those situations in which a subject is exposed to two or more therapeutic regimens (e.g., two or more therapeutic agents) to treat a single disease (e.g., a cancer). The two or more regimens/agents may be administered simultaneously or sequentially. When administered simultaneously, a dose of the first agent and a dose of the second agent are administered at about the same time, such that both agents exert an effect on the patient at the same time or, if the first agent is faster- or slower-acting than the second agent, during an overlapping period of time. When administered sequentially, the doses of the first and second agents are separated in time, such that they may or may not exert an effect on the patient at the same time. For example, the first and second agents may be given within the same hour or same day, in which case the first agent would likely still be active when the second is administered. Alternatively, a much longer period of time may elapse between administration of the first and second agents, such that the first agent is no longer active when the second is administered (e.g., all doses of a first regimen are administered prior to administration of any dose(s) of a second regimen by the same or a different route of administration, as may occur in treating a refractory cancer). For clarity, combination therapy does not require that two agents be administered together in a single composition or at the same time, although in some embodiments, two or more agents, including a compound of Formula (I), (Ia), a species thereof, or a specified form thereof and a second agent described herein, may be administered within the same period of time (e.g., within the same hour, day, week, or month).

[0025] The terms "cutoff" and "cutoff value" mean a value measured in an assay that defines the dividing line between two subsets of a population (e.g., likely responders and non-responders (e.g., responders and non-responders to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof)). In some instances, values that are equal to or above the cutoff value define one subset of the population, and values that are lower than the cutoff value define the other subset of the population. In other instances, values that are equal to or below the cutoff value define on subset of the population, and values above the cutoff value define the other. As described further below, the cutoff or cutoff value can define the threshold value.

[0026] As used herein, "diagnostic information" is information that is useful in determining whether a patient has a disease and/or in classifying (stratifying) the disease into a genotypic or phenotypic category or any category having significance with regard to the prognosis of the disease or its likely response to treatment (either treatment in general or any particular treatment described herein). Similarly, "diagnosis" refers to obtaining or providing any type of diagnostic information, including, but not limited to, whether a patient is likely to have or develop a disease; whether that disease has or is likely to reach a certain state or stage or to exhibit a particular characteristic (e.g., resistance to a therapeutic agent); information related to the nature or classification of a tumor; information related to prognosis (which may also concern resistance); and/or information useful in selecting an appropriate treatment (e.g., selecting a compound of Formula (I), (Ia), a species thereof, or a specified form thereof for a patient identified as having a cancer that is likely to respond to such an inhibitor or other treatment). A patient classified (stratified) according to a method described herein and selected for treatment with a compound of Formula (I), (Ia), a species thereof, or a specified form thereof is likely to respond well to the treatment, meaning that such a patient is more likely to be successfully treated than a patient with the same type of cancer who has not been so identified and is not in the same strata. Available treatments include therapeutic agents and other treatment modalities such as surgery, radiation, etc., and selecting an appropriate treatment encompasses the choice of withholding a particular therapeutic agent; the choice of a dosing regimen; and the choice of employing a combination therapy. Diagnostic information can be used to stratify patients and is thus useful in identifying and classifying a given patient according to, for example, biomarker status. Obtaining diagnostic information can constitute a step in any of the patient stratification methods described herein.

[0027] One of ordinary skill in the art will appreciate that the term "dosage form" may be used to refer to a physically discrete unit of an active agent (e.g., a therapeutic or diagnostic agent) for administration to a patient. Typically, each such unit contains a predetermined quantity of active agent. In some embodiments, such quantity is a unit dosage amount (or a whole fraction thereof) appropriate for administration in accordance with a dosing regimen that has been determined to correlate with a desired or beneficial outcome when administered to a relevant population (i.e., with a therapeutic dosing regimen). Those of ordinary skill in the art appreciate that the total amount of a therapeutic composition or agent administered to a particular patient is determined by one or more attending physicians and may involve administration of multiple dosage forms.

[0028] One of ordinary skill in the art will appreciate that the term "dosing regimen" may be used to refer to a set of unit doses (typically more than one) that are administered individually to a patient, separated by equal or unequal periods of time. A given therapeutic agent typically has a recommended dosing regimen, which may involve one or

more doses, each of which may contain the same unit dose amount or differing amounts. In some embodiments, a dosing regimen comprises a first dose in a first dose amount, followed by one or more additional doses in a second dose amount that is different from the first dose amount. In some embodiments, a dosing regimen is correlated with a desired or beneficial outcome when administered across a relevant population (i.e., the regimen is a therapeutic dosing regimen).

[0029] As used herein, an "effective amount" of an agent (e.g., a chemical compound described herein), such as a compound of Formula (I), refers to an amount that produces or is expected to produce the desired effect for which it is administered. The effective amount will vary depending on factors such as the desired biological endpoint, the pharmacokinetics of the compound administered, the condition being treated, the mode of administration, and characteristics of the patient, as discussed further below and recognized in the art. The term can be applied to therapeutic and prophylactic methods. For example, a therapeutically effective amount is one that reduces the incidence and/or severity of one or more signs or symptoms of the disease. For example, in treating a cancer, an effective amount may reduce the tumor burden, stop tumor growth, inhibit metastasis or prolong patient survival. One of ordinary skill in the art will appreciate that the term does not in fact require successful treatment be achieved in any particular individual. Rather, a therapeutically effective amount is that amount that provides a particular desired pharmacological response in a significant number of patients when administered to patients in need of such treatment. In some embodiments, reference to a therapeutically effective amount may be a reference to an amount administered or an amount measured in one or more specific tissues (e.g., a tissue affected by the disease) or fluids (e.g., blood, saliva, serum, sweat, tears, urine, etc.). Effective amounts may be formulated and/or administered in a single dose or in a plurality of doses, for example, as part of a dosing regimen.

[0030] As used herein, an "enhancer" is a region of genomic DNA that helps regulate the expression of genes; enhancers have been found up to 1 Mbp away from a gene they regulate. An enhancer may overlap, but is often not composed of, gene coding regions. An enhancer is often bound by transcription factors and designated by specific histone marks.

[0031] The term "patient" refers to any organism that is or may be subjected to a diagnostic method described herein or to which a compound described herein, or a specified form thereof, is or may be administered for, e.g., experimental, diagnostic, prophylactic, and/or therapeutic purposes. Typical patients include animals (e.g., mammals such as mice, rats, rabbits, non-human primates, and humans; domesticated animals, such as dogs and cats; and livestock or any other animal of agricultural or commercial value). A patient may be suffering from or susceptible to (i.e., have a higher than average risk of developing) a disease described herein and may display one or more signs or symptoms thereof.

[0032] The term "pharmaceutically acceptable," when applied to a carrier used to formulate a composition disclosed herein (e.g., a pharmaceutical composition), means a carrier that is compatible with the other ingredients of the composition and not deleterious to a patient (e.g., it is non-toxic in the amount required and/or administered (e.g., in a unit dosage form)).

[0033] The term "pharmaceutically acceptable," when applied to a salt, solvate, stereoisomer, tautomer, or isotopic form of a compound described herein, refers to a salt, solvate, stereoisomer, tautomer, or isotopic form that is, within the scope of sound medical judgment, suitable for use in contact with the tissues of humans (e.g., patients) and lower animals (including, but not limited to, mice and rats used in laboratory studies) without unacceptable toxicity, irritation, allergic response and the like, and that can be used in a manner commensurate with a reasonable benefit/risk ratio. Many pharmaceutically acceptable salts are well known in the art (see, e.g., Berge et al., *J. Pharm. Sci.* 66:1-19, 1977). Pharmaceutically acceptable salts of the compounds described herein include those derived from suitable inorganic and organic acids and bases. Examples of pharmaceutically acceptable, nontoxic acid addition salts are salts of an amino group formed with inorganic acids such as hydrochloric acid, hydrobromic acid, phosphoric acid, sulfuric acid, and perchloric acid or with organic acids such as acetic acid, oxalic acid, maleic acid, tartaric acid, citric acid, succinic acid, or malonic acid or by using other methods known in the art such as ion exchange. Other pharmaceutically acceptable salts include adipate, alginate, ascorbate, aspartate, benzenesulfonate, benzoate, bisulfate, borate, butyrate, camphorate, camphorsulfonate, citrate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, formate, fumarate, glucoheptonate, glycerophosphate, gluconate, hemisulfate, heptanoate, hexanoate, hydroiodide, 2-hydroxy-ethanesulfonate, lactobionate, lactate, laurate, lauryl sulfate, MALAT1e, maleate, malonate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, nitrate, oleate, oxalate, palmitate, pamoate, pectinate, persulfate, 3-phenylpropionate, phosphate, picrate, pivalate, propionate, stearate, succinate, sulfate, tartrate, thiocyanate, p toluenesulfonate, undecanoate, valerate salts, and the like. Salts derived from appropriate bases include alkali metal, alkaline earth metal, ammonium and $N^+(C_{1-4}$ alkyl$)_4$ salts. Representative alkali or alkaline earth metal salts include sodium, lithium, potassium, calcium, magnesium, and the like. Further pharmaceutically acceptable salts include, when appropriate, nontoxic ammonium, quaternary ammonium, and amine cations formed using counterions such as halide, hydroxide, carboxylate, sulfate, phosphate, nitrate, lower alkyl sulfonate, and aryl sulfonate.

[0034] As used herein, the term "population" means some number of items (e.g., at least 30, 40, 50, or more) sufficient to reasonably reflect the distribution, in a larger group, of the value being measured in the population. Within the context of the present invention, the population can be a discrete group of humans, laboratory animals, or cells lines (for example) that are identified by at least one common characteristic for the purposes of data collection and analysis. For example, a "population of samples" refers to a plurality of samples that is large enough to reasonably reflect the distribution of a value (e.g., a value related to the state of a biomarker) in a larger group of samples. The items in the population may be biological samples, as described herein. For example, each sample in a population of samples may be cells of a cell line or a biological sample obtained from a patient or a xenograft (e.g., a tumor grown in a mouse by implanting a tumorigenic cell line or a patient sample into the mouse). As noted, individuals within a population can be a discrete group identified by a common characteristic, which can be the same disease (e.g., the same type of

cancer), whether the sample is obtained from living beings suffering from the same type of cancer or a cell line or xenograft representing that cancer.

[0035] The term "prevalence cutoff," as used herein in reference to a specified value (e.g., the strength of a SE associated with a biomarker gene) means the prevalence rank that defines the dividing line between two subsets of a population (e.g., a subset of "responders" and a subset of "non-responders," which, as the names imply include patients who are likely or unlikely, respectively, to experience a beneficial response to a therapeutic agent or agents). Thus, a prevalence rank that is equal to or higher (e.g., a lower percentage value) than the prevalence cutoff defines one subset of the population; a prevalence rank that is lower (e.g., a higher percentage value) than the prevalence cutoff defines the other subset.

[0036] As used herein, the term "prevalence rank" for a specified value (e.g., the mRNA level of a specific biomarker) means the percentage of a population that are equal to or greater than that specific value. For example, a 35% prevalence rank for the amount of mRNA of a specific biomarker in a test cell means that 35% of the population have that level of biomarker mRNA or greater than the test cell.

[0037] As used herein, the terms "prognostic information" and "predictive information" are used to refer to any diagnostic information that may be used to indicate any aspect of the course of a disease or condition either in the absence or presence of treatment. Such information may include, but is not limited to, the average life expectancy of a patient, the likelihood that a patient will survive for a given amount of time (e.g., 6 months, 1 year, 5 years, etc.), the likelihood that a patient will be cured of a disease, the likelihood that a patient's disease will respond to a particular therapy (wherein response may be defined in any of a variety of ways). Diagnostic information can be prognostic or predictive.

[0038] As used herein, the term "rank ordering" means the ordering of values from highest to lowest or from lowest to highest.

[0039] As used herein, the terms "RB-E2F pathway" and "RB-E2F family" refer to a set of genes and the proteins encoded thereby, as the context will make clear, whose expression or activity regulates the activity of the RB gene family and in turn regulates the activity of the E2F family of transcription factors that are required for entry into and progression through the cell cycle. The table below contains a list of genes in the RB-E2F family, an indication of a currently understood function of the encoded proteins and the status of these biomarkers in cancer. We use the shorthand "activated or overexpressed" to indicate that an attribute of a gene (e.g., its copy number or level of expression) or the protein it encodes (e.g., its level of expression or activity) is higher in some patients with certain cancers than it is in healthy subjects. A pre-determined threshold for such activated or overexpressed RB-E2F family members can be determined by comparative analysis and is a level (e.g., mRNA level, protein level, gene copy number, strength of enhancer associated with the gene) that, when found or exceeded in a cancer patient, identifies that patient as a candidate for treatment as described herein. We use the shorthand "inactivated or underexpressed" to indicate that an attribute of a gene (e.g., its copy number, or level of expression) or a protein it encodes (e.g., its level of expression or activity) is lower in some patients with certain cancers than it is in healthy subjects. A pre-determined threshold for such inactivated or underexpressed RB-E2F family members can be determined by comparative analysis and is a level (e.g., mRNA level, protein level, CNV, strength of enhancer associated with the gene) that, when unattained in a cancer patient, identifies that patient as a candidate for treatment as described herein.

Gene	Function	Status in Cancer
E2F1	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F2	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F3	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F4	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F5	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F6	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F7	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
E2F8	E2F family - transcriptional control of cell cycle entry	Activated or overexpressed
RB1	RB family - E2F family inhibition	Inactivated or underexpressed
RBL1	RB family - E2F family inhibition	Inactivated or underexpressed
RBL2	RB family - E2F family inhibition	Inactivated or underexpressed
CDK4	RB family inhibition	Activated or overexpressed
CDK6	RB family inhibition	Activated or overexpressed
CDK2	RB family inhibition	Activated or overexpressed
CCND1	CDK4/6 regulation	Activated or overexpressed

-continued

Gene	Function	Status in Cancer
CCND2	CDK4/6 regulation	Activated or overexpressed
CCND3	CDK4/6 regulation	Activated or overexpressed
CDKN2A	CDK4/6 regulation	Inactivated or underexpressed
CDKN2B	CDK4/6 regulation	Inactivated or underexpressed
CDKN2C	CDK4/6 regulation	Inactivated or underexpressed
CDKN2D	CDK4/6 regulation	Inactivated or underexpressed
CCNE1	CDK2 regulation	Activated or overexpressed
CCNE2	CDK2 regulation	Activated or overexpressed
CDKN1A	CDK2 regulation	Inactivated or underexpressed
CDKN1B	CDK2 regulation	Inactivated or underexpressed
CDKN1C	CDK2 regulation	Inactivated or underexpressed
FBXW7	CCNE regulation	Inactivated or underexpressed

It will be readily apparent to one of ordinary skill in the art that for those genes in the RB-E2F pathway that are activated or overexpressed in cancer, one would select those patients that had (1) an alteration in the DNA encoding such gene that resulted in increased expression (e.g. elevated gene copy number, mutation that led to increased activity, change in methylation that led to increased expression); (2) an epigenetic alteration associated with that gene that resulted in increased expression (e.g. histone methylation or histone acetylation pattern that led to increased expression); or (3) an increase in the level of expression of mRNA or protein encoded by that gene. For those genes in the RB-E2F pathway that are inactivated or under-expressed in cancer, one would select from those patients that had (1) an alteration in the DNA encoding that gene that resulted in decreased expression or activity (e.g. reduced gene copy number, mutation that led to decreased activity or inactivity, change in methylation that led to decreased expression); (2) an epigenetic alteration associated with that gene that resulted in decreased expression (e.g. histone methylation or histone acetylation pattern that led to decreased expression); or (3) an decrease in the level of expression of mRNA or protein encoded by that gene.

[0040] As used herein, a "reference" refers to a standard or control relative to which a comparison is performed. For example, an agent, patient, population, sample, sequence, or value of interest is compared with a reference agent, patient, population, sample, sequence or value. The reference can be analyzed or determined substantially simultaneously with the analysis or determination of the item of interest or it may constitute a historical standard or control, determined at an earlier point in time and optionally embodied in a tangible medium. One of ordinary skill in the art is well trained in selecting appropriate references, which are typically determined or characterized under conditions that are comparable to those encountered by the item of interest. One of ordinary skill in the art will appreciate when sufficient similarities are present to justify reliance on and/or comparison to a particular possible reference as a standard or control.

[0041] As used herein, a "response" to treatment is any beneficial alteration in a patient's condition that results from, or that correlates with, treatment. The alteration may be stabilization of the condition (e.g., inhibition of deterioration that would have taken place in the absence of the treatment), amelioration of, delay of onset of, and/or reduction in frequency of one or more signs or symptoms of the condition, improvement in the prospects for cure of the condition, greater survival time, and etc. A response may be a patient's response or a tumor's response.

[0042] As used herein, when the term "strength" is used to refer to a portion of an enhancer or a SE, it means the area under the curve of the number of H3K27Ac or other genomic marker reads plotted against the length of the genomic DNA segment analyzed. Thus, "strength" is an integration of the signal resulting from measuring the mark at a given base pair over the span of the base pairs defining the region being chosen to measure.

[0043] As used herein, the term "super-enhancer" (SE) refers to a subset of enhancers that contain a disproportionate share of histone marks and/or transcriptional proteins relative to other enhancers in a particular cell or cell type. Genes regulated by SEs are predicted to be of high importance to the function of a cell. SEs are typically determined by rank ordering all of the enhancers in a cell based on strength and determining, using available software such as ROSE (bitbucket.org/young_computation/rose), the subset of enhancers that have significantly higher strength than the median enhancer in the cell (see, e.g., U.S. Pat. No. 9,181,580, which is hereby incorporated by reference herein in its entirety).

[0044] The terms "threshold" and "threshold level" mean a level that defines the dividing line between two subsets of a population (e.g., responders and non-responders). A threshold or threshold level can define a prevalence cutoff or a cutoff value.

[0045] As used herein, the terms "treatment," "treat," and "treating" refer to reversing, alleviating, delaying the onset of, and/or inhibiting the progress of a "pathological condition" (e.g., a disease, such as cancer) described herein. In some embodiments, "treatment," "treat," and "treating" require that signs or symptoms of the disease have developed or have been observed. In other embodiments, treatment may be administered in the absence of signs or symptoms of the disease or condition (e.g., in light of a

history of symptoms and/or in light of genetic or other susceptibility factors). Treatment may also be continued after symptoms have resolved, for example, to delay or inhibit recurrence.

[0046] As the invention relates to compositions and methods for diagnosing and treating patients who have cancer, the terms "active agent," "anti-cancer agent," "pharmaceutical agent," and "therapeutic agent" are used interchangeably (unless the context clearly indicates otherwise) and compounds of Formula (I), (Ia), a species thereof, or a specified form thereof, would be understood by one of ordinary skill in the art as active, anti-cancer, pharmaceutical, or therapeutic agents. As noted, the treatment methods and uses encompass combination therapies/uses in which a compound of Formula (I), (Ia), a species thereof, or a specified form thereof is administered or used in combination with one or more additional agents (e.g., an additional anti-cancer therapeutic), as described herein. In keeping with convention, in any embodiment requiring two agents, we may refer to one as the "first" agent and to the other as the "second" agent to underscore that the first and second agents are distinct from one another. Where three agents are employed, we refer to the "third agent."

[0047] As indicated, each therapeutic method and any diagnostic method that employs a compound of Formula (I), (Ia), a species thereof, or a specified form thereof may also be expressed in terms of use and vice versa. For example, the invention encompasses the use of a compound or composition described herein for the treatment of a disease described herein (e.g., cancer); a compound or composition for use in diagnosing and/or treating or a disease (e.g., cancer); and the use of the compound or composition for the preparation of a medicament for treating a disease described herein (e.g., cancer).

[0048] A patient subjected to a diagnostic or therapeutic method described herein may have a blood cancer, which may also be referred to as a hematopoietic or hematological cancer or malignancy, and any of the methods described herein can entail analyzing a biomarker described herein in a biological sample of, e.g., blood or lymph, obtained from the patient. More specifically and in various embodiments, the blood cancer can be a leukemia such as acute lymphocytic leukemia (ALL; e.g., B cell ALL or T cell ALL), acute myelocytic leukemia (AML; e.g., B cell AML or T cell AML), chronic myelocytic leukemia (CML; e.g., B cell CML or T cell CML), chronic lymphocytic leukemia (CLL; e.g., B cell CLL (e.g., hairy cell leukemia) or T cell CLL), chronic neutrophilic leukemia (CNL), or chronic myelomonocytic leukemia (CMML). The blood cancer can also be a lymphoma such as Hodgkin lymphoma (HL; e.g., B cell HL or T cell HL), non-Hodgkin lymphoma (NHL, which can be deemed aggressive; e.g., B cell NHL or T cell NHL), follicular lymphoma (FL), chronic lymphocytic leukemia/small lymphocytic lymphoma (CLL/SLL), mantle cell lymphoma (MCL), a marginal zone lymphoma (MZL), such as a B cell lymphoma (e.g., splenic marginal zone B cell lymphoma), primary mediastinal B cell lymphoma (e.g., splenic marginal zone B cell lymphoma), primary mediastinal B cell lymphoma, Burkitt lymphoma (BL), lymphoplasmacytic lymphoma (i.e., Waldenstrom's macroglobulinemia), immunoblastic large cell lymphoma, precursor B lymphoblastic lymphoma, or primary central nervous system (CNS) lymphoma. The B cell NHL can be diffuse large cell lymphoma (DLCL; e.g., diffuse large B cell lymphoma (DLBCL; e.g., germinal center B cell-like (GCB) DLBCL or activated B-cell like (ABC) DLBCL)), and the T cell NHL can be precursor T lymphoblastic lymphoma or a peripheral T cell lymphoma (PTCL). In turn, the PTCL can be a cutaneous T cell lymphoma (CTCL) such as mycosis fungoides or Sezary syndrome, angioimmunoblastic T cell lymphoma, extranodal natural killer T cell lymphoma, enteropathy type T cell lymphoma, subcutaneous anniculitis-like T cell lymphoma, or anaplastic large cell lymphoma.

[0049] In other embodiments, the cancer is characterized by a solid tumor considered to be either of its primary location or metastatic. For example, in various embodiments, the cancer or tumor treated or prevented as described herein is an acoustic neuroma; adenocarcinoma; adrenal gland cancer; anal cancer; angiosarcoma (e.g., lymphangiosarcoma, lymphangio-endotheliosarcoma, hemangiosarcoma); appendix cancer; benign monoclonal gammopathy (also known as monoclonal gammopathy of unknown significance (MGUS); biliary cancer (e.g., cholangiocarcinoma); bladder cancer; breast cancer (e.g., adenocarcinoma of the breast, papillary carcinoma of the breast, mammary cancer, medullary carcinoma of the breast; any of which may be present in subjects having a particular profile, such as an HR+(ER+ or PR+), HER2+, HR− (having neither estrogen nor progesterone receptors), a triple negative breast cancer (TNBC; ER−/PR−/HER2−), or a triple-positive breast cancer (ER+/PR+/HER2+); a brain cancer (e.g., meningioma, glioblastoma, glioma (e.g., astrocytoma, oligodendroglioma), medulloblastoma); bronchus cancer; carcinoid tumor, which may be benign; cervical cancer (e.g., cervical adenocarcinoma); choriocarcinoma; chordoma; craniopharyngioma; a cancer present in the large intestine, such as colorectal cancer (CRC, e.g., colon cancer, rectal cancer, or colorectal adenocarcinoma); connective tissue cancer; epithelial carcinoma; ependymoma; endothelio-sarcoma (e.g., Kaposi's sarcoma or multiple idiopathic hemorrhagic sarcoma); endometrial cancer (e.g., uterine cancer, uterine sarcoma); esophageal cancer (e.g., adenocarcinoma of the esophagus, Barrett's adenocarcinoma); Ewing's sarcoma (or other pediatric sarcoma, such as embryonal rhabdomyosarcoma or alveolar rhabdomyosarcoma); eye cancer (e.g., intraocular melanoma, retinoblastoma); familiar hypereosinophilia; gallbladder cancer; gastric cancer (e.g., stomach adenocarcinoma); gastrointestinal stromal tumor (GIST); germ cell cancer; head and neck cancer (e.g., head and neck squamous cell carcinoma, oral cancer (e.g., oral squamous cell carcinoma), throat cancer (e.g., laryngeal cancer, pharyngeal cancer, nasopharyngeal cancer, oropharyngeal cancer)); hypopharynx cancer; inflammatory myofibroblastic tumors; immunocytic amyloidosis; kidney cancer (e.g., nephroblastoma a.k.a. Wilms' tumor, renal cell carcinoma); liver cancer (e.g., hepatocellular cancer (HCC), malignant hepatoma); lung cancer (e.g., bronchogenic carcinoma, small cell lung cancer (SCLC), non-small cell lung cancer (NSCLC), adenocarcinoma, squamous cell carcinoma, or large cell carcinoma of the lung); leiomyosarcoma (LMS); mastocytosis (e.g., systemic mastocytosis); mouth cancer; muscle cancer; myelodys-plastic syndrome (MDS); mesothelioma; myeloproliferative disorder (MPD) (e.g., polycythemia vera (PV), essential thrombocytosis (ET), agnogenic myeloid metaplasia (AMM) a.k.a. myelofibrosis (MF), chronic idiopathic myelofibrosis, hypereosinophilic syndrome (HES)); neuroblastoma; neurofibroma (e.g., neurofibromatosis (NF) type 1 or type 2, schwannomatosis);

neuroendocrine cancer (e.g., gastroentero-pancreatic neuroendocrine tumor (GEP-NET), carcinoid tumor); osteosarcoma (e.g., bone cancer); ovarian cancer (e.g., cystadenocarcinoma, ovarian embryonal carcinoma, ovarian adenocarcinoma, HGSOC, LGSOC, epithelial ovarian cancer (e.g., ovarian clear cell carcinoma or mucinous carcinoa), sex cord stromal tumors (granulosa cell), and endometroid tumors); papillary adenocarcinoma; pancreatic cancer (whether an exocrine tumor (e.g., pancreatic adenocarcinoma, pancreatic ductal adenocarcinoma (PDAC)), intraductal papillary mucinous neoplasm (IPMN), or a neuroendocrine tumor (e.g., PNETs or islet cell tumors); penile cancer (e.g., Paget's disease of the penis and scrotum); pinealoma; primary peritoneal cancer, primitive neuroectodermal tumor (PNT); plasma cell neoplasia; paraneoplastic syndromes; prostate cancer, which may be castration-resistant (e.g., prostate adenocarcinoma); rhabdomyosarcoma; salivary gland cancer; skin cancer (e.g., squamous cell carcinoma (SCC), keratoacanthoma (KA), melanoma, basal cell carcinoma (BCC)); small bowel or small intestine cancer; soft tissue sarcoma (e.g., malignant fibrous histiocytoma (MFH), liposarcoma, malignant peripheral nerve sheath tumor (MPNST), chondrosarcoma, fibrosarcoma, myxosarcoma); sebaceous gland carcinoma; sweat gland carcinoma; synovioma; testicular cancer (e.g., seminoma, testicular embryonal carcinoma); thyroid cancer (e.g., papillary carcinoma of the thyroid, papillary thyroid carcinoma (PTC), medullary thyroid cancer); urethral cancer; vaginal cancer; and vulvar cancer (e.g., Paget's disease of the vulva). We use the term "gastrointestinal (GI) tract cancer" to refer to a cancer present anywhere in the GI tract, including cancers of the mouth, throat, esophagus, stomach, large or small intestine, rectum, and anus. As noted above, the cancer can be a neuroendocrine cancer, and such tumors can be treated as described herein regardless of the organ in which they present. A biomarker described herein can be analyzed in a biological sample containing tumor cells or ctDNA of any of the cancer types just listed. Further, a patient identified by analyzing a biomarker as described herein can be "newly diagnosed" and therefor previously unexposed to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof and, similarly, previously unexposed to a second agent as described herein. We may refer to such a patient as treatment naïve.

[0050] The methods of the invention that concern diagnosing and/or treating a cancer described herein (or use of a compound or compounds for such purposes) may specifically exclude any one or more of the types of cancers described herein. For example, the invention features methods of treating cancer by administering a compound of Formula (I), (Ia), a species thereof, or a specified form thereof, with the proviso that the cancer is not a breast cancer; with the proviso that the cancer is not a breast cancer or a leukemia; with the proviso that the cancer is not a breast cancer, a leukemia, or an ovarian cancer; and so forth, with exclusions selected from any of the cancers listed herein and with the same notion of variable exclusion from lists of elements relevant to other aspects of the invention (e.g., chemical substituents of a compound described herein or components of kits and pharmaceutical compositions). Thus, where elements are presented as lists (e.g., in Markush group format), every possible subgroup of the elements is also disclosed, and any element(s) can be removed from the group.

[0051] In one aspect, the invention features the use of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof in treating cancer in a patient who has been identified by analyzing the biomarker BCL2, RB1, RBL1, RBL2, CDKN2A, CDKN2B, CDKN2C, CDKN2D, CDKN1A, CDKN1B, CDKN1C, or FBXW7 in a biological sample containing cancer cells or ctDNA from the patient. Analyzing the biomarker can include analyzing its sequence to detect a mutation or determining CNA, association with a SE, RNA expression level (e.g., mRNA expression) or another feature described above as indicating the state of the biomarker. A patient identified by analyzing BCL2, RB1, RBL1, RBL2, CDKN2A, CDKN2B, CDKN2C, CDKN2D, CDKN1A, CDKN1B, CDKN1C, or FBXW7 can be: treated with a platinum-based therapeutic agent (e.g., carboplatin, cisplatin, or oxaliplatin) as a second agent; a patient whose cancer has developed resistance to a platinum-based therapeutic agent (e.g., carboplatin, cisplatin, or oxaliplatin); or a patient undergoing treatment with a CDK4/6 inhibitor used alone or in combination with one or more of an aromatase inhibitor, a selective estrogen receptor modulator (SERM), selective estrogen receptor degrader (SERD), or estrogen suppressant, any of which may be selected from the descriptions of such agents provided herein or known in the art. The patient's cancer may have become resistant to the CDK4/6 inhibitor or be at risk of becoming so. In the context of these uses (e.g., where the patient has been identified by analyzing the biomarker BCL2, RB1, RBL1, RBL2, CDKN2A, CDKN2B, CDKN2C, CDKN2D, CDKN1A, CDKN1B, CDKN1C, or FBXW7), the cancer can be a breast cancer (e.g., a triple negative breast cancer (TNBC), HR+, or other type of breast cancer described herein), an ovarian cancer (e.g., HGSOC), a lung cancer (e.g., SCLC, NSCLC or other lung cancer described herein), retinoblastoma, or a blood cancer (e.g., acute myeloid leukemia (AML)).

[0052] The methods of treating such a patient include a step of administering an effective amount of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, optionally within a pharma-ceutical composition described herein and/or according to a dosing regimen described herein.

[0053] In another aspect, the invention features the use of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof in treating cancer in a patient who has been identified by analyzing the biomarker CCNE1, CCNE2, RB1, CDK6, CCND1, CCND2, CCND3, or CCKN2A in a biological sample containing cancer cells or ctDNA from the patient. Analyzing the biomarker can include analyzing its sequence to detect a mutation or determining CNA, association with a SE, RNA expression level (e.g., mRNA expression) or another feature described above as indicating the state of the biomarker. A patient identified by analyzing CCNE1, CCNE2, RB1, CDK6, CCND1, CCND2, CCND3, or CCKN2A can be a patient who has undergone, is presently undergoing, or who will undergo (e.g., has been prescribed) treatment with a Bcl-2 inhibitor, such as venetoclax, a SERM, such as tamoxifen, a SERD, such as fulvestrant, or a PARP inhibitor, such as olaparib or niraparib. In the context of these methods, the patient may have a breast cancer (e.g., TNBC or an HR+ breast cancer), lymphoma, melanoma (e.g., familial melanoma), ovarian cancer (e.g., HGSOC), or pancreatic cancer (e.g., PDAC). For example, where the biomarker is CDKN2A, the patient may have TNBC, PDAC, or HGSOC.

For example, where the biomarker is CCNE1, the patient may have TNBC, HGSOC, melanoma (e.g., familial melanoma), or lymphoma. As noted above, one of ordinary skill will recognize, as is well established in the art, the relationship between a given gene and the protein it encodes. Thus, it will be clear that our reference to, for example, "the biomarker BCL2" encompasses analysis of the biomarker gene BCL2-like 1 and the biomarker protein (BCL2) encoded thereby; "the biomarker CCNE1" encompasses analysis of the biomarker gene CCNE1 and the biomarker protein (cyclin E1) encoded thereby; and so forth. The methods of treating such a patient include a step of administering an effective amount of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, optionally within a pharmaceutical composition described herein and/or according to a dosing regimen described herein.

[0054] In another aspect, the invention features the use of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof in treating cancer in a patient who has been identified by analyzing the biomarker MYC (see Kalkat et al., *Genes* 8(6):151, 2017), CDK1, CDK2, CDK4, CDK17, CDK18, CDK19, CCNA1, CCNB1, ESR-1 or FGFR1 in a biological sample containing cancer cells or ctDNA from the patient. Analyzing the biomarker can include analyzing any mutations within MYC, CDK1, CDK2, CDK4, CDK17, CDK18, CDK19, CCNA1, CCNB1, ESR-1 or FGFR1 or determining CNA, association with a SE, RNA expression level (e.g., mRNA expression) or another feature described above as indicating the state of the biomarker. The patient may have a breast cancer (e.g., TNBC or an ovarian cancer (e.g., HGSOC) and may be resistant to a platinum-based anti-cancer agent, such as carboplatin, cisplatin, or oxaliplatin, resistant to gemcitabine, resistant to a PARP inhibitor, such as olaparib or niraparib, or resistant to a taxane, such as paclitaxel. The methods of treating such a patient include a step of administering an effective amount of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, optionally within a pharmaceutical composition described herein and/or according to a dosing regimen described herein. C-myc encodes at least two phosphoproteins with apparent molecular weights of 62,000 and 66,000 (see Ramsay et al., *Proc. Natl. Acad. Sci.* (*USA*) 81(24):7742-7746, 1984), and it has been determined through H3K27Ac ChIP-seq (ChIP-sequencing) methods that there is a SE locus associated with the MYC gene at chr8:128628088-128778308 (Gencode v19 annotation of the human genome build hg19/GRCh37).

[0055] In another aspect, the invention features the use of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof in treating cancer in a patient who has been identified by analyzing the biomarker CDK7 or CDK9. Analyzing the biomarker can include analyzing any mutations within CDK7 or CDK9 or determining CNA, association with a SE, RNA expression level (e.g., mRNA expression) or another feature described above as indicating the state of the biomarker. Where the biomarker is CDK7 or CDK9, the patient may have a lymphoma and the diagnosing/identifying step may more specifically be based on analysis of CDK7 (e.g., the level of CDK7 mRNA); the patient may have a breast cancer (e.g., TNBC), with the diagnosing/identifying step more specifically based on CDK9 (e.g., the level of CDK9 mRNA); the patient may have a TNBC or a lung cancer (e.g., SCLC), with the diagnosing step more specifically be based CDK19 (e.g., on the level of CDK19 mRNA). The methods of treating such a patient include a step of administering an effective amount of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, optionally within a pharmaceutical composition described herein and/or according to a dosing regimen described herein.

[0056] In another aspect, the invention features the use of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof in treating cancer in a patient who has been identified by analyzing the biomarker BRAF, E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, or E2F8 in a biological sample containing cancer cells or ctDNA from the patient. Analyzing the biomarker can include analyzing its sequence to detect a mutation or determining CNA, association with a SE, RNA expression level (e.g., mRNA expression) or another feature described above as indicating the state of the biomarker. A patient identified by analyzing BRAF, E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, or E2F8 (by virtue of having a feature equal to or above a pre-determined threshold, as described herein) can be a patient who has undergone, is presently undergoing, or who will undergo (e.g., has been prescribed) treatment with a PI3K inhibitor, such as alpelisib or capecitabine, a platinum-based anti-cancer agent, such as carboplatin, cisplatin, or oxaliplatin, or vincristine. In the context of these methods, the patient may have a melanoma, lung cancer (e.g., NSCLC), GI tract cancer (e.g., CRC), thyroid cancer, retinoblastoma, or leukemia (e.g., hairy cell leukemia). The methods of treating such a patient include a step of administering an effective amount of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, optionally within a pharmaceutical composition described herein and/or according to a dosing regimen described herein.

[0057] A compound or other composition described herein (e.g., a pharmaceutical composition comprising a compound of Formula (I), (Ia), a species thereof or specified form thereof) can be administered in a combination therapy (e.g., as defined and further described herein) with a second agent described herein or a plurality thereof (i.e., a patient identified as described herein may be treated with first, second, and third agents). The additional/second agent employed in a combination therapy is most likely to achieve a desired effect for the same disorder (e.g., the same cancer), however it may achieve different effects that aid the patient. Accordingly, the invention features pharmaceutical compositions containing a compound of Formula (I), (Ia), a species thereof, or a specified form thereof (e.g., a pharmaceutically acceptable salt), optionally in a therapeutically effect amount, for use in treating a patient identified as described herein. The pharmaceutical compositions may optionally include any of the additional/second agents described herein and will include a pharmaceutically acceptable carrier. The second/additional agent can be selected from a Bcl-2 inhibitor such as venetoclax, a PARP inhibitor such as olaparib or niraparib, a platinum-based anti-cancer agent such as carboplatin, cisplatin, or oxaliplatin, a taxane such as docetaxel or paclitaxel (or paclitaxel protein-bound (available as Abraxane®)), a CDK4/6 inhibitor such as palbociclib, ribociclib, abemaciclib, or trilaciclib, a selective estrogen receptor modulator (SERM) such as tamoxifen (available under the brand names Nolvadex™ and Soltamox™), raloxifene (available under the brand name Evista™), and toremifene (available as Fareston™) and a selective estrogen receptor

degrader such as fulvestrant (available as Faslodex™), each in a therapeutically effective amount.

[0058] Unless otherwise specified, when employing a combination of a compound of Formula (I), (Ia), a species thereof, or a specified variant thereof and a second therapeutic agent in a method of the invention, the second therapeutic agent can be administered concurrently with, prior to, or subsequent to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof. The second therapeutic pharmaceutical agent may be administered at a dose and/or on a time schedule determined for that pharmaceutical agent. An additional/second therapeutic agent may also be administered together with the compound of Formula (I), (Ia), a species thereof, or a specified form thereof in a single dosage form or administered separately in different dosage forms. In general, and without limitation, it is expected that the second therapeutic agents utilized in combination with a compound of Formula (I), (Ia), a species thereof, or a specified form thereof will be utilized at levels that do not exceed the levels at which they are utilized individually. In some embodiments, the levels of the second therapeutic agent utilized in combination will be lower than those utilized in a monotherapy due to synergistic effects.

[0059] In particular combination therapies for a patient identified as described herein: (a) the cancer is TNBC, an ER+ breast cancer, pancreatic cancer (e.g., PDAC), or a squamous cell cancer of the head or neck and the second agent is a CDK4/6 inhibitor; (b) the cancer is a breast cancer (e.g., TNBC) or an ovarian cancer and the second agent is a PARP inhibitor; (c) the cancer is a leukemia (e.g., AML) and the second agent is a FLT3 inhibitor; (d) the cancer is an ovarian cancer (e.g., HGSOC) and the second agent is a platinum-based anti-cancer agent; (e) the cancer is a breast cancer (e.g., TNBC), a leukemia (e.g., AML), Ewing's sarcoma, or an osteosarcoma and the second agent is a BET inhibitor; (f) the cancer is a breast cancer (e.g., TNBC), a leukemia (e.g., AML), an ovarian cancer (e.g., HGSOC), or a lung cancer (e.g., NSCLC) and the second agent is a Bcl-2 inhibitor. In particular embodiments, the cancer is AML and the second agent is a Bcl-2 inhibitor, such as venetoclax; the cancer is an epithelial ovarian cancer, a fallopian tube cancer, a primary peritoneal cancer, a triple negative breast cancer or a $Her2^{+}/ER^{-}/PR^{-}$ breast cancer and the second agent is a PARP inhibitor, such as olaparib or niraparib; the cancer is an ovarian cancer and the second agent is a platinum-based anti-cancer agent, such as carboplatin, cisplatin, or oxaliplatin. As noted above, and regardless of the biomarker analyzed or the type of cancer in question, a method of treatment can either be carried out on an identified patient without an explicit step of analyzing the biomarker or with an explicit step in which the biomarker is analyzed (e.g., by obtaining a biological sample from a patient).

[0060] With regard to combination therapies, a patient identified as described herein can be treated with a combination of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof and one or more of a second agent that can be, but is not limited to, a Bcl-2 inhibitor such as APG-1252, APG-2575, BP1002 (prexigebersen), the antisense oligonucleotide known as oblimersen (G3139), S55746/BCL201, or venetoclax (e.g., venetoclax tablets marketed as Venclexta®); a CDK9 inhibitor such as alvocidib/DSP-2033/flavopiridol, AT7519, AZD5576, BAY1251152, BAY1143572, CYC065, nanoflavopiridol, NVP2, seliciclib (CYC202), TG02, TP-1287, VS2-370 or voruciclib (formerly P1446A-05); a hormone receptor (e.g., estrogen receptor) degradation agent, such as fulvestrant (e.g., marketed as Faslodex® and others); a Flt3 (FMS-like tyrosine kinase 3) inhibitor such as CDX-301, CG'806, CT053PTSA, crenolanib (e.g., crenolanib besylate), ENMD-2076, FF-10101-01, FLYSYN, gilteritinib (ASP2215), HM43239, lestautinib, ponatinib (e.g., marketed as Iclusig®, previously AP24534), NMS-088, sorafenib (e.g., marketed as Nexavar®), sunitinib, pacritinib, pexidartinib/PLX3397, quizartinib, midostaurin (e.g., marketed as Rydapt®), SEL24, SKI-G-801, or SKLB1028; a PARP inhibitor such as olaparib (e.g., marketed as Lynparza®), rucaparib (e.g., marketed as Rubraca®), talazoparib (e.g., marketed as Talzenna®), veliparib (ABT-888), or niraparib (e.g., marketed as Zejula®); a BET inhibitor such as ABBV-075, BAY-299, BAY-1238097, BMS-986158, CPI-0610, CPI-203, FT-1101, GS-5829, GSK-2820151, GSK-525762, I-BET151, I-BET762, INCB054329, JQ1, MS436, OTX015, PFI-1, PLX51107, RVX2135, TEN-010, ZEN-3694, or a compound disclosed in U.S. application Ser. No. 12/810,564 (now U.S. Pat. No. 8,476,260), which is hereby incorporated herein by reference in its entirety; a platinum-based therapeutic agent such as cisplatin, oxaliplatin (e.g., marketed as Eloxatin®), nedaplatin, carboplatin (e.g., marketed as Paraplatin®), phenanthriplatin, picoplatin, satraplatin (JM216), or triplatin tetranitrate; a CDK4/6 inhibitor such as BPI-1178, G1T38, palbociclib (e.g., marketed as Ibrance®), ribociclib (e.g., marketed as Kisqali®), ON 123300, trilaciclib, or abemaciclib (e.g., marketed as Verzenio®); a MEK inhibitor such as trametinib (e.g., marketed as Mekinist®), cobimetinib (available as Cotellic®), or binimetinib (Braftovi®), useful in combination with a compound of Formula (I), (Ia), a species thereof, or specified form thereof, in treating, e.g., melanoma); or a phosphoinositide 3-kinase (PI3 kinase) inhibitor, optionally of Class I (e.g., Class IA) and/or optionally directed against a specific PI3K isoform. The PI3K inhibitor can be apitolisib (GDC-0980), idelalisib (e.g., marketed as Zydelig®), copanlisib (e.g., marketed as Aliqopa®), duvelisib (e.g., marketed as Copiktra®), pictilisib (GDC-0941), or alpelisib (e.g., marketed as Piqray®). In other embodiments, the additional/second agent can be capecitabine (e.g., marketed as Xeloda®). Such PI3K inhibitors can be combined with a compound of Formula (I), (Ia), a species thereof or specified form thereof in treating, e.g., HR+ breast cancer, TNBC, lymphoma (e.g., follicular lymphoma or non-Hodgkin lymphoma), or leukemia (e.g., CLL). In other embodiments, the additional/second agent can be gemcitabine (combined with a compound of the invention to treat, e.g., TNBC, CRC, SCLC, or a pancreatic cancer (e.g., PDAC)). In other embodiments, the additional/second agent can be an antimetabolite, such as the pyrimidine analog 5-fluorouracil (5-FU), which may be used in combination with a compound of Formula (I), (Ia), a species thereof, or a specified form thereof, and one or more of leucovorin, methotrexate, or oxaliplatin. In other embodiments, the additional/second agent can be an aromatase inhibitor, such as exemestane or anastrasole. In other embodiments, the additional/second agent is an inhibitor of the PI3K/AKT/mTOR pathway (e.g., gedatolisib). In one embodiment, the methods encompass the use of or administration of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, to a patient identified as described herein, in combination with a MEK

inhibitor, such as trametinib (available as Mekinist®), cobimetinib (available as Cotellic®), or binimetinib (available as Braftovi®)

[0061] APG-1252 is a dual Bcl-2/Bcl-xL inhibitor that has shown promise in early clinical trials when patients having SCLC or another solid tumor were dosed between 10-400 mg (e.g., 160 mg) intravenously twice weekly for three weeks in a 28-day cycle (see Lakhani et al., *J. Clin. Oncol.* 36:15_suppl, 2594, and ClinicalTrials.gov identifier NCT03080311). APG-2575 is a Bcl-2 selective inhibitor that has shown promise in preclinical studies of FL and DLBCL in combination with ibrutinib (see Fang et al., AACR Annual Meeting 2019, *Cancer Res.* 79(13 Suppl):Abstract No. 2058) and has begun clinical trials as a single-agent treatment for patients with blood cancers; in a dose escalation study, patients are given 20 mg, once daily, by mouth, for four consecutive weeks as one cycle. Escalations to 50, 100, 200, 400, 600 and 800 mg are planned to identify the MTD (see ClinicalTrials.gov identifier NCT03537482). BP1002 is an uncharged P-ethoxy antisense oligodeoxynucleotide targeted against Bcl-2 mRNA that may have fewer adverse effects than other antisense analogs and has shown promise in inhibiting the growth of human lymphoma cell lines inclubated with BP1002 for four days and of CJ cells (transformed FL cells) implanted into SCID mice (see Ashizawa et al., AACR Annual Meeting 2017, *Cancer Res.* 77(13 Suppl):Abstract No. 5091). BP1002 has also been administered in combination with cytarabine (LDAC) to patients having AML (see ClinicalTrials.gov identifier NCT04072458). S55746/BCL201 is an orally available, selective Bcl-2 inhibitor that, in mice, demonstrated antitumor efficacy in two blood cancer xenograft models (Casara et al., *Oncotarget* 9(28):20075-88, 2018). A phase I dose-escalation study was designed to administer film-coated tablets containing 50 or 100 mg of S55746, in doses up to 1500 mg, to patients with CLL or a B cell NHL including FL, MCL, DLBCL, SLL, MZL, and MM (see ClinicalTrials.gov identifier NCT02920697). Venetoclax tablets have been approved for treating adult patients with CLL or SLL and, in combination with azacytidine, or decitabine, or low-dose cytarabine, for treating newly-diagnosed AML in patients who are at least 75 years old or who have comorbidities that preclude the use of intensive induction chemotherapy. Dosing for CLL/SLL can follow the five-week ramp-up schedule and dosing for AML can follow the four-day ramp-up, both described in the product insert, together with other pertinent information (see also U.S. Pat. Nos. 8,546,399; 9,174,982; and 9,539,251, which are hereby incorporated by reference in their entireties). Alvocidib was studied in combination with cytarabine/mitoxantrone or cytarabine/daunorubicin in patients with AML, with the details of administration being available at ClinicalTrials.gov with the identifier NCT03563560 (see also Yeh et al., *Oncotarget* 6(5):2667-2679, 2015, Morales et al., *Cell Cycle* 15(4):519-527, 2016, and Zeidner et al., *Haematologica* 100(9):1172-1179, 2015). AT7519 has been administered in a dose escalation format to eligible patients having refractory solid tumors. While there was some evidence of clinical activity, the appearance of QTc prolongation precluded further development at the dose schedule described by Mahadevan et al. (*J. Clin. Oncol*. ASCO Abstract No. 3533; see also Santo et al., *Oncogene* 29:2325-2336, 2010, describing the preclinical activity of AT7519 in MM). AZD5576 induced apoptosis in breast and lung cancer cell lines at the nanomolar level (see Li et al., *Bioorg. Med. Chem. Lett.* 27(15):3231-3237, 2017) and has been examined alone and in combination with acalabrutinib for the treatment of NHL (see AACR 2017 Abstract No. 4295). BAY1251152 was the subject of a phase I clinical trial to characterize the MTD in patients with advanced blood cancers; the agent was infused weekly in 21-day cycles (see ClinicalTrials.gov identifier NCT02745743; see also Luecking et al., AACR 2017 Abstract No. 984). Voruciclib is a clinical stage oral CDK9 inhibitor that represses MCL-1 and sensitizes high-risk DLBCL to BCL2 inhibition. Dey et al. (*Scientific Reports* 7:18007, 2017) suggest that the combination of voruciclib and venetoclax is promising for a subset of high-risk DLBCL patients (see also ClinicalTrials.gov identifier NCT03547115). Fulvestrant has been approved for administration to postmenopausal women with advanced hormone receptor (HR)-positive, HER2-negative breast cancer, with HR-positive metastatic breast cancer whose disease progressed after treatment with other anti-estrogen therapies, and in combination with palbociclib (Ibrance®). Fulvestrant is administered by intramuscular injection at 500 or 250 mg (the lower dose being recommended for patients with moderate hepatic impairment) on days 1, 15, and 29, and once monthly thereafter (see the product insert for additional information; see also U.S. Pat. Nos. 6,744,122; 7,456,160; 8,329,680; and 8,466,139, each of which are hereby incorporated by reference herein in their entireties). Ponatinib has been administered in clinical trials to patients with CML or ALL (see ClinicalTrials.gov identifiers NCT0066092072, NCT012074401973, NCT02467270, NCT03709017, NCT02448095, NCT03678454, and NCT02398825) as well as solid tumors, such as biliary cancer and NSCLC (NCT02265341, NCT02272998, NCT01813734, NCT02265341, NCT02272998, NCT01813734, NCT02265341, NCT02272998, NCT01813734, NCT01935336, NCT03171389, and NCT03704688; see also the review article by Tan et al., *Onco. Targets Ther.* 12:635-645, 2019). Additional information regarding the dosing regimen can be found in the product insert; see also U.S. Pat. Nos. 8,114,874; 9,029,533; and 9,493,470, each of which is hereby incorporated by reference herein in its entirety. Sorafenib has been approved for the treatment of kidney and liver cancers, AML, and radioactive iodine resistant advanced thyroid cancer, and a clinical trial was initiated in patients with desmoid-type fibromatosis (see ClinicalTrials.gov identifier NCT02066181). Information regarding dosage can be found in the product insert, which advises administration of two, 400 mg tablets twice daily; see also U.S. Pat. Nos. 7,235,576; 7,351,834; 7,897,623; 8,124,630; 8,618,141; 8,841,330; 8,877,933; and 9,737,488, each of which is hereby incorporated by reference herein in its entirety. Midostaurin has been administered to patients having AML, MDS, or systemic mastocytosis, and has been found to significantly prolong survival of FLT3-mutated AML patients when combined with conventional induction and consolidation therapies (see Stone et al., ASH 57th Annual Meeting, 2015 and Gallogly et al., *Ther. Adv. Hematol.* 8(9):245-251, 2017; din see also the product insert, ClinicalTrials.gov identifier NCT03512197, and U.S. Pat. Nos. 7,973,031; 8,222,244; and 8,575,146, each of which is hereby incorporated by reference herein in its entirety. Alpelisib is a kinase inhibitor indicated in combination with fulvestrant for the treatment of postmenopausal women, and men, with HR+/HER2−/

PIK3CA-mutated, advanced or metastatic breast cancer as deteted by an FDA-approved test following progression on or after an endocrine-based regimen. The recommended dose is 300 mg (two 150 mg tablets) taken orally once daily with food, which, as for all chemotherapeutic agents, may be interrupted, reduced, or discontinued to manage adverse reactions. Paclitaxel is supplied as a nonaqueous solution intended for dilution with a suitable parenteral fluid prior to intravenous infusion. Under the brand name Taxol®, it is supplied in 30 mg, 100 mg, and 300 mg vials and can be used in a combination therapy described herein to treat a variety of cancers, including those of the bladder, breast, esophagus, fallopian tube or ovary, lung, skin (melanoma), and prostate. Palbociclib has been approved for use in HR+/HER2− advanced or metastatic breast cancer at a recommended dose of 125 mg daily, by mouth. It can be used to treat a patient as identified herein with a compound of Formula (I), (Ia), a species thereof, or a specified form thereof, either alone or in combination with an aromatase inhibitor or fulvestrant. The information provided here and publicly available can be used to practice the methods and uses of the invention. In case of doubt, the invention encompasses combination therapies that require a compound of the invention or a specified form thereof and any one or more additional/second agents, which may be administered at or below a dosage currently approved for single use (e.g., as described above), to a patient as described herein. Triplet combinations include a compound of Formula (I), (Ia), a species thereof, or a specified form thereof with: alpelisib and fulvestrant or alpelisib and a taxane (for, e.g., treating NSCLC).

[0062] Where the combination therapy employs a compound of the invention and: a CDK4/6 inhibitor, the patient can have a breast cancer (e.g., TNBC or an ER+ breast cancer), pancreatic cancer, lung cancer (e.g., SCLC or NSCLC), or squamous cell cancer of the head and neck; a CDK9 inhibitor, the patient can have a breast cancer and, more specifically, a $Her2^{+}/ER^{-}/PR^{-}$ breast cancer; a Flt3 inhibitor (e.g., midostaurin), the patient can have a hematological cancer (e.g., AML); a BET inhibitor, the patient can have a hematological cancer (e.g., AML), a breast cancer (e.g., TNBC), an osteosarcoma or Ewing's Sarcoma; a Bcl-2 inhibitor (e.g., venetoclax), the patient can have a breast cancer (e.g., TNBC), an ovarian cancer, a lung cancer (e.g., NSCLC) or a hematological cancer (e.g., AML); or a PARP inhibitor (e.g., niraparib or olaparib), the patient can have a breast cancer (e.g., TNBC or $Her2^{+}/ER^{-}/PR^{-}$ breast cancer), an ovarian cancer (e.g., an epithelial ovarian cancer), a fallopian tube cancer, or a primary peritoneal cancer.

[0063] In some aspects relating to using RB-E2F pathway genes (or the proteins they encode) as biomarkers, the invention provides a method of treating a patient having a cancer and identified as described herein, which comprises administering to a patient identified as having either (a) a level of CCNE1 mRNA or protein in the cancer equal to or above a pre-determined threshold; and/or (b) a level of RB1 mRNA or protein in the cancer equal to or below a pre-determined threshold, an effective amount of a CDK7 inhibitor of Formula (I). In some aspects of these embodiments, the method further comprises determining a level of RB1 and/or CCNE1 mRNA or protein present in a sample of cancer cells from the patient. In various embodiments, the human patient is diagnosed as having a cancer sensitive to a CDK7 inhibitor responsive to the determination; is suffering from ovarian cancer; or is suffering from a breast cancer (e.g., TNBC or an HR^{+} breast cancer). In some embodiments, a compound of Formula (I), (Ia), a species thereof or a specified form thereof is co-administered with a PARP inhibitor. In some embodiments, the compound of Formula (I), (Ia), a species thereof or a specified form thereof is co-administered with a SERM (e.g., tamoxifen, raloxifene, or toremifene), a SERD such fulvestrant, or an agent that inhibits the production of estrogen (e.g., an aromatase inhibitor such as anastrozole (available as Arimidex®), exemestane (available as Aromasin®), and letrozole (available as Femara®), optionally to treat a cancer that is refractory to palbociclib. Our data indicate that a compound of Formula (I), (Ia), a species thereof, or a specified form thereof (e.g., Compound 101) can induce deep and sustained TGI in combination with fulvestrant in palbociclib-resistant (PBR) ER+ breast cancer PDX models. Further, based on data with Compound 101, we believe compounds of the invention can resensitize palbociclib- and fulvestrant-resistant (PBR/FSR) ER+ breast cancer PDX tumors to fulvestrant treatment. In other embodiments, the invention provides methods of treating cancer in a patient identified as described herein by administering to the patient a combination of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof and a platinum-based standard of care (SOC) anti-cancer agent for such cancer or a taxane. The cancer can be an ovarian cancer and the SOC anti-cancer agent can be a platinum-based anti-cancer agent (e.g., carboplatin, cisplatin, or oxaliplatin). In some embodiments, the human patient is, has been determined to be, or has become resistant (after some initial responsiveness) to the platinum-based anti-cancer agent when administered as either a monotherapy or in combination with an anti-cancer agent other than a CDK7 inhibitor. In some aspects of this embodiment, the human patient is determined to have become resistant to the platinum-based anti-cancer agent when administered as a monotherapy or in combination with an anti-cancer agent other than a CDK7 inhibitor after some initial efficacy of that prior treatment. In some aspects of this embodiment, the SOC anti-cancer agent is a taxane (e.g., paclitaxel).

[0064] The invention also provides methods of treating a biomarker-identified HR^{+} breast cancer in a human patient selected on the basis of being resistant to treatment with a CDK4/6 inhibitor comprising the step of administering to the patient a compound of Formula (I), (Ia), a species thereof, or a specified form thereof. In some embodiments, prior to administration of the compound of Formula (I), (Ia), a species thereof, or a specified form thereof, the patient is, has been determined to be, or has become resistant (after some initial responsiveness) to a prior treatment with a CDK4/6 inhibitor alone or in combination with another SOC agent for breast cancer other than a CDK7 inhibitor, such as an aromatase inhibitor (e.g., letrozole, anastrozole) or a SERM (e.g., tamoxifen, raloxifene, or toremifene), SERD (e.g., fulvestrant), or estrogen suppressant such as anastrozole (available as Arimidex®), exemestane (available as Aromasin®), or letrozole (available as Femara®). In other words, the identified patient is selected for treatment with a compound of Formula (I), (Ia), a species thereof, or a specified form thereof on the basis of being resistant to prior treatment with a CDK4/6 inhibitor alone or in combination with another SOC agent for breast cancer other than a CDK7 inhibitor. In some embodiments, the compound of Formula

(I), (Ia), a species thereof, or a specified form thereof is co-administered with another SOC agent, such as an aromatase inhibitor (e.g. anastrozole, exemestane, or letrozole) and/or a SERM or SERD, e.g., as described herein, or a second line treatment after failure on an aromatase inhibitor or fulvestrant. In some embodiments, prior to administration of the compound of Formula (I), (Ia), a species thereof, or a specified form thereof, the patient is, has been determined to be, or has become resistant (after some initial responsiveness) to treatment with a CDK4/6 inhibitor alone or in combination with another SOC agent for breast cancer other than a CDK7 inhibitor, such as an aromatase inhibitor (e.g., anastrozole, exemestane, or letrozole), or a SERM or SERD such as tamoxifen or fulvestrant; and the compound of Formula (I), (Ia), a species thereof, or a specified form thereof is co-administered with a SOC agent for breast cancer (e.g., a second line treatment after failure of an aromatase inhibitor or a SERM or SERD such as tamoxifen or fulvestrant.

[0065] An enhancer or SE can be identified by various methods known in the art (see Hinsz et al., *Cell,* 155:934-947, 2013; McKeown et al., *Cancer Discov.,* 7(10):1136-53, 2017; and PCT/US2013/066957, each of which are hereby incorporated herein by reference in their entireties). Identifying a SE can be achieved by obtaining a biological sample from a patient (e.g., from a biopsy or other source, as described herein). The important metrics for enhancer measurement occur in two dimensions: along the length of the DNA over which genomic markers (e.g., H3K27Ac) are contiguously detected and the compiled incidence of genomic marker at each base pair along that span of DNA, the compiled incidence constituting the magnitude. The measurement of the area under the curve ("AUC") resulting from integration of length and magnitude analyses determines the strength of the enhancer. The strength of the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2) SEs relative to an appropriate reference can be used to diagnose (stratify) a patient and thereby determine whether a patient is likely to respond well to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof. It will be readily apparent to one of ordinary skill in the art, particularly in view of the instant specification, that if the length of DNA over which the genomic markers is detected is the same for each of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2) and the reference/control, then the ratio of the magnitude of the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2) SE relative to the control will be equivalent to the strength and may also be used to determine whether a patient will be responsive to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof. The strength of the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2) SE in a cell can be normalized before comparing it to other samples. Normalization is achieved by comparison to a region in the same cell known to comprise a ubiquitous SE or enhancer that is present at similar levels in all cells. One example of such a ubiquitous super-enhancer region is the MALAT1 super-enhancer locus (chr11:65263724-65266724) (genome build hg19).

[0066] It has been determined through H3K27Ac ChIP-seq (ChIP-sequencing) methods that there is a SE locus associated with the CDK18 gene at chr1:205399084-205515396; a SE locus associated with the CDK19 gene at chr6:110803523-110896277; a SE locus associated with the CCNE1 gene at chr19:30418503-30441450; and a SE locus associated with the FGFR1 gene at chr8:38233326-38595483. All loci are based on the Gencode v19 annotation of the human genome build hg19/GRCh37.

[0067] ChIP-seq is used to analyze protein interactions with DNA by combining chromatin immunoprecipitation (ChIP) with massively parallel DNA sequencing to identify the binding sites of DNA-associated proteins. It can be used to map global binding sites precisely for any protein of interest. Previously, ChIP-on-chip was the most common technique utilized to study these protein-DNA relations. Successful ChIP-seq is dependent on many factors including sonication strength and method, buffer compositions, antibody quality, and cell number (see, e.g., Furey, Nature Reviews Genetics 13:840-852, 2012); Metzker, Nature Reviews Genetics 11:31-46, 2010; and Park, Nature Reviews Genetics 10:669-680, 2009). Genomic markers other than H3K27Ac that can be used to identify SEs using ChIP-seq include P300, CBP, BRD2, BRD3, BRD4, components of the mediator complex (Loven et al., *Cell,* 153 (2):320-334, 2013), histone 3 lysine 4 monomethylated (H3K4me1), and other tissue-specific enhancer tied transcription factors (Smith and Shilatifard, *Nature Struct. Mol. Biol.,* 21(3):210-219, 2014; and Pott and Lieb, *Nature Genetics,* 47(1):8-12, 2015). Quantification of enhancer strength and identification of SEs can be determined using SE scores (McKeown et al., *Cancer Discov.* 7(10):1136-1153, 2017; DOI: 10.1158/2159-8290.CD-17-0399).

[0068] In some instances, H3K27Ac or other marker ChIP-seq data SE maps of the entire genome of a cell line or a patient sample already exist. One would then simply determine whether the strength or ordinal rank of the enhancer or SE in such maps at the chr8:128628088-128778308 (genome build hg19) locus was equal to or above the pre-determined threshold level. In some embodiments, one would simply determine whether the strength, or ordinal rank of the enhancer or super-enhancer in such maps at the chr1:205399084-205515396 (genome build hg19) locus was equal to or above the pre-determined threshold level.

[0069] It should be understood that the specific chromosomal location of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2) and MALAT1 may differ for different genome builds and/or for different cell types. The

same is true for BCL2-like 1, CDK7, CDK9, CDKN2A, and RB1 or another E2F pathway member that is underexpressed in cancer (E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CCND1, CCND2, CCND3, CCNE1, or CCNE2). However, one skilled in the art, particularly in view of the instant specification, can determine such different locations by locating in such other genome builds specific sequences corresponding to the loci in genome build hg 19.

[0070] Other methods that can be used to identify SEs in the context of the present methods include chromatin immunoprecipitation (Delmore et al., *Cell,* 146(6):904-917, 2011), chip array (ChIP-chip), and chromatin immunoprecipitation followed by qPCR (ChIP-qPCR) using the same immunoprecipitated genomic markers and oligonucleotide sequences that hybridize to the chr8:128628088-128778308 (genome build hg19) MYC locus or chr1:205399084-205515396 (genome build hg19) CDK18 locus (for example). In the case of ChIP-chip, the signal is typically detected by intensity fluorescence resulting from hybridization of a probe and input assay sample as with other array-based technologies. For ChIP-qPCR, a dye that becomes fluorescent after intercalating the double stranded DNA generated in the PCR reaction is used to measure amplification of the template.

[0071] In some embodiments, determination of whether a cell has a BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) SE strength equal to or above a requisite threshold level is achieved by comparing BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength in a test cell to the corresponding BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) strength in a population of cell samples, wherein each of the cell samples is obtained from a different source (e.g., a different patient, a different cell line, a different xenograft) reflecting the same disease to be treated. In some embodiments, only primary tumor cell samples from patients are used to determine the threshold level. In some aspects of these embodiments, at least some of the samples in the population will have been tested for responsiveness to a specific CDK7 inhibitor (e.g., a compound of Formula (I), (Ia), a species thereof or a specified form thereof) to establish: (a) the lowest M BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength of a sample in the population that responds to that specific compound ("lowest responder"); and, optionally, (b) the highest BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength of a sample in the population that does not respond to that specific compound ("highest non-responder"). In these embodiments, a cutoff of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength above which a test cell would be considered responsive to that specific compound is set: i) equal to or up to 5% above the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength in the lowest responder in the population; or ii) equal to or up to 5% above the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength in the highest non-responder in the population; or iii) a value in between the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength of the lowest responder and the highest non-responder in the population.

[0072] In the above embodiments, not all of the samples in a population necessarily are to be tested for responsiveness to a specific CDK7 inhibitor (e.g., a compound of Formula (I), (Ia), a species thereof or a specified form thereof), but all samples are measured for BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PICKCA, or certain genes encoding an E2F pathway member (see above) enhancer strength. In some embodiments, the samples are rank ordered based on M BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength. The choice of which of the three methods set forth above to use to establish the cutoff will depend upon the difference in BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength between the lowest responder and the highest non-responder in the population and whether the goal is to minimize the number of false positives or to minimize the chance of missing a potentially responsive sample or patient. When the difference between the lowest responder and highest non-responder is large (e.g., when there are many samples not tested for responsiveness that fall between the lowest responder and the highest non-responder in a rank ordering of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength), the cutoff is typically set equal to or is up to 5% above the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength in the lowest responder in the population. This cutoff maximizes the number of potential responders. When this difference is small (e.g., when there are few or no samples untested for responsiveness that fall between the lowest responder and the highest non-responder in a rank ordering of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength), the cutoff is typically set to a value in between the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19,

CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength of the lowest responder and the highest non-responder. This cutoff minimizes the number of false positives. When the highest non-responder has a BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see the Table herein) enhancer strength that is greater than the lowest responder, the cutoff is typically set to a value equal to or up to 5% above the BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) enhancer strength in the highest non-responder in the population. This method also minimizes the number of false positives.

[0073] In some embodiments, the methods discussed above can be employed to simply determine if a diseased cell (e.g., a cancer cell) from a patient has a SE associated with a biomarker gene described herein (e.g., BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) or a protein encoded thereby). The presence of the SE indicates that the patient is likely to respond well to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof. The cell is determined to have a SE associated with the biomarker (e.g., BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) or a protein encoded thereby) when the enhancer has a strength that is equal to or above the enhancer associated with MALAT-1. In alternate embodiments, the cell is determined to have a SE associated with BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) when the BRAF−, MYC−, CDK1−, CDK2−, CDK4−, CDK6−, CDK17−, CDK18−, CDK19−, CCNA1−, CCNB1−, CCNE1−, ESR−1−, FGFR1−, PIK3CA−, or certain genes encoding an E2F pathway member- (see above) associated enhancer has a strength that is at least 10-fold greater than the median strength of all of the enhancers in the cell. In other embodiments, the cell is determined to have a SE associated with an aforementioned gene when the gene-associated enhancer has a strength that is above the point where the slope of the tangent is 1 in a rank-ordered graph of strength of each of the enhancers in the cell.

[0074] For any biomarker (e.g., in embodiments involving CDK18), the cutoff value for enhancer strength can be converted to a prevalence cutoff, which can then be applied to biomarker RNA levels (e.g., CDK18 mRNA) levels to determine a mRNA cutoff value in a given mRNA assay.

[0075] In some embodiments, biomarker mRNA levels in a patient (as assessed, e.g., in a biological sample obtained from the patient) are compared, using the same assay, to the same gene of interest/biomarker mRNA levels in a population of patients having the same disease or condition to identify likely responders to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof. Analogous comparisons can be made when another feature of the biomarker is selected for analysis (e.g., its copy number, chromosomal location, primary RNA level, or expressed protein level). In embodiments where a biomarker (e.g., CDK18, CDK19, and CCNE1) correlates with (e.g., is one whose mRNA expression correlates with) responsiveness to a compound of the invention, at least some of the samples in the population will have been tested for responsiveness to the inhibitor (e.g., a compound of Formula (I), (Ia), a species thereof, or specified form thereof) to establish: (a) the lowest level (e.g., mRNA level) in a sample in the population that responds to that specific compound ("lowest mRNA responder"); and, optionally, (b) the highest level (e.g., highest mRNA level) in a sample in the population that does not respond to that specific compound ("highest mRNA non-responder"). In these embodiments, a cutoff of biomarker mRNA level above which a test cell would be considered responsive to that specific compound is set: i) equal to or up to 5% above the level (e.g., the mRNA level) in the lowest mRNA responder in the population; or ii) equal to or up to 5% above the level (e.g., the mRNA level) in the highest mRNA non-responder in the population; or iii) a value in between the level (e.g., mRNA level) of the lowest responder (e.g., lowest mRNA responder) and the highest responder (e.g., highest mRNA) non-responder in the population.

[0076] In embodiments where mRNA levels positively correlate with sensitivity to a compound of Formula (I), (Ia), a species thereof or a specified form thereof, not all of the samples in a population need to be tested for responsiveness to the compound, but all samples are measured for the gene of interest mRNA levels. In some embodiments, the samples are rank ordered based on gene of interest mRNA levels. The choice of which of the three methods set forth above to use to establish the cutoff will depend upon the difference in gene of interest mRNA levels between the lowest mRNA responder and the highest mRNA non-responder in the population and whether the cutoff is designed to minimize false positives or maximize the potential number of responders. When this difference is large (e.g., when there are many samples not tested for responsiveness that fall between the lowest mRNA responder and the highest mRNA non-responder in a rank ordering of mRNA levels), the cutoff is typically set equal to or up to 5% above the mRNA level in the lowest mRNA responder. When this difference is small (e.g., when there are few or no samples untested for responsiveness that fall between the lowest mRNA responder and the highest mRNA non-responder in a rank ordering of mRNA levels), the cutoff is typically set to a value in between the mRNA levels of the lowest mRNA responder and the highest mRNA non-responder. When the highest mRNA non-responder has a mRNA level that is greater than the lowest mRNA responder, the cutoff is typically set to a value equal to or up to 5% above the mRNA levels in the highest mRNA non-responder in the population.

[0077] In embodiments where a biomarker is one whose mRNA expression inversely correlates with responsiveness to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof (i.e., BCL-xL, CDK7, CDK9, or an RB1 family member), at least some of the samples in the population will have been tested for responsiveness to the compound in order to establish: (a) the highest mRNA level of a sample in the population that responds to that specific compound ("highest mRNA responder"); and, optionally, (b) the lowest mRNA level of a sample in the population that does not respond to that specific compound ("lowest mRNA

non-responder"). In these embodiments, a cutoff of mRNA level above which a test cell would be considered responsive to that specific compound is set: i) equal to or up to 5% below the mRNA level in the highest mRNA responder in the population; or ii) equal to or up to 5% below the mRNA level in the lowest mRNA non-responder in the population; or iii) a value in between the mRNA level of the lowest mRNA non-responder and the highest mRNA responder and in the population.

[0078] In embodiments where mRNA levels inversely correlate with sensitivity to a compound of the invention, not all of the samples in a population need to be tested for responsiveness to the compound, but all samples are measured for the gene of interest mRNA levels. In some embodiments, the samples are rank ordered based on gene of interest mRNA levels. The choice of which of the three methods set forth above to use to establish the cutoff will depend upon the difference in gene of interest mRNA levels between the highest mRNA responder and the lowest mRNA non-responder in the population and whether the cutoff is designed to minimize false positives or maximize the potential number of responders. When this difference is large (e.g., when there are many samples not tested for responsiveness that fall between the highest mRNA responder and the lowest mRNA non-responder in a rank ordering of mRNA levels), the cutoff is typically set equal to or up to 5% below the mRNA level in the highest mRNA responder. When this difference is small (e.g., when there are few or no samples untested for responsiveness that fall between the highest mRNA responder and the lowest mRNA non-responder in a rank ordering of mRNA levels), the cutoff is typically set to a value in between the mRNA levels of the highest mRNA responder and the lowest mRNA non-responder. When the highest mRNA responder has a mRNA level that is lower than the lowest mRNA responder, the cutoff is typically set to a value equal to or up to 5% below the mRNA levels in the lowest mRNA non-responder in the population.

[0079] In embodiments involving CDK18, the cutoff for CDK18 mRNA levels may be determined using the prevalence cutoff established based on CDK18 enhancer strength, as described above. In some embodiments, a population is measured for mRNA levels and the prior determined prevalence cutoff is applied to that population to determine an mRNA cutoff level. In some aspects of these embodiments a rank-order standard curve of CDK18 mRNA levels in a population is created, and the pre-determined prevalence cutoff is applied to that standard curve to determine the CDK18 mRNA cutoff level.

[0080] In some aspects of embodiments where a test cell or sample is compared to a population, the cutoff mRNA level value(s) obtained for the population is converted to a prevalence rank and the mRNA level cutoff is expressed as a percent of the population having the cutoff value or higher, e.g., a prevalence cutoff. Without being bound by theory, applicants believe that the prevalence rank of a test sample and the prevalence cutoff in a population will be similar regardless of the methodology used to determine mRNA levels.

[0081] A patient can be identified as likely to respond well to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof if the state of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above) as determined by, e.g., RNA (e.g., mRNA levels) in a biological sample from the patient) corresponds to (e.g., is equal to or greater than) a prevalence rank in a population of about 80%, 79%, 78%, 77%, 76%, 75%, 74%, 73%, 72%, 71%, 70%, 69%, 68%, 67%, 66%, 65%, 64%, 63%, 62%, 61%, 60%, 59%, 58%, 57%, 56%, 55%, 54%, 43%, 42%, 51%, 50%, 49%, 48%, 47%, 46%, 45%, 44%, 43%, 42%, 41%, 40%, 39%, 38%, 37%, 36%, 35%, 34%, 33%, 32%, 31%, 30%, 29%, 28%, 27%, 26%, 25%, 24%, 23%, 22%, 21%, or 20% as determined by the state of BRAF, MYC, CDK1, CDK2, CDK4, CDK6, CDK7, CDK17, CDK18, CDK19, CCNA1, CCNB1, CCNE1, ESR-1, FGFR1, PIK3CA, or certain genes encoding an E2F pathway member (see above), respectively, determined by assessing the same parameter (e.g., mRNA level(s)) in the population. A patient can be identified as likely to respond well to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof if the state of BCL2-like 1, CDK7, CDK9, CDKN2A, and RB (as determined by, e.g., RNA (e.g., mRNA) levels or corresponding protein levels in a biological sample from the patient) is below a prevalence rank in a population of about 80%, 79%, 78%, 77%, 76%, 75%, 74%, 73%, 72%, 71%, 70%, 69%, 68%, 67%, 66%, 65%, 64%, 63%, 62%, 61%, 60%, 59%, 58%, 57%, 56%, 55%, 54%, 43%, 42%, 51%, 50%, 49%, 48%, 47%, 46%, 45%, 44%, 43%, 42%, 41%, 40%, 39%, 38%, 37%, 36%, 35%, 34%, 33%, 32%, 31%, 30%, 29%, 28%, 27%, 26%, 25%, 24%, 23%, 22%, 21%, or 20% as determined by the state of BCL2-like 1, CDK7, CDK9, CDKN2A, and RB, respectively, determined by assessing the same parameter (e.g., mRNA level(s)) in the population. In some embodiments, the cutoff value or threshold is established based on the biomarker (e.g., mRNA) prevalence value.

[0082] In still other embodiments, a population may be divided into three groups: responders, partial responders and non-responders, and two cutoff values (or thresholds) or prevalence cutoffs (or thresholds) are set or determined. The partial responder group may include responders and non-responders as well as those patients whose response to a compound of Formula (I), (Ia), a species thereof, or a specified form thereof was not as high as the responder group. This type of stratification may be particularly useful when, in a population, the highest mRNA non-responder has an mRNA level that is greater than that of the lowest mRNA responder. In this scenario, for CDK18 or CDK19, the cutoff level or prevalence cutoff between responders and partial responders is set equal to or up to 5% above the CDK18 or CDK19 mRNA level of the highest CDK18 or CDK19 mRNA non-responder; and the cutoff level or prevalence cutoff between partial responders and non-responders is set equal to or up to 5% below the CDK18 or CDK19 mRNA level of the lowest CDK18 or CDK19 mRNA responder. For BCL-xL, CDK7 or CDK9, this type of stratification may be useful when the highest mRNA responder has a mRNA level that is lower than that of the lowest mRNA non-responder. In this scenario, for BCL-xL, CDK7 or CDK9, the cutoff level or prevalence cutoff between responders and partial responders is set equal to or up to 5% below the mRNA level of the lowest mRNA non-responder; and the cutoff level or prevalence cutoff between partial responders and non-responders is set equal to or up to 5% above the mRNA level of the highest mRNA responder. The determination of whether partial responders should be administered a com-

pound of Formula (I), (Ia), a species thereof, or a specified form thereof will depend upon the judgment of the treating physician and/or approval by a regulatory agency.

[0083] Methods that can be used to quantify specific RNA sequences in a biological sample are known in the art and include, but are not limited to, fluorescent hybridization such as utilized in services and products provided by NanoString Technologies, array based technology (Affymetrix), reverse transcriptase qPCR as with SYBR® Green (Life Technologies) or TaqMan® technology (Life Technologies), RNA sequencing (e.g., RNA-seq), RNA hybridization and signal amplification as utilized with RNAscope® (Advanced Cell Diagnostics), or Northern blot. In some cases, mRNA expression values for various genes in various cell types are publicly available (see, e.g., broadinstitute.org/ccle; and Barretina et al., *Nature,* 483:603-607, 2012).

[0084] In some embodiments, the state of a biomarker (as assessed, for example, by the level of RNA transcripts) in both the test biological sample (i.e., from the patient) and the reference standard or all members of a population is normalized before comparison. Normalization involves adjusting the determined level of an RNA transcript by comparison to either another RNA transcript that is native to and present at equivalent levels in both of the cells (e.g., GADPH mRNA, 18S RNA), or to a fixed level of exogenous RNA that is "spiked" into samples of each of the cells prior to super-enhancer strength determination (Lovén et al., *Cell,* 151(3):476-82, 2012; Kanno et al., *BMC Genomics* 7:64, 2006; Van de Peppel et al., *EMBO Rep.,* 4:387-93, 2003).

[0085] A patient (e.g., a human) suffering from a cancer described herein and identified as described herein based on biomarker status may have been determined to be resistant (or to be acquiring resistance after some initial efficacy) to a therapeutic agent that was administered prior to the compound of Formula (I), (Ia), a species thereof, or a specified form thereof. For example, the cancer may be resistant or refractory to a chemotherapeutic agent, e.g., a Bcl-2 inhibitor such as venetoclax, a BET inhibitor, a CDK4/6 inhibitor such as palbociclib or ribociclib, a CDK9 inhibitor such as alvocidib, a FLT3 inhibitor, a MEK inhibitor such a trametinib, a PARP inhibitor, such as olaparib or niraparib, a PI3K inhibitor, such as alpelisib or capecitabine, a platinum-based therapeutic agent such as cisplatin, oxaliplatin, nedaplatin, carboplatin, phenanthriplatin, picoplatin, satraplatin (JM216), or triplatin tetranitrate, a SERM, such as tamoxifen faloxifene, or toremifene, or a steroid receptor degrading agent (e.g., a SERD, such as fulvestrant). Combination therapies including one or more of these agents are also within the scope of the invention and are discussed further herein. For example, in one embodiment, the methods encompass the use of or administration of a compound of Formula (I), (Ia), a species thereof or a specified form thereof, in combination with a SERD, such as fulvestrant, to treat a cancer (e.g., a breast cancer (e.g., an ER+ breast cancer)) resistant to treatment with a CDK4/6 inhibitor such as palbociclib or ribociclib.

[0086] In some embodiments, the prior therapeutic agent may be a platinum-based anti-cancer agent administered as a monotherapy or in combination with a SOC agent. Most cancer patients eventually develop resistance to platinum-based therapies by one or more of the following mechanisms: (i) molecular alterations in cell membrane transport proteins decrease uptake of the platinum agent; (ii) molecular alterations in apoptotic signaling pathways that prevent a cell from inducing cell death; (iii) molecular alterations of certain genes (e.g. BRCA1/2, CHEK1, CHEK2, RAD51) that restore the ability of the cell to repair platinum agent-induced DNA damage. K. N. Yamamoto et al., 2014, PloS ONE 9(8):e105724. The term "molecular alterations" includes increased or decreased mRNA expression from the genes involved in these functions; increased or decreased expression of protein from such genes; and mutations in the mRNA/proteins expressed from those genes.

[0087] Resistance is typically determined by disease progression (e.g., an increase in tumor size and/or numbers) during treatment or a decrease in the rate of shrinkage of a tumor. In some instances, a patient will be considered to have become resistant to a platinum-based agent when the patient's cancer responds or stabilizes while on treatment, but which progresses within 1-6 months following treatment with the agent. Resistance can occur after any number of treatments with platinum agents. In some instances, disease progression occurs during, or within 1 month of completing treatment. In this case, the patient is considered to have never demonstrated a response to the agent. This is also referred to a being "refractory" to the treatment. Resistance may also be determined by a treating physician when the platinum agent is no longer considered to be an effective treatment for the cancer.

[0088] In some embodiments, the patient is or has been determined to be resistant to treatment with a CDK4/6 inhibitor administered as a monotherapy or in combination with a SOC agent.

[0089] CDK4/6 inhibitors in cancer (e.g., HR^+ breast cancer) are known to block entry into S phase of the cell cycle by inducing G1 arrest. Resistance to CDK4/6 inhibitors in cancer (e.g., HR^+ metastatic breast cancer) has been shown to be mediated, in part, by molecular alterations that: 1) enhance CDK4/6 activity, such as amplifications of CDK6, CCND1, or FGFR1 (Formisano et al., *SABCS* 2017, Publication Number GS6-05; Cruz et al., *SABCS* 2017 Publication Number PD4-05), or 2) reactivate cell cycle entry downstream of CDK4/6, such as RB1 loss and CCNE1 amplification (Condorelli, *Ann. Oncol.,* 2017 PMID: 29236940; Herrera-Abreu, *Cancer Research* 2016 PMID: 27020857).

[0090] Unlike platinum-based agents which are typically administered for a period of time followed by a period without treatment, CDK4/6 inhibitors, such as palbociclib, ribociclib or abemaciclib, are administered until disease progression is observed. In some instances, a patient will be considered to have become resistant to a CDK4/6 inhibitor when the patient's cancer initially responds or stabilizes while on treatment, but which ultimately begins to progress while still on treatment. In some instances, a patient will be considered to be resistant (or refractory) to treatment with a CDK4/6 inhibitor if the cancer progresses during treatment without demonstrating any significant response or stabilization. Resistance may also be determined by a treating physician when the CDK4/6 inhibitor is no longer considered to be an effective treatment for the cancer.

[0091] In case of any doubt, any of the specified forms of a compound of Formula (I), (Ia), or a species thereof can be included in a pharmaceutical composition used or administered (e.g., in an effective amount (e.g., a therapeutically effective amount) according to a method of the invention. Pharmaceutical compositions useful in the methods of the invention can be prepared by relevant methods known in the

art of pharmacology. In general, such preparatory methods include the steps of bringing a compound described herein, including compounds of Formula (I), (Ia), a species thereof, or a specified form thereof (e.g., a pharmaceutically acceptable salt, solvate, stereoisomer, tautomer, or isotopic form thereof) into association with a carrier and/or one or more other active ingredients (e.g., a second agent described herein) and/or accessory ingredients, and then, if necessary and/or desirable, shaping and/or packaging the product into a desired single-dose or multi-dose unit (e.g., for oral dosing). The accessory ingredient may improve the bioavailability of a compound of Formula (I), (Ia), a species thereof, or a specified form thereof, may reduce and/or modify its metabolism, may inhibit its excretion, and/or may modify its distribution within the body (e.g., by targeting a diseased tissue (e.g., a tumor). The pharmaceutical compositions can be packaged in various ways, including in bulk containers and as single unit doses (containing, e.g., discrete, predetermined amounts of the active agent) or a plurality thereof, and any such packaged or divided dosage forms are within the scope of the invention. The amount of the active ingredient can be equal to the amount constituting a unit dosage or a convenient fraction of a dosage such as, for example, one-half or one-third of a dose.

[0092] Relative amounts of the active agent/ingredient, the pharmaceutically acceptable carrier(s), and/or any additional ingredients in a pharmaceutical composition of the invention can vary, depending upon the identity, size, and/or condition of the subject treated and further depending upon the route by which the composition is to be administered and the disease to be treated. By way of example, the composition may comprise between about 0.1% and 99.9% (w/w or w/v) of an active agent/ingredient.

[0093] Pharmaceutically acceptable carriers useful in the manufacture of the pharmaceutical compositions described herein are well known in the art of pharmaceutical formulation and include inert diluents, dispersing and/or granulating agents, surface active agents and/or emulsifiers, disintegrating agents, binding agents, preservatives, buffering agents, lubricating agents, and/or oils. Pharmaceutically acceptable carriers useful in the manufacture of the pharmaceutical compositions described herein include, but are not limited to, ion exchangers, alumina, aluminum stearate, lecithin, serum proteins, such as human serum albumin, buffer substances such as phosphates, glycine, sorbic acid, potassium sorbate, partial glyceride mixtures of saturated vegetable fatty acids, water, salts or electrolytes, such as protamine sulfate, disodium hydrogen phosphate, potassium hydrogen phosphate, sodium chloride, zinc salts, colloidal silica, magnesium trisilicate, polyvinyl pyrrolidone, cellulose-based substances, polyethylene glycol, sodium carboxymethylcellulose, polyacrylates, waxes, polyethylene-polyoxypropylene-block polymers, polyethylene glycol and wool fat.

[0094] Pharmaceutical compositions used as described herein may be administered orally. Such orally acceptable dosage forms may be solid (e.g., a capsule, tablet, sachet, powder, granule, and orally dispersible film) or liquid (e.g., an ampoule, semi-solid, syrup, suspension, or solution (e.g., aqueous suspensions or dispersions and solutions). In the case of tablets, carriers commonly used include lactose and corn starch. Lubricating agents, such as magnesium stearate, can also be included. In the case of capsules, useful diluents include lactose and dried cornstarch. When aqueous suspensions are formulated, the active agent/ingredient can be combined with emulsifying and suspending agents. In any oral formulation, sweetening, flavoring or coloring agents may also be added. In any of the various embodiments described herein, an oral formulation can be formulated for immediate release or sustained/delayed release and may be coated or uncoated. A provided composition can also be micro-encapsulated.

[0095] Compositions suitable for buccal or sublingual administration include tablets, lozenges and pastilles. Formulations can also be prepared for subcutaneous, intravenous, intramuscular, intraocular, intravitreal, intra-articular, intra-synovial, intrasternal, intrathecal, intrahepatic, intraperitoneal intralesional and by intracranial injection or infusion techniques. Preferably, the compositions are administered orally, subcutaneously, intraperitoneally or intravenously. Sterile injectable forms of the compositions of this invention may be aqueous or oleaginous suspension. These suspensions may be formulated according to techniques known in the art using suitable dispersing or wetting agents and suspending agents. The sterile injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally acceptable diluent or solvent, for example as a solution in 1,3-butanediol. Among the acceptable vehicles and solvents that may be employed are water, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium.

[0096] Although the descriptions of pharmaceutical compositions provided herein are principally directed to pharmaceutical compositions which are suitable for administration to humans, it will be understood by one of ordinary skill in the art that such compositions are generally suitable for administration to animals of all sorts. Modification of pharmaceutical compositions suitable for administration to humans in order to render the compositions suitable for administration to various animals is well understood, and the ordinarily skilled veterinary pharmacologist can design and/or perform such modification.

[0097] Compounds described herein are typically formulated in dosage unit form, e.g., single unit dosage form, for ease of administration and uniformity of dosage. The specific therapeutically or prophylactically effective dose level for any particular subject or organism will depend upon a variety of factors including the disease being treated and the severity of the disorder; the activity of the specific active ingredient employed; the specific composition employed; the age, body weight, general health, sex and diet of the subject; the time of administration, route of administration, and rate of excretion of the specific active ingredient employed; the duration of the treatment; drugs used in combination or coincidental with the specific active ingredient employed; and like factors well known in the medical arts.

[0098] The amount of a compound required to achieve an optimum clinical outcome can vary from subject to subject, depending, for example, on species, age, and general condition of a subject, severity of the side effects, cancer to be treated, identity of the particular compound(s) to be administered, and mode of administration. The desired dosage can be delivered two or three times a day, once a day, every other day, every third day, every week, every two weeks, every three weeks, or every four weeks. In certain embodiments, the desired dosage can be delivered using multiple admin-

istrations (e.g., two, three, four, five, six, seven, eight, nine, ten, eleven, twelve, thirteen, fourteen, or more administrations).

[0099] In certain embodiments, an effective amount of a compound for administration one or more times a day (e.g., once) to a 70 kg adult human may comprise about 1-100 mg, about 1-50 mg, about 1-35 mg (e.g., about 1-5, 1-10, 1-15, 1-20, 1-25, or 1-30 mg), about 2-20 mg, about 3-15 mg or about 10-30 mg (e.g., 10-20 or 10-25 mg). Here, and wherever ranges are referenced, the end points are included. The dosages provided in this disclosure can be scaled for patients of differing weights or body surface and may be expressed per m^2 of the patient's body surface. In certain embodiments, compositions of the invention may be administered once per day. The dosage of a compound of Formula (I), (Ia), a species thereof or a specified form thereof (e.g., a salt thereof) can be about 1-100 mg, about 1-50 mg, about 1-25 mg, about 2-20 mg, about 5-15 mg, about 10-15 mg, or about 13-14 mg. In certain embodiments, a composition of the invention may be administered twice per day. In some embodiments, the dosage of a compound of Formula I or a subgenus or species thereof for each administration is about 0.5 mg to about 50 mg, about 0.5 mg to about 25 mg, about 0.5 mg to about 1 mg, about 1 mg to about 10 mg, about 1 mg to about 5 mg, about 3 mg to about 5 mg, or about 4 mg to about 5 mg.

[0100] As noted, the invention provides pharmaceutical kits configured for treating cancer that include a compound of Formula (I), (Ia), a species thereof, or a specified form thereof and, optionally, an additional/second therapeutic agent (e.g., second and third agents) selected from the second/additional agents described herein. For example, the second/additional agent can be: (a) a Bcl-2 inhibitor or dual Bcl-2/BCL-xL inhibitor, (b) a CDK inhibitor (e.g., a CDK4/6, CDK7, or CDK9 inhibitor), (c) a Flt3 inhibitor, (d) a PARP inhibitor, (e) a BET inhibitor, (f) an aromatase inhibitor, (g) a SERM, SERD, or estrogen suppressant, (h) a MEK inhibitor, or (i) a PI3 kinase inhibitor, which, as noted, may be selected from those disclosed herein. The kit can include optional instructions for: (a) reconstituting (if necessary) a compound of Formula (I), (Ia), a species thereof, or a specified form thereof and/or the second therapeutic agent; (b) administering each of the compound of Formula (I), (Ia), a species thereof, or a specified form thereof and/or the second therapeutic agent; and/or (c) a list of specific cancers for which the kit is useful or diagnostic methods by which they may be determined. The kits can also include any type of paraphernalia useful in administering the active agent(s) contained therein (e.g., tubing, syringes, needles, sterile dressings, tape, and the like). Such kits, whether configured to deliver a monotherapy consisting of a compound of Formula (I), (Ia), a species thereof, or a specified variant thereof, or a combination therapy including an additional/second agent selected from any one of those described herein, find utility in the diagnostic and treatment methods described herein. In some instances, the first and second agents will be in separate vessels (e.g., with the first agent confined to a first container and the second agent confined to a second container) and/or formulated in a pharmaceutically acceptable composition, optionally in unit dosage form, that includes the first agent, the second agent, and a pharmaceutically acceptable carrier. In some instances, the kits include a written insert or label with instructions to use the two (or more) therapeutic agents in a patient suffering from a cancer (e.g., as described herein) and identified as amenable to treatment by a method described herein. The instructions may be adhered or otherwise attached to a vessel or vessels comprising the therapeutic agents. Alternatively, the instructions and the vessel(s) can be separate from one another but present together in a single kit, package, box, bag, or other type of container. Alternatively, or in addition, the written instructions can specify and direct the user to a website or other media. The instructions in the kit will typically be mandated or recommended by a governmental agency approving the therapeutic use of the combination (e.g., in a patient population identified as described herein). The instructions may optionally comprise dosing information for each therapeutic agent, the types of cancer for which treatment of the combination was approved or may be prescribed, physicochemical information about each of the therapeutics, pharmacokinetic information about each of the therapeutics, drug-drug interaction information, or diagnostic information (e.g., based on a biomarker or a method of identifying a patient for treatment as described herein). The kits of the invention can also include reagents useful in the diagnostic methods described herein.

EXAMPLES

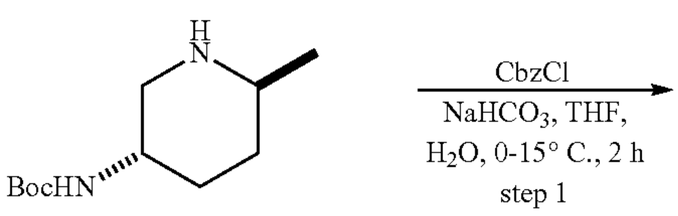
[0101] The compounds described herein can be prepared from readily available starting materials and according to the synthetic protocols described below. Alternatively, one of ordinary skill in the art may readily modify the disclosed protocols. For example, it will be appreciated that where process conditions (e.g., reaction temperatures, reaction times, mole ratios of reactants, solvents, pressures, etc.) are given, other process conditions can also be used. Additionally, and as will be apparent to one of ordinary skill in the art, protecting groups may be used to prevent certain functional groups from undergoing undesired reactions. The choice of a suitable protecting group for a particular functional group as well as suitable conditions for protection and deprotection are well known in the art. For example, numerous protecting groups and guidance for their introduction and removal are disclosed by Greene et al. (*Protecting Groups in Organic Synthesis*, Second Edition, Wiley, New York, 1991, and references cited therein).

[0102] Also included within the Examples below are studies demonstrating that daily oral dosing of Compound 101 can induce dose-dependent TGI in ovarian and breast tumor xenografts, with tumor regression observed at doses as low as one-fifth of MTD. We also observed Compound 101 plasma exposures that were dose proportional without accumulation upon repeated dialing dosing at therapeutic doses in mice (1-6 mg/kg). Compound 101 induced rapid (4 hours) and sustained (24 hours) dose-dependent pharmacodynamic responses in xenograft tumor tissue that correlated with TGI, supporting but not mandating a QD dosing regimen. We also observed tumor regressions that were sustained after treatment with Compound 101 was discontinued, at well-tolerated doses in multiple PDX models from SCLC, TNBC, and HGSOC. Sustained regressions were associated with RB pathway alterations. In a study of combination therapy, Compound 101 induced robust anti-tumor activity in combination with fulvestrant in treatment-resistant PDX models of ER+ breast cancer. Collectively, these studies highlight the broad potential for compounds of the invention in a variety of solid tumor types.

Example 1: Synthesis of Benzyl (2R, 5R)-5-amino-2-methyl-piperidine-1-carboxylate and benzyl (2S, 5S)-5-amino-2-methyl-piperidine-1-carboxylate

Step 1: Benzyl 5-(tert-butoxycarbonylamino)-2-methyl-piperidine-1-carboxylate

[0103]



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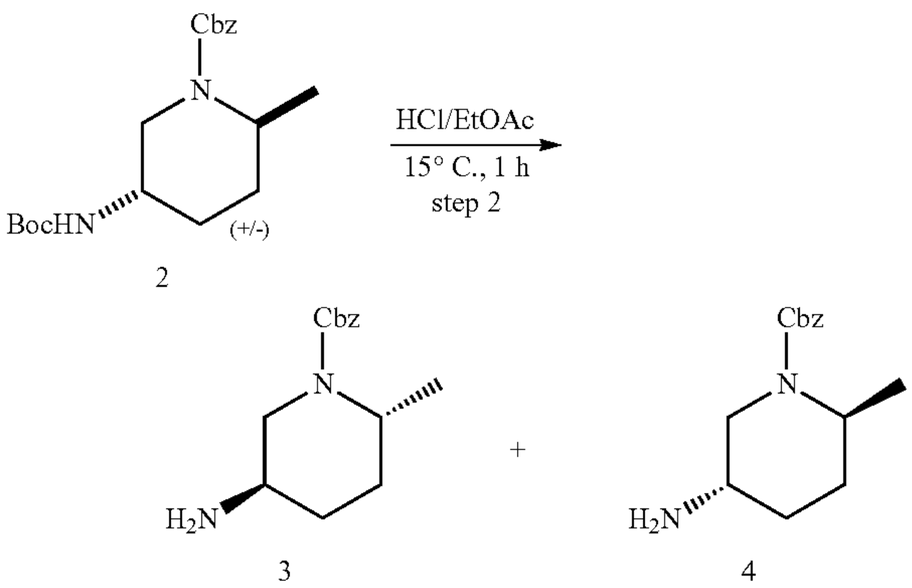
[0104] To a solution containing commercially available racemic trans tert-butyl N-(6-methyl-3-piperidyl)carbamate (5 g, 23.33 mmol, 1 eq,) and $NaHCO_3$ (13.72 g, 163.32 mmol, 7 eq) in tetrahydrofuran (THF; 50 mL) and H_2O (50 mL), we added CbzCl (5.97 g, 35.00 mmol, 4.98 mL, 1.5 eq) dropwise at 0° C. The mixture was stirred at 15° C. for 2 hours then poured into water (50 mL) and extracted with ethyl acetate (EtOAc; 50 mL×3). The combined organic layer was washed with brine (50 mL×3), dried over Na_2SO_4, and filtered. The filtrate was concentrated under reduced pressure, and the residue was purified by medium pressure liquid chromatography (MPLC; SiO_2, PE:EtOAc=5:1 to 1:1) to give the title compound as a yellow solid (9.7 g, 18.04 mmol, 77.32% yield, 64.8% purity).

Step 2: Benzyl (2R, 5R)-5-amino-2-methyl-piperidine-1-carboxylate and benzyl (2S, 5S)-5-amino-2-methyl-piperidine-1-carboxylate

[0105]



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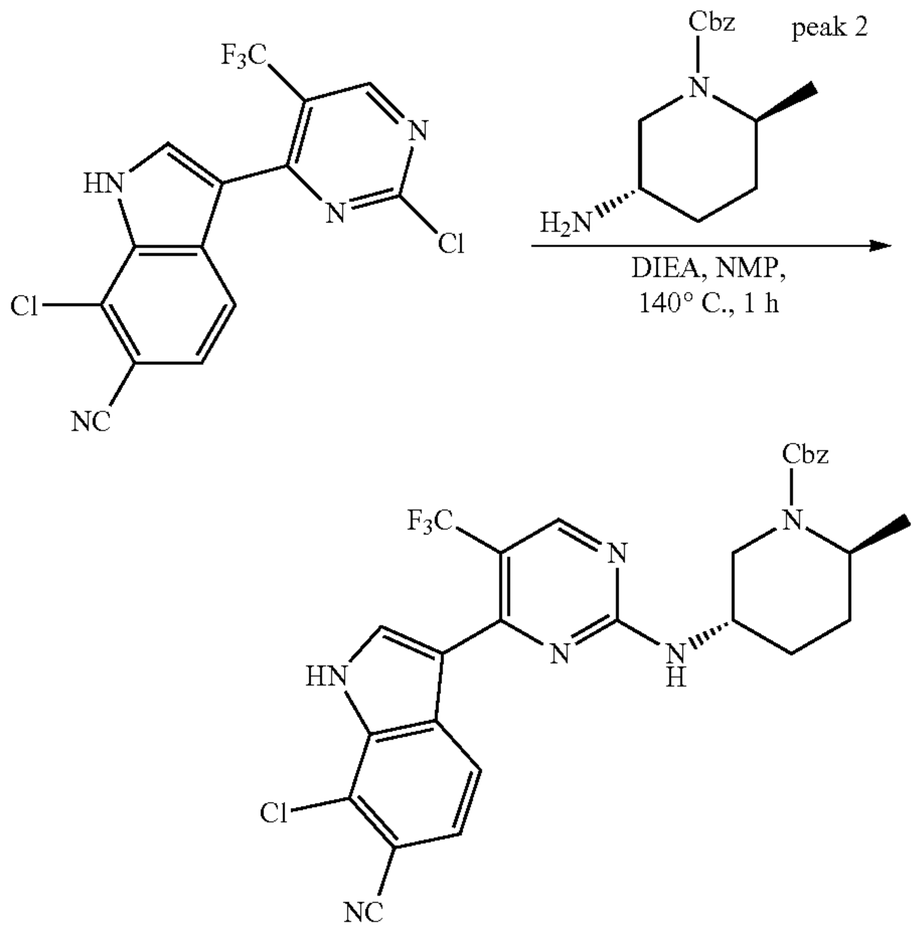
4

[0106] To a mixture of racemic trans benzyl 5-(tert-butoxycarbonylamino)-2-methyl-piperidine-1-carboxylate (9.7 g, 27.84 mmol, 1 eq) in EtOAc (100 mL) we added HCl/EtOAc (15 mL, 4 M), and the mixture was stirred at 15° C. for 1 hour. We then filtered the mixture and collected the filter cake. The solid was dissolved in methanol (MeOH; 15 mL) and the pH was adjusted to 9 using a strongly acidic cation exchange resin (here, AMBERLYST® A21) before the mixture was filtered and the filtrate was concentrated. The residue was separated by supercritical fluid chromatography (SFC; column: marketed by Daicel as CHIRALCEL® (chemicals for use in chromatography) ODH (250 mm×30 mm, 5 μm); mobile phase: [0.1% $NH_3.H_2O$ MeOH]; B %: 28%-28%, 16 min) to afford title compound 1 (1.9 g, SFC: Rt=2.264 min, 93.2% ee, peak 1) and title compound 2 (1.9 g, SFC: Rt=2.593 min, 98.6% ee, peak 2), both as light yellow solids. Peak 1 is structure 3. Peak 2 is structure 4.

Example 2: Synthesis of 7-dimethylphosphoryl-3-[2-[[(3S, 6S)-6-methyl-3-piper-idyl]amino]-5-(trifluoromethyl)pyrimidin-4-yl]-1H-indole-6-carbonitrile (Compound 100)

Step 1: Benzyl (2S, 5S)-5-[[4-(7-chloro-6-cyano-1H-indol-3-yl)-5-(trifluoromethyl) pyrimidin-2-yl] amino]-2-methyl-piperidine-1-carboxylate

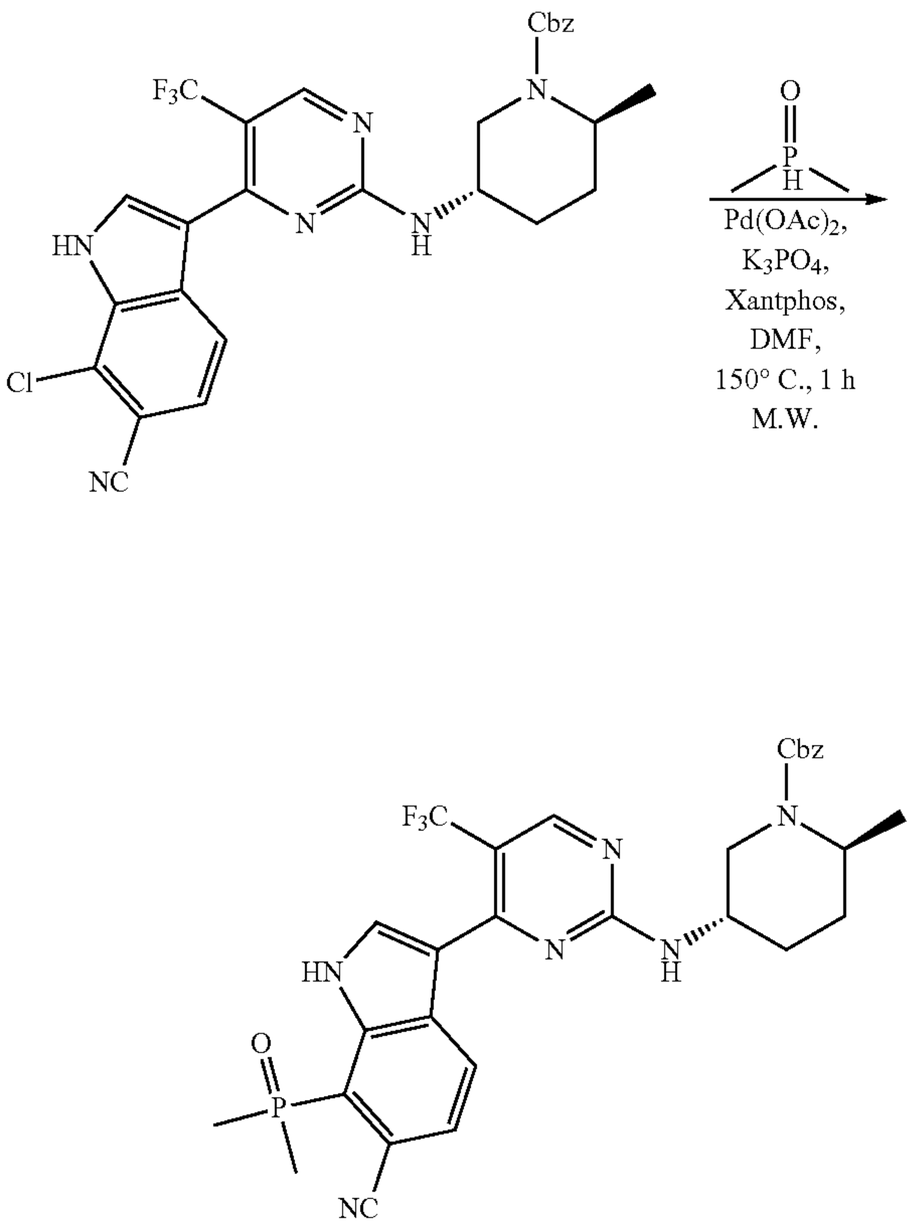
[0107]



[0108] We stirred a mixture of 7-chloro-3-[2-chloro-5-(trifluoromethyl)pyrimidin-4-yl]-1H-indole-6-carbonitrile (0.81 g, 2.27 mmol, 1 eq), benzyl (2S,5S)-5-amino-2-methyl-piperidine-1-carboxylate (732.20 mg, 2.95 mmol, 1.3 eq) and N,N-diisopropylethylamine (DIEA or DIPEA; 879.41 mg, 6.80 mmol, 1.19 mL, 3 eq) in N-methyl-2-pyrrolidone (NMP; 8 mL) at 140° C. for 1 hour. The reaction mixture was diluted with H_2O (100 mL) and extracted with EtOAc (50 mL×2). The combined organic layers were washed with brine (100 mL×2), dried over Na_2SO_4, filtered, and concentrated under reduced pressure to give a residue that was purified by column chromatography (SiO_2, petroleum ether/ethyl acetate=10:1 to 4:1) to afford title compound as a yellow solid (1.1 g).

Step 2: Benzyl (2S, 5S)-5-[[4-(6-cyano-7-dimethylphosphoryl-1H-indol-3-yl)-5-(trifluoromethyl) pyrimidin-2-yl]amino]-2-methyl-piperidine-1-carboxylate

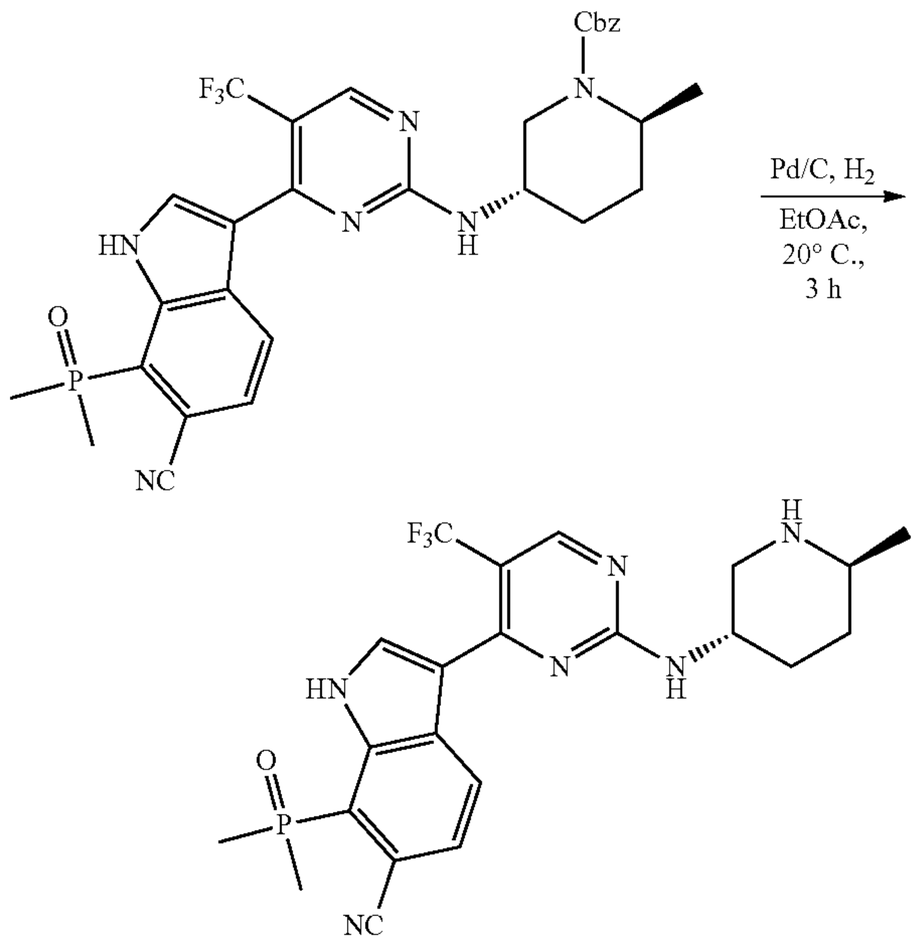
[0109]



[0110] We prepared a mixture of benzyl (2S,5S)-5-[[4-(7-chloro-6-cyano-1H-indol-3-yl)-5-(trifluoromethyl) pyrimidin-2-yl]amino]-2-methyl-piperidine-1-carboxylate (1.05 g, 1.85 mmol, 1 eq), methylphosphonoylmethane (720.17 mg, 9.23 mmol, 5 eq), K_3PO_4 (783.45 mg, 3.69 mmol, 2 eq), $Pd(OAc)_2$ (41.43 mg, 184.54 μmol, 0.1 eq), xantphos ($C_{39}H_{32}OP_2$; 106.78 mg, 184.54 μmol, 0.1 eq) and dimethylformamide (DMF; 10 mL) in a microwave sealed tube, degassed it, and purged it with N_2 (×3). The mixture was then stirred at 150° C. for 1 hour in microwave. The reaction mixture was diluted with H_2O (100 mL) and extracted with ethyl acetate (EtOAc; 50 mL×3). The combined organic layers were washed with brine (150 mL×2), dried over Na_2SO_4, filtered, and concentrated under reduced pressure to give a residue that we purified by column chromatography (SiO_2, petroleum ether/ethyl acetate=10:1 to 1:1) to afford the title compound as a yellow oil (490 mg).

Step 3: 7-dimethylphosphoryl-3-[2-[[(3S, 6S)-6-methyl-3-piperidyl]amino]-5-(trifluoromethyl)pyrimidin-4-yl]-1H-indole-6-carbonitrile

[0111]

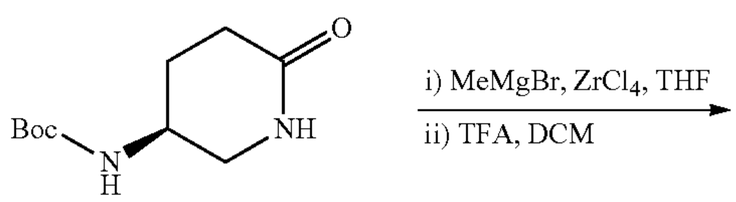


[0112] To a solution of benzyl(2S,5S)-5-[[4-(6-cyano-7-dimethylphosphoryl-1H-indol-3-yl)-5-(trifluoromethyl)pyrimidin-2-yl]amino]-2-methyl-piperidine-1-carboxylate (440 mg, 720.64 μmol, 1 eq) in EtOAc (5 mL), we added Pd/C (200 mg, 10% purity) under N_2. We degassed the suspension under vacuum, purged it with H_2 several times, then stirred the mixture under H_2 (15 psi) at 20° C. for 3 hours before filtering it. The filtrate was concentrated to give a residue we purified by prep-HPLC (high performance liquid chromatography; neutral condition) to yield the title compound as a white solid (142.2 mg).

[0113] The reaction was combined with another reaction in 50 mg scale for purification by liquid chromatography mass spectrometry (LCMS). LCMS: ET6034-1492-P1C: ($M+H^+$): 477.1 @2.572 (10-80% ACN (acetonitrile) in H_2O 4.5 minutes). ^1H NMR (400 MHz, DMSO (dimethylsulfoxide)-d6) δ 8.74 (br d, J=7.89 Hz, 1H), 8.65-8.44 (m, 2H), 8.17 (br d, J=15.35 Hz, 1H), 7.84 (brt, J=8.11 Hz, 1H), 7.67 (brt, J=7.02 Hz, 1H), 3.81 (br s, 1H), 3.10 (br d, J=11.40 Hz, 1H), 2.45-2.38 (m, 1H), 2.02 (d, J=13.59 Hz, 8H), 1.64 (br d, J=11.40 Hz, 1H), 1.49-1.34 (m, 1H), 1.11 (br d, J=10.96 Hz, 1H), 0.97 (br d, J=5.70 Hz, 3H).

Example 3: Synthesis of (S)-6,6-dimethylpiperidin-3-amine

[0114]



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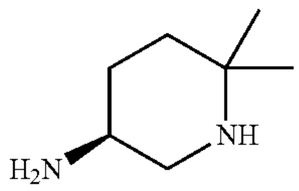
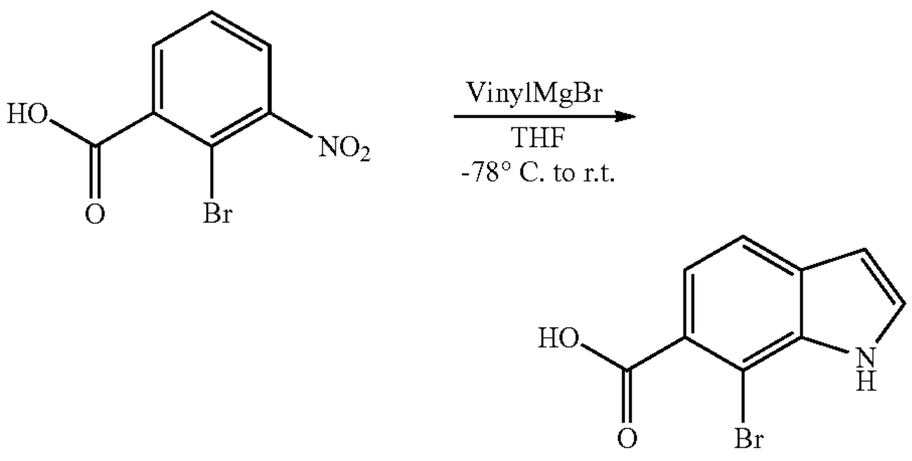


[0115] We dissolved (S)-tert-butyl (6-oxopiperidin-3-yl) carbamate (1.00 g, 4.67 mmol) (Tetrahedron Letters, 36:8205, 1995) in THF (47 mL) and cooled the solution to −10° C. Zirconium (IV) chloride (2.61 g, 11.22 mmol) was added, and the mixture was stirred for 30 minutes at this temperature. A methylmagnesium bromide solution (3M in ether, 20.25 mL, 60.75 mmol) was added, and the reaction mixture was allowed to slowly warm up to room temperature, at which it was stirred overnight. The solution was quenched with 30% aqueous NaOH, diluted with EtOAc, filtered, and then extracted 3 times with EtOAc. The organics were combined, dried over sodium sulfate, filtered, and concentrated in vacuo to provide the crude product as a yellow oil that was used without purification. The oil was dissolved in dichloromethane (DCM; 47 mL) and trifluoroacetic acid (TFA; 3.58 mL, 46.73 mmol) was added. We stirred the reaction mixture at room temperature for 16 hours, concentrated it in vacuo and co-evaporated it a few times with DCM to provide the crude title compound as a brown oil, which we used in the next step without further purification.

Example 4: Synthesis of (S)-7-(dimethylphosphoryl)-3-(2-((6,6-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-1H-indole-6-carbonitrile (Compound 101)

Step 1: 7-Bromo-1H-indole-6-carboxylic Acid

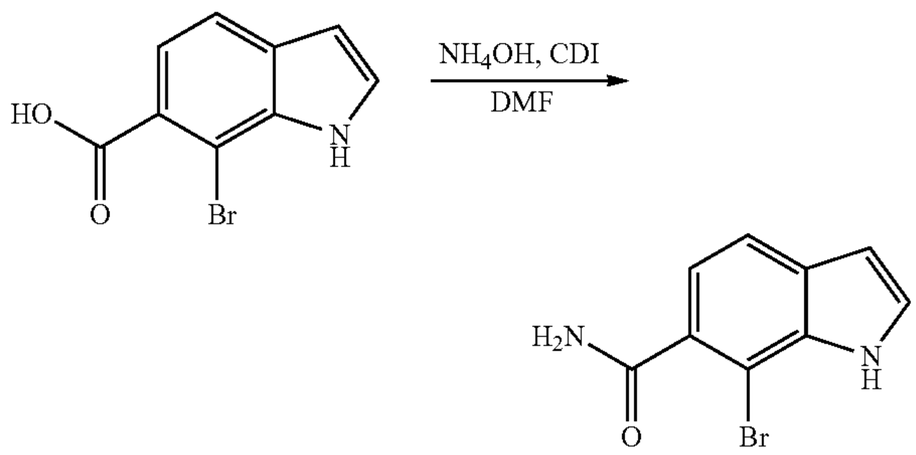
[0116]



[0117] We stirred a solution of vinylmagnesium bromide (1.0 M in THF (159 mL, 159 mmol) at −78° C. and added to it, dropwise, over a period of 1 hour, a solution of 2-bromo-3-nitrobenzoic acid (10.0 g, 39.8 mmol) in THF (159 mL). The reaction mixture was allowed to reach room temperature and was stirred at that temperature overnight. The reaction mixture was then poured over saturated aqueous ammonium chloride (150 mL) and acidified to a pH 2, using aqeous 1M HCl. We extracted the crude product with EtOAc (3×200 mL), dried the extract over sodium sulfate, filtered it, and concentrated it in vacuo. The residue was then triturated in DCM (100 mL) and dried overnight with a flow of air to provide the title compound as a light brown solid (7.58 g, 31.58 mmol, 79% yield).

Step 2: 7-Bromo-1H-indole-6-carboxamide

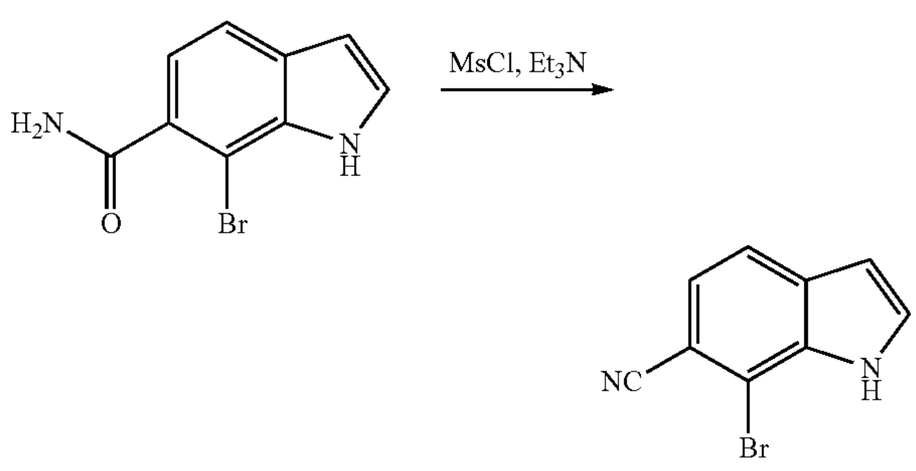
[0118]



[0119] We stirred a solution of 7-bromo-1H-indole-6-carboxylic acid (6.58 g, 27.4 mmol) in DMF (54.8 mL) at 0° C. and added 1,1'-carbonyldiimidazole (CDI; 8.89 g. 54.8 mmol) to it portion wise. The mixture was stirred for 5 minutes, and the intermediate was observed by LCMS. We then added NH_4OH (39.5 mL, 274 mmol) at 0° C., and the solution was stirred for 5 minutes. The reaction was quenched with saturated aqueous ammonium chloride (25 mL) and saturated aqueous sodium chloride (25 mL) then diluted with 2-methyltetrahydroftiran (MeTHF; 50 mL). We separated the phases and washed the organic layer again with saturated aqueous ammonium chloride (25 mL) and saturated aqueous sodium chloride (25 mL). The organic layer was then dried over sodium sulfate, filtered, and concentrated in vacuo to provide the title compound, which was carried over to the next step assuming the quantitative yield.

Step 3: 7-Bromo-1H-indole-6-carbonitrile

[0120]

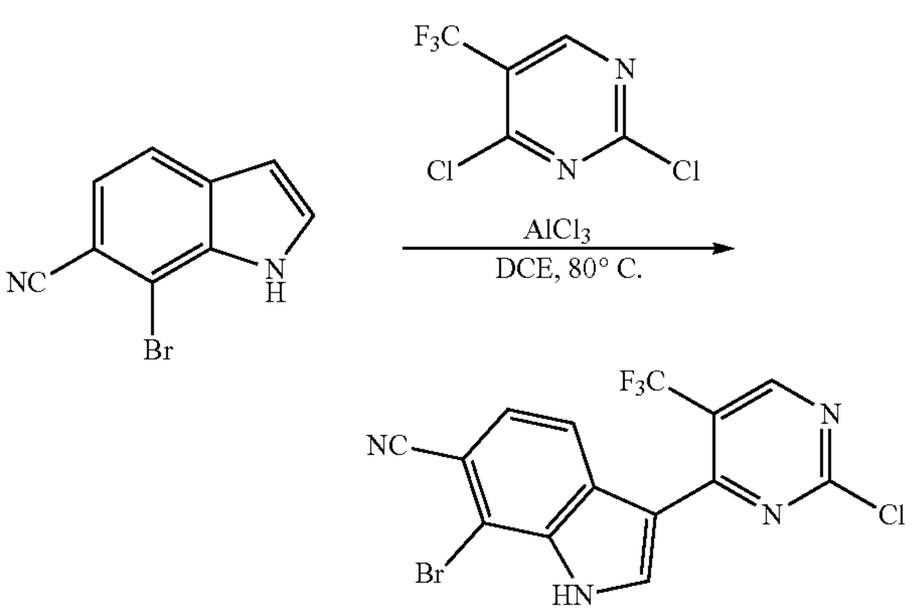


[0121] We added Et_3N (triethylamine; 44.1 mL, 315 mmol) to a suspension of 7-bromo-1H-indole-6-carboxamide (7.53 g, 31.5 mmol) in DCM (315 mL) at 0° C. and stirred the resulting orange solution at that temperature until we obtained a homogeneous solution. MsCl (12.2 mL, 157 mmol) was then added dropwise, and the solution was stirred at 0° C. for 5 minutes. We diluted the mixture with ethyl acetate and washed it with saturated aqueous sodium bicarbonate before extracting the aqueous layer twice more with ethyl acetate. The organic layers were combined, washed with brine, dried over sodium sulfate, filtered, and concentrated in vacuo. The residue was purified by filtering

it through a pad of silica (eluting with ethyl acetate) to provide the title compound as a brown solid (5.80 g, 26.24 mmol, 83% yield).

Step 4: 7-Bromo-3-(2-chloro-5-(trifluoromethyl) pyrimidin-4-yl)-1H-indole-6-carbonitrile

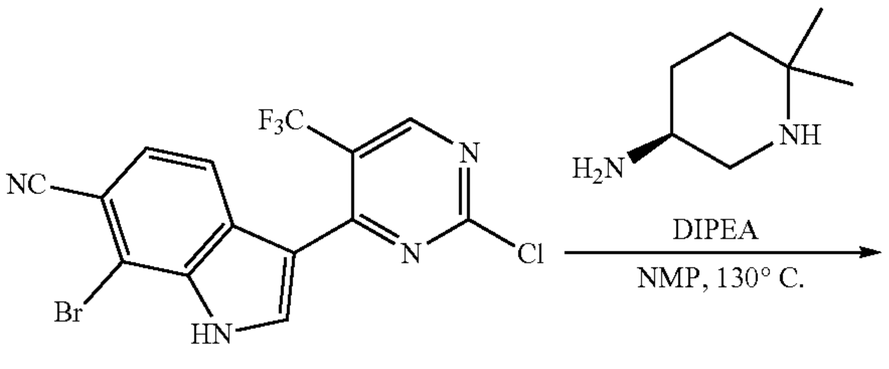
[0122]



[0123] We added $AlCl_3$ (1.83 g, 13.6 mmol) to a solution of 2,4-dichloro-5-trifluoromethylpyrim-idine (3.66 mL, 27.2 mmol) in 1,2-dichloroethane (DCE; 36.2 mL) and stirred the resulting suspension at 80° C. for 30 minutes. We added 7-bromo-1H-indole-6-carbonitrile (2.00 g, 9.05 mmol) to the mixture and stirred the resulting red solution at 80° C. until full conversion (4 hours). The reaction mixture was then diluted with MeTHF (100 mL) and washed with water (100 mL). The aqueous layer was extracted with 2-MeTHF (100 mL), and the organic extracts were combined, dried over sodium sulfate, filtered, and concentrated in vacuo. Formation of two possible regioisomers was observed in a ratio of 3:1 (desired/undesired). We purified the residue by reverse phase chromatography on C18 (MeCN (acetonitrile) in water, 15 to 80% gradient) to provide the title compound as a beige solid (1.51 g, 3.76 mmol, 42% yield). ^1H NMR (500 MHz, DMSO) δ 13.00 (brs, 1H), 9.17 (s, 1H), 8.35 (d, J=8.4 Hz, 1H), 8.16 (d, J=2.6 Hz, 1H), 7.71 (d, J=8.4 Hz, 1H).

Step 5: (S)-7-Bromo-3-(2-((6,6-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-1H-indole-6-carbonitrile

[0124]



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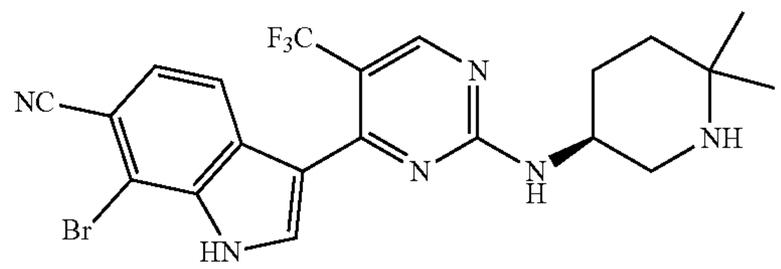
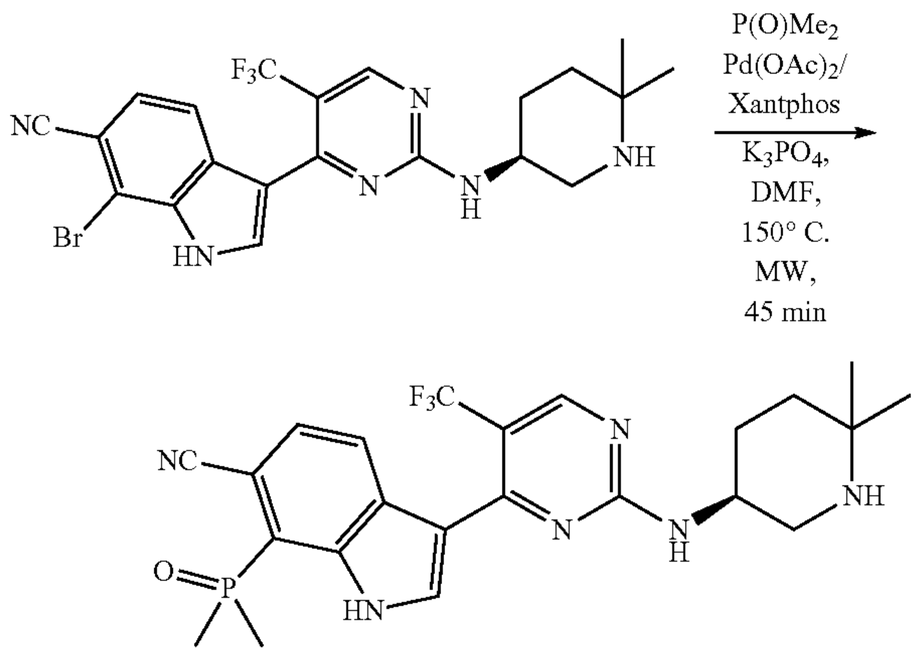


[0125] We dissolved 7-bromo-3-(2-chloro-5-(trifluoromethyl)pyrimidin-4-yl)-1H-indole-6-carbonitrile (200 mg, 0.498 mmol), (S)-6,6-dimethylpiperidin-3-amine (95.8 mg, 0.747 mmol), and DIPEA (174 μL, 0.996 mmol) in NMP (4 mL) then stirred the reaction mixture at 130° C. in an oil bath until full conversion (3 hours). The mixture Was cooled to room temperature, loaded directly onto a C18 column and purified by reverse phase chromatography (MeCN with 0.1% FA (formic acid) in water also containing 0.1% FA, 0 to 100% gradient). The title compound was obtained as a beige solid (245 mg. 0497 mmol, quantitative yield).

Step 6: (S)-7-(dimethylphosphoryl)-3-(2-((6-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)-pyrimidin-4-yl)-1H-indole-6-carbonitrile

[0126]

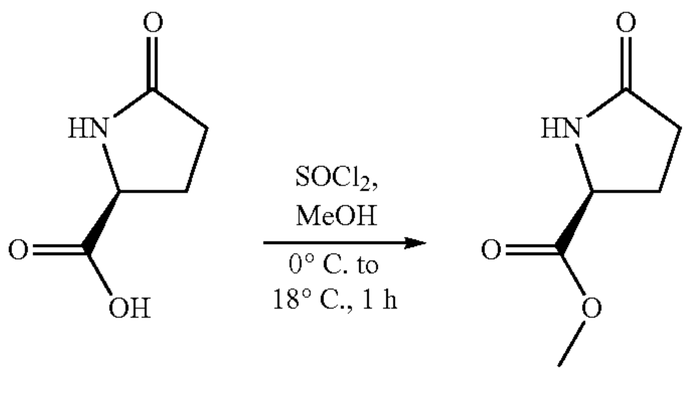


[0127] We combined (S)-7-bromo-3-(2-((6,6-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)-pyrimidin-4-yl)-1H-indole-6-carbonitrile (180.0 mg, 0.365 mmol), Xantphos (21.5 trig, 36.5 μmol), palladium (II) acetate (4.14 mg 18.2 μmol), and K_3PO_4 (85.2 mg. 0.401 mmol) in a 2.5 mL microwave vial under nitrogen. Dimethylphosphine oxide (73 mg, 0.912 mmol) was dissolved in anhydrous DMF (1 mL), and the solution was degassed before combining with the other reactants in a microwave vial. The sealed vial with the reaction mixture was then submitted to heat in a microwave reactor at 150° C. for 45 minutes. The reaction mixture was cooled to room temperature, loaded directly onto a C18 column, and purified by reverse phase chromatography (MeCN in aqueous 10 mM ammonium formate pH 3.8, 15 to 35% gradient). The title compound was obtained as an off-white solid (76 mg, 0.155 mmol, 42% yield).

Example 5: Synthesis of (3S)-1-benzyl-5, 5-dimethyl-piperidin-3-amine

Step 1: Methyl (2S)-5-oxopyrrolidine-2-carboxylate

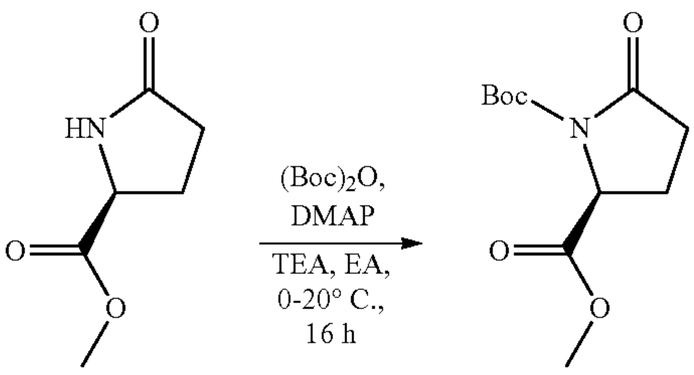
[0128]



[0129] We added $SOCl_2$ (215.62 g, 1.81 mol, 131.47 mL, 2 eq) to a solution of (2S)-5-oxopyrrolidine-2-carboxylic acid (117 g, 906.18 mmol, 1 eq) in MeOH (500 mL) at 0° C. The mixture was stirred at 18° C. for 1 hour before the reaction mixture was concentrated. We diluted the residue with EtOAc (1000 mL) and TEA (triethylamine; 150 mL) and filtered the solid that was formed. The filtrate was evaporated to afford the title compound as a light yellow oil (147 g, crude) to be used directly in the next step without any further purification.

Step 2: (S)-1-tert-butyl 2-methyl 5-oxopyrrolidine-1,2-dicarboxylate

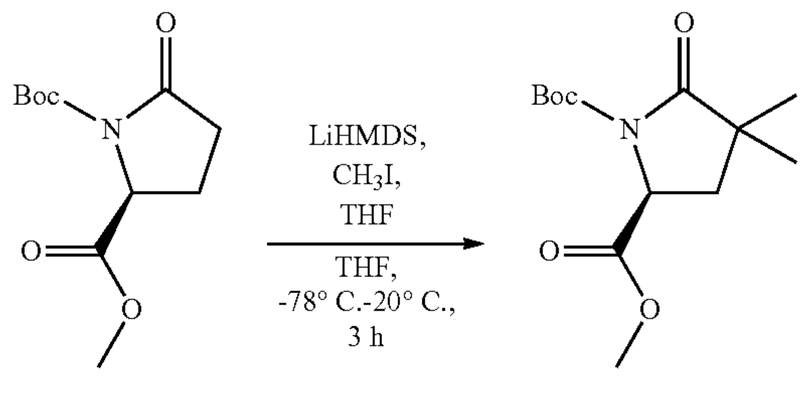
[0130]



[0131] To a solution of methyl (2S)-5-oxopyrrolidine-2-carboxylate (147 g, 1.03 mol, 1 eq), DMAP (4-dimethylaminopyridine; 15.06 g, 123.24 mmol, 0.12 eq) and TEA (259.80 g, 2.57 mol, 357.35 mL, 2.5 eq) in EtOAc (500 mL) we added tert-butoxycarbonyl tert-butyl carbonate (291.37 g, 1.34 mol, 306.71 mL, 1.3 eq), dropwise, at 0° C. The mixture was then stirred at 20° C. for 16 hours. We then washed the reaction mixture with HCl (0.5 M, 1000 mL), saturated $NaHCO_3$ (1000 mL), brine (1500 mL), dried it over Na_2SO_4, and filtered and concentrated it under reduced pressure to give a residue that was then purified by recrystallization from methyl tert-butyl ether (MTBE; 250 mL). The reaction mixture was filtered and evaporated to afford the title compound as a white solid (2 batches obtained; Batch 1: 108 g, 100% HPLC purity; Batch 2: 53 g, 90% ^1H NMR purity).

Step 3: (S)-1-tert-butyl 2-methyl 4,4-dimethyl-5-oxopyrrolidine-1,2-dicarboxylate

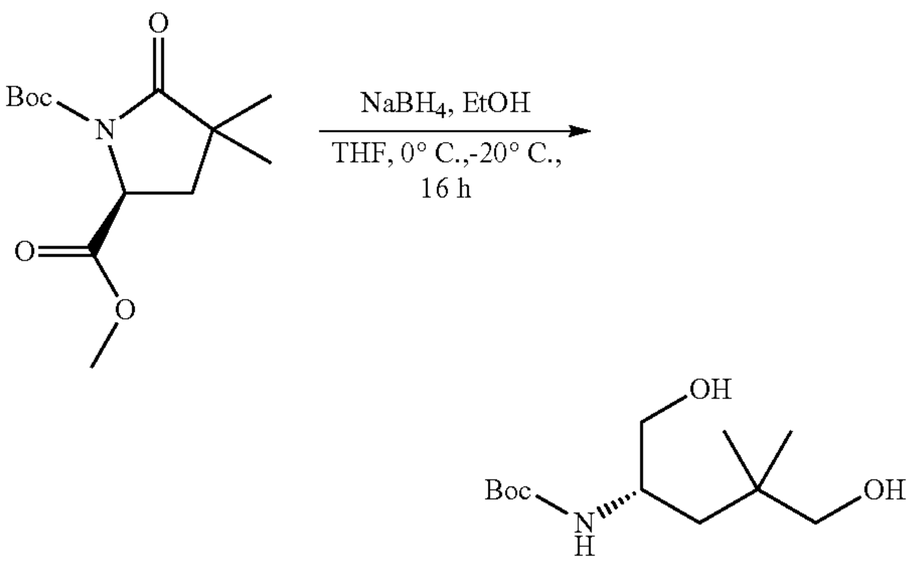
[0132]



[0133] We added LiHMDS (lithium hexamethyldisilazide; 1 M, 172.66 mL, 2.1 eq), dropwise, to a solution of (S)-1-tert-butyl 2-methyl 5-oxopyrrolidine-1, 2-dicarboxylate (20 g, 82.22 mmol, 1 eq) in THF (500 mL) at −78° C. under N_2 atmosphere. After addition, the mixture was stirred at that temperature for 30 minutes before we added CH_3I (35.01 g, 246.65 mmol, 15.36 mL, 3 eq), dropwise, at −78° C. under N_2 atmosphere. The resulting mixture was stirred at 20° C. for 2.5 hours. The reaction mixture was diluted with saturated aqueous NH_4Cl (1000 mL) and extracted with EtOAc (300 mL×3). The combined organic layers were washed with brine (500 mL), dried over Na_2SO_4, filtered, and concentrated under reduced pressure to give a residue that was purified by MPLC (SiO_2, PE:EtOAc=4:1-3:1) to afford the title compound as a light yellow solid (8 g, 25.95 mmol, 31.56% yield, 88% purity).

Step 4: tert-butyl N-[(1S)-4-hydroxy-1-(hydroxymethyl)-3,3-dimethyl-butyl]carbamate

[0134]

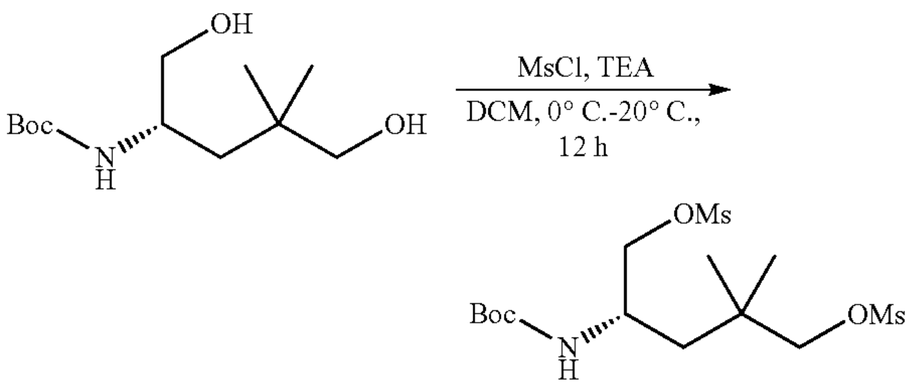


[0135] To a solution of (S)-1-tert-butyl 2-methyl 4, 4-dimethyl-5-oxopyrrolidine-1,2-dicarboxylate (4.3 g, 15.85 mmol, 1 eq) in THF (35 mL) we added $NaBH_4$ (1.80 g, 47.55 mmol, 3 eq), by portions, at 0° C. under N_2. After addition, EtOH (ethanol; 8.25 g, 179.09 mmol, 10.47 mL, 11.3 eq) was added dropwise at 0° C. The resulting mixture was stirred at 20° C. for 16 hours then poured into saturated aqueous NH_4Cl (250 mL) and extracted with EtOAc (100 mL×3). The combined organic layers were washed with brine (250 mL), dried over Na_2SO_4, filtered and concentrated under reduced pressure to afford the title compound as

a colorless oil (3.67 g, crude), which was used directly in the next step without any further purification

Step 5: [(2S)-2-(tert-butoxycarbonylamino)-4,4-dimethyl-5-methylsulfonyloxy-pentyl]methane-sulfonate

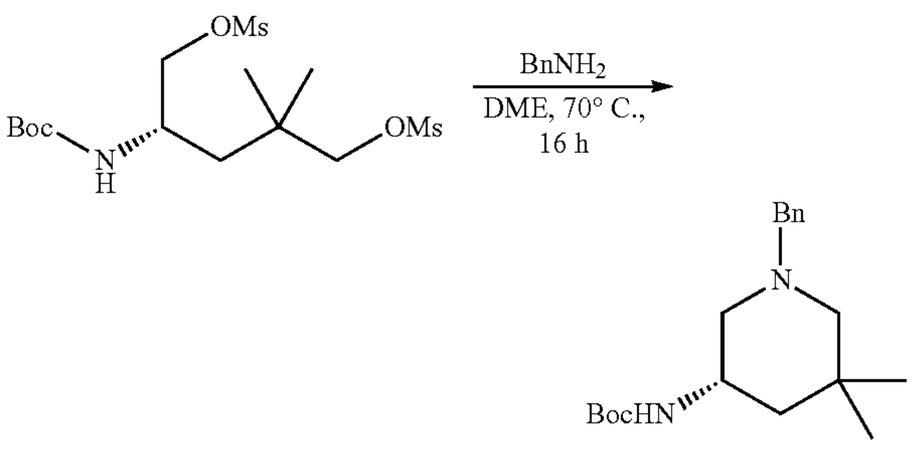
[0136]



[0137] To a solution of tert-butyl N-[(1S)-4-hydroxy-1-(hydroxymethyl)-3,3-dimethyl-butyl]carbamate (3.67 g, 14.84 mmol, 1 eq) and TEA (6.01 g, 59.35 mmol, 8.26 mL, 4 eq) in EtOAc (25 mL) we added methanesulfonyl chloride (5.10 g, 44.52 mmol, 3.45 mL, 3 eq), dropwise, at 0° C. The resulting mixture was stirred at 20° C. for 12 hours then poured into H_2O (200 mL). EtOAc (50 mL×3) was used to extract the product. The organic layer was washed with brine (30 mL), dried over Na_2SO_4, filtered and evaporated to afford the title compound as a colorless oil (6.06 g crude) that was used directly in the next step without any further purification.

Step 6: Tert-butyl N-[(3S)-1-benzyl-5,5-dimethyl-3-piperidyl] carbamate

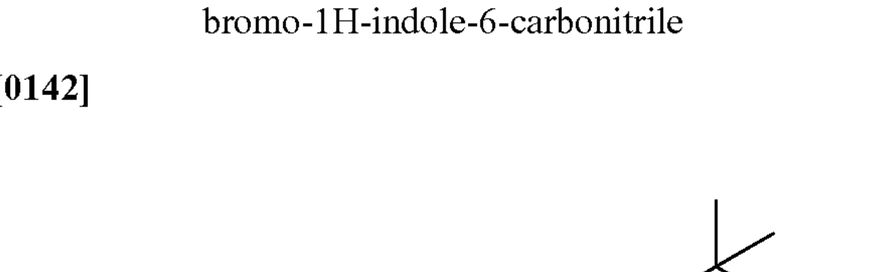
[0138]



[0139] A flask was fitted with [(2S)-2-(tert-butoxycarbonylamino)-4, 4-dimethyl-5-methyl-sulfonyloxypentyl] methanesulfonate (6.06 g, 15.02 mmol, 1 eq), phenylmethanamine (5.15 g, 48.06 mmol, 5.24 mL, 3.2 eq) and dimethoxyethane (DME; 50 mL). We heated the reaction mixture to 70° C. for 16 hours then poured it into H_2O (40 mL). DCM (40 mL×3) was used to extract the product. The organic layer was washed with brine (30 mL), dried over Na_2SO_4, filtered and evaporated to afford the crude product, which was purified twice by MPLC (SiO_2, PE:EtOAc=20:1-10:1) to afford the title compound as a colorless oil (580 mg, 1.49 mmol, 9.91% yield, 81.7% purity).

Step 7: (3S)-1-benzyl-5, 5-dimethyl-piperidin-3-amine

[0140]

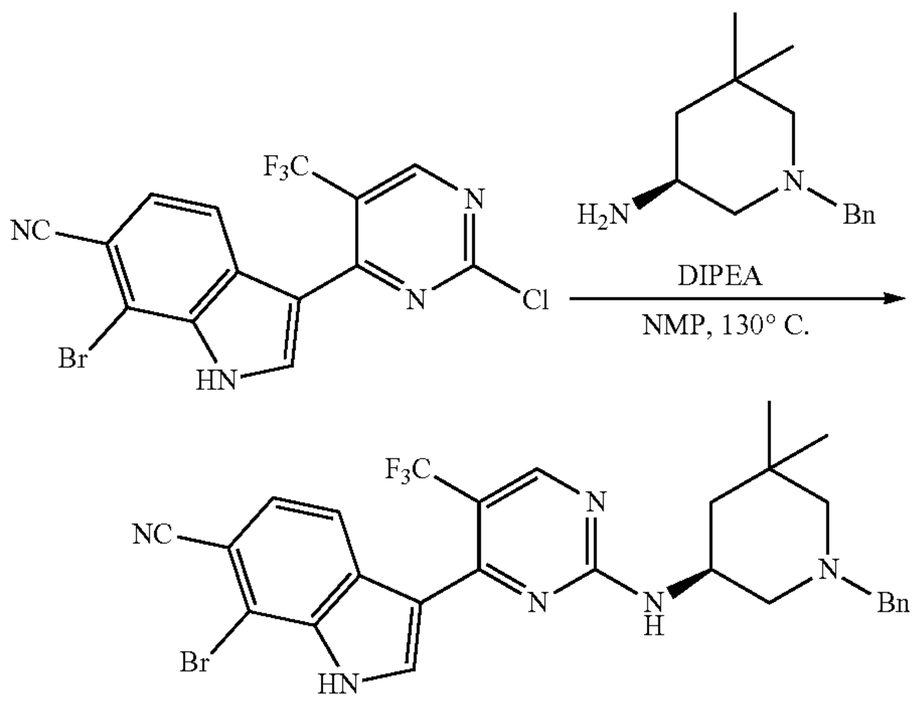


[0141] A flask was fitted with tert-butyl N-[(3S)-1-benzyl-5, 5-dimethyl-3-piperidyl]carbamate (300 mg, 942.05 μmol, 1 eq) in HCl/EtOAc (15 mL). The mixture was stirred at 25° C. for 1 hour, after which some white precipitate formed. We filtered the mixture, and the cake was washed by EtOAc (5 mL), collected and dried over vacuum to afford the title compound as a white solid (220 mg, 738.23 μmol, 78.36% yield, 85.5% purity, HCl) as a white solid to be used directly in the next step.

Example 6: Synthesis of (S)-7-(dimethylphosphoryl)-3-(2-((5,5-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-1H-indole-6-carbonitrile (Compound 102)

Step 1: (S)-3-(2-((1-benzyl-5,5-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-7-bromo-1H-indole-6-carbonitrile

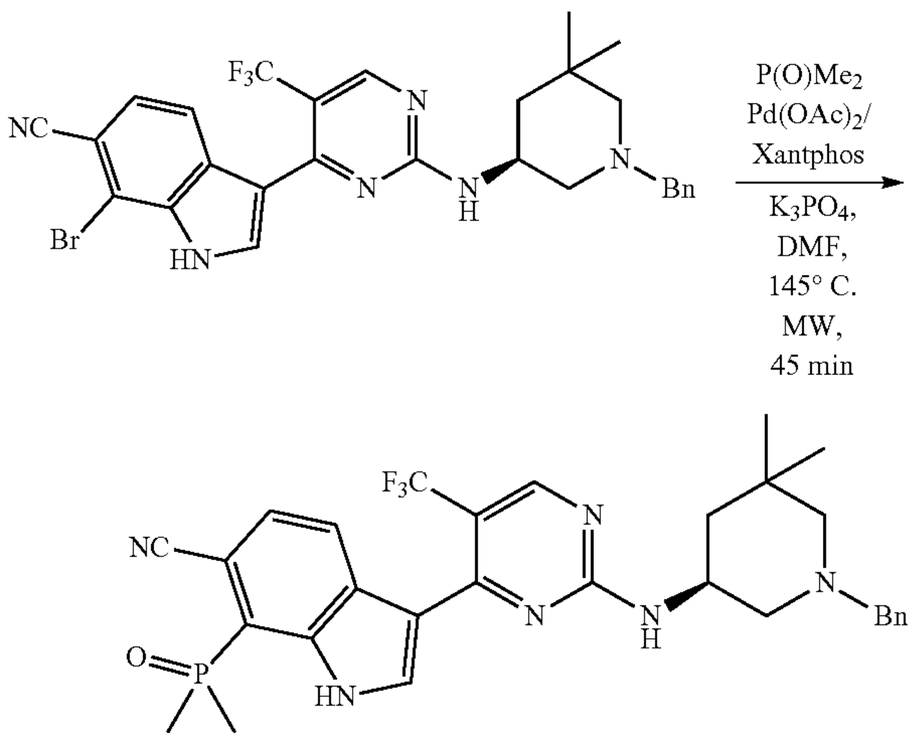
[0142]



[0143] We dissolved 7-bromo-3-(2-chloro-5-(trifluoromethyl)pyrimidin-4-yl)-H-indole-6-carbonitrile (168 mg, 0.418 mmol), (S)-1-benzyl-5,5-dimethylpiperidin-3-amine (128 mg, 0.585 mmol), and DIPEA (221 μL, 1.26 mmol) in NMP (2 mL). We stirred the reaction mixture at 130° C. in an oil bath until full conversion (4 hours). The mixture was cooled to room temperature, diluted with EtOAc and washed with saturated aqueous LiCl. The organic layer was separated, dried over sodium sulfate, filtered, and concentrated in vacuo to provide the crude title compound (240 mg, 0.411 mmol, quant. yield), which was used in the next step without further purification.

Step 2: (S)-3-(2-((1-benzyl-5,5-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-7-(dimethylphosphoryl)-1H-indole-6-carbonitrile

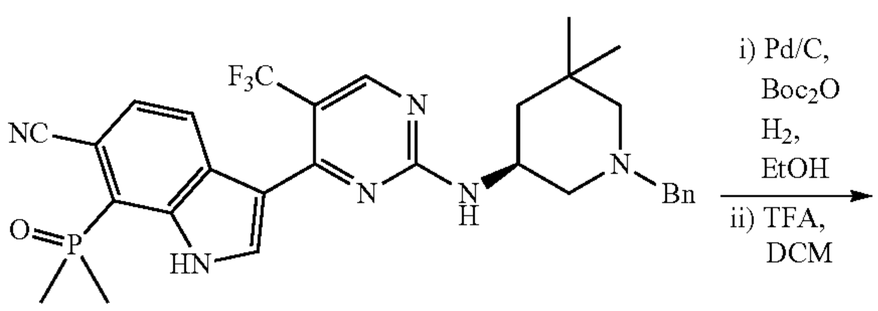
[0144]



[0145] We combined (S)-3-(2-((1-benzyl-5,5-dimethylpiperidin-3-yl)amino)-5-(trifluoro-methyl)pyrimidin-4-yl)-7-bromo-1H-indole-6-carbonitrile (240 mg, 0.411 mmol), Xantphos (24.3 mg, 41.1 μmol), palladium (II) acetate (4.66 mg, 20.6 μmol), and K_3P0_4 (96.0 mg, 0.452 mmol) in a 2.5 mL microwave vial under nitrogen. Dimethylphosphine oxide (39.2 mg, 0.494 mmol) was dissolved in anhydrous DMF (1 mL), and the solution was degassed before combining with the other reactants in a microwave vial. The sealed vial with the reaction mixture was then submitted to heat in a microwave reactor at 145° C. for 45 minutes. The reaction mixture was then cooled to room temperature, diluted with 2-MeTHF and washed with saturated aqueous $NaHCO_3$ and brine. The organic layer was separated, dried over sodium sulfate, filtered, and concentrated in vacuo before the residue was purified by reverse phase chromatography on C18 (MeCN in aqueous 10 mM ammonium formate pH 3.8, 0 to 100% (gradient). The title compound was obtained as a pale brown oil (58.0 mg, 0.10 mmol, 24% yield).

Step 3: (S)-7-(dimethylphosphoryl)-3-(2-((5,5-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-1H-indole-6-carbonitrile

[0146]



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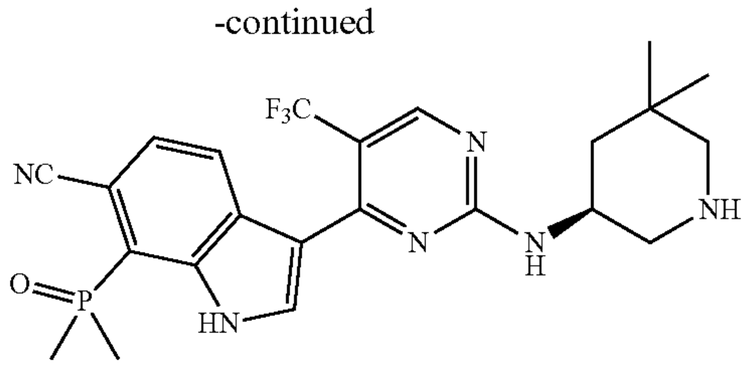

[0147] Under a nitrogen atmosphere, to a stirring solution of (S)-3-(2-((1-benzyl-5,5-dimethylpiperidin-3-yl)amino)-5-(trifluoromethyl)pyrimidin-4-yl)-7-(dimethylphosphoryl)-1H-indole-6-carbonitrile (58.0 mg, 0.10 mmol) in EtOH (12.5 mL), we added Pd/C 10% w/w (1.1 mg, 0.01 mmol) and Boc_2O (di-t-butyl decarbonate; 65.5 mg, 0.30 mmol). The reaction mixture was evacuated and back-filled with nitrogen (×3) before being filled with hydrogen. The reaction mixture was then stirred at room temperature overnight under hydrogen atmosphere. After 16 hours, we observed an incomplete conversion and therefore filtered the reaction mixture through a pad of CELITE® and concentrated it under reduced pressure. The reaction was then repeated with the residue as described above. After almost complete consumption of starting material (48 hours), the reaction mixture was filtered through a pad of CELITE® and concentrated in vacuo to provide the crude product, which was engaged in the next step. Thus, the obtained oil was redissolved in DCM (5 mL), and TFA (0.23 mL, 3.0 mmol) was added. The reaction mixture was stirred at room temperature overnight. The mixture was then concentrated in vacuo, and the residue was purified by reverse phase chromatography on C18 (MeCN in aqueous 10 mM ammonium formate pH 3.8, 0 to 100% gradient) to provide the title compound as a white solid (11.11 mg, 0.023 mmol, 23% yield over two steps).

Example 7: Inhibition of CDK Kinase Activity

[0148] We assayed some compounds for inhibition of CDK7, CDK9, CDK12, and CDK2 activity at Biortus Biosciences (Jiangyin, Jiangsu Province, P.R. of China) using kinase assays for each CDK developed with a Caliper/LabChip EZ Reader (Perkin Elmer, Waltham, Mass.). These assays measure the amount of phosphorylated peptide substrate produced as a fraction of the total peptide following an incubation period at 27° C. with the following components: test compounds (variable concentrations from 10 μM down to 0.508 nM in a series of 3-fold serial dilutions), active CDK protein (with the indicated cyclin, listed below for each CDK), ATP (at either the K_m concentrations listed below for each CDK/cyclin or 2 mM ATP), and substrate peptide (listed below) in the following buffer: 2-(N-morpholino)ethanesulfonate (MES buffer, 20 mM), pH 6.75, 0.01% (v/v) Tween 20 detergent, 0.05 mg/mL bovine serum albumin (BSA), and 2% DMSO.

[0149] Specifically, the CDK7 inhibition assay used CDK7/Cyclin H/MAT1 complex (6 nM) and "5-FAM-CDK7tide" peptide substrate (2 μM, synthesized fluorophore-labeled peptide with the sequence 5-FAM-YSPTSP-SYSPTSPSYSPTSPSKKKK (SEQ ID NO:1), where "5-FAM" is 5-carboxyfluorescein) with 6 mM MgCl2 in the buffer composition listed above where the apparent ATP K_m for CDK7/Cyclin H/MAT1 under these conditions is 50 μM.

The CDK9 inhibition assay used CDK9/Cyclin T1 complex (8 nM) and "5-FAM-CDK9tide" peptide substrate (2 μM, synthesized fluorophore-labeled peptide with the sequence: 5-FAM-GSRTPMY-NH_2 (SEQ ID NO:2), where 5-FAM is defined above and NH_2 signifies a C-terminal amide with 10 mM $MgCl_2$ in the buffer composition listed above. The CDK12 inhibition assay used CDK12 (aa686-1082)/Cyclin K complex (50 nM) and "5-FAM-CDK9tide" (2 μM) as defined above, with 2 mM $MgCl_2$ in the buffer composition above. The CDK2 inhibition assay used CDK2/Cyclin E1 complex (0.5 nM) and "5-FAM-CDK7tide" (2 μM) as defined above, with 2 mM $MgCl_2$ in the buffer composition listed above.

[0150] The incubation period at 27° C. for each CDK inhibition assay was chosen such that the fraction of phosphorylated peptide product produced in each assay, relative to the total peptide concentration, was approximately 20% (±5%) for the uninhibited kinase (35 minutes for CDK7, 35 minutes for CDK2, 3 hours for CDK12, and 15 minutes for CDK9). In cases where the compound titrations were tested and resulted in inhibition of peptide product formation, these data were fit to produce best-fit IC_{50} values. The best-fit IC_{50} values at K_m ATP for each CDK/Cyclin, except for CDK7/Cyclin H/MAT1, were used to calculate K_i values, or the apparent affinity of each inhibitor for each CDK/Cyclin from the kinase activity inhibition assay, according to the Cheng-Prusoff relationship for ATP substrate-competitive inhibition (Cheng and Prusoff, *Biochem. Pharmacol.,* 22(23):3099-3108, 1973), with a correction term for inhibitor depletion due to the enzyme concentration (Copeland, "Evaluation of Enzyme Inhibitors in Drug Discover: A Guide for Medicinal Chemists and Pharmacologists," Second Edition, March, 2013; ISBN: 978-1-118-48813-3):

$$IC_{50} = K_i\left(1 + \frac{[\text{Substrate}]}{K_m}\right) + \frac{[\text{Enzyme}]}{2}$$

[0151] Due to tight-binding inhibition and the limits of the CDK7/Cyclin H/MAT1 assay, instead of calculating the apparent K_i values for each inhibitor, the K_d, or direct compound binding affinity, was measured using surface plasmon resonance (SPR) as described below.

Example 8: CDK7/Cyclin H Surface Plasmon Resonance (SPR) Assay Method

[0152] We measured binding kinetics and affinities of selected compounds to the CDK7/Cyclin H dimer using a Biacore T200 surface plasmon resonance (SPR) instrument (GE Healthcare). The dimer was amine-coupled to a CM5 sensor chip at pH 6.5 in 10 mM MES buffer at a concentration of 12.5 μg/mL with a flow rate of 10 μL/min. Target protein was immobilized on two flow cells for 12-16 seconds to achieve immobilized protein levels of 200-400 Response Units.

[0153] Compounds were titrated from 0.08-20 nM in a 9-step, 2-fold serial dilution in 10 mM HEPES buffer at pH 7.5 with 150 mM NaCl, 0.05% Surfactant P20, and 0.0002% DMSO. Each compound concentration cycle was run at 100 μL/min with 70 second contact time, 300 second dissociation time, 60 second regeneration time with 10 mM glycine pH 9.5, and 400 second stabilization time. For each compound, 0 nM compound controls and reference flow-cell binding were subtracted to remove background and normalize data. Compound titrations were globally fit by Biacore T200 Evaluation Software (GE Healthcare) using kinetics mode. Best-fit values for compound binding on-rate (k_{on}) and dissociation off-rate (k_{off}) for CDK7/Cyclin H were determined and these values were used to calculate the compound affinity (K_d) for CDK7/Cyclin H using the following equation:

$$K_d(M) = \frac{k_{off}(s^{-1})}{k_{on}(M^{-1}s^{-1})}.$$

Compound selectivity for CDK7 over CDK2, CDK9, or CDK12 were determined based on the ratios of K_i values for the off-target CDKs relative to the direct compound binding K_d for CDK7 measured by SPR according to:

$$\text{Selectivity} = \frac{K_{i,off\,target}}{K_{d,CDK7}}.$$

The inhibitory and dissociation constants and selectivity of the indicated compounds (three compounds of the invention and four comparators) against CDK2, CDK7, CDK9, and CDK12 are shown in the table of FIG. **1**. As can be seen, each of the compounds of the invention is at least 1300-fold and up to 40,000-fold more specific for CDK7 than for the other CDKs tested.

Example 9: Inhibition of Cell Proliferation (Compounds 100-102)

[0154] The HCC70 cell line was derived from human TNBC, and we tested representative compounds of the invention, at different concentrations (from 4 μM to 126.4 μM; 0.5 log serial dilutions), for their ability to inhibit the proliferation of those cells. More specifically, we tested the same compounds tested above for CDK7 selectivity (the structures of which are shown in FIG. **1**), and we used the known CDK inhibitors dinaciclib (or N-((1S,3R)-3-((5-chloro-4-(1H-indol-3-yl) pyrimidin-2-yl)amino)cyclohexyl)-5-((E)-4-(dimethylamino)but-2-enamido)picolinamide) and triptolide as positive controls. The cells were grown in ATCC-formulated RPMI-1640 medium (ATCC 30-2001) supplemented with 10% fetal bovine serum (FBS), at 37° C. in a humidified chamber in the presence of 5% CO_2. We conducted proliferation assays over a 72-hour time period using a CyQUANT® Direct Cell Proliferation Assay (Life Technologies, Chicago, Ill. USA) according to the manufacturer's directions and utilizing the reagents supplied with the kit. The results of the assay are shown in the Table below.

Compound	HCC70 EC_{50} (nM)
Compound 100	0.98
Compound 101	5.6
Compound 102	2.1
Comparator 1	0.53
Comparator 2	260
Comparator 3	24
Comparator 4	110

Example 10: TGI in Patient-Derived Xenograft (PDX) Models

[0155] Tumor growth inhibition was evaluated in estrogen receptor-positive breast cancer (ER+BC) PDX models selected in vivo for resistance to the CDK4/6 inhibitor palbociclib (ST1799, n=1) or resistance to both palbociclib and fulvestrant (ST941, n=1). Dosing was initiated when tumors were 100-200 mm^3. Mice were treated with either Compound 101, QD (6 mg/kg, once daily, by mouth); fulvestrant, SC (2.5 mg/kg, once weekly dosing, by subcutaneous injection); palbociclib, QD (50 mpk, once daily, by mouth) or in combination of Compound 101 (6 mg/kg, once daily, by mouth) and fulvestrant (2.5 mg/kg, once weekly, by subcutaneous injection) over the course of 28 days, followed by 21 days of observation. Tumor growth inhibition (TGI) was calculated on the last day of dosing using the formula: $TGI=(V_{c1}-V_{t1})/(V_{c0}-V_{t0})$, where V_{c1} and V_{t1} are the mean volumes of control and treated groups at the time of tumor extraction, while V_{c0} and V_{t0} are the same groups at the start of dosing.

[0156] In the palbociclib-resistant ER+BC PDX (ST1799) model, the combination of Compound 101 and fulvestrant induced significant TGI (89%), with no evident tumor regrowth up to 21 days after dosing cessation, distinguishing the observed effects from Compound 101 (83%), fulvestrant (60%) or palbociclib (21%) when administered as single agents. Additionally, the combination of Compound 101 and fulvestrant was superior to the SOC combination of palbociblib and fulvestrant (75%). In a palbociclib and fulvestrant double-resistant ER+BC PDX model (ST941), Compound 101 administered as a single agent resulted in 33% TGI and fulvestrant and palbociclib as single agents or fulvestrant and palbociclib in combination had no activity. In contrast, the combination of Compound 101 and fulvestrant demonstrated significant TGI (68%; $p<0.0001$ vs fulvestrant as a single agent), suggesting re-sensitization to fulvestrant.

[0157] FIG. **2** illustrates the TGI results from the palbociclib resistant HR+BC PDX model ST1799, and FIG. **3** illustrates the TGI results from the palbociclib and fulvestrant resistant HR+BC PDX model ST941. We also observed TGI in four additional PDX models; BR5010 (modeling TNBC), LU5178 (modeling small cell lung cancer (SCLC)), OV15398 (modeling high grade serous ovarian cancer (HGSOC)), and ST390 (modeling pancreatic ductal adenocarcinoma (PDAC)). In the TNBC model, Compound 101 was orally administered to tumor-bearing NOD/SCID mice at 10 mg/kg QD or 5 mg/kg BID for 21 days. In the SCLC and HGSOC models, Compound 101 was orally administered to tumor-bearing NOD/SCID mice at 3 mg/kg BID for 21 days. In the PDAC model, Compound 101 was orally administered to tumor-bearing NOD/SCID mice at 6 mg/kg QD. In the TNBC, SCLC, and HGSOC models, tumor volume was measured during the treatment period and for an additional 21 days after treatment ceased. The % TGI observed at the end of treatment (day 21) was calculated as: 1−[(Mean TV Compound 101 @ EOT−Mean TV Compound 101 @ Day 0)/(Mean TV Veh @ EOT−Mean TV Veh @ Day 0)]×100. The % regression was calculated as: (Mean TV Compound 101 @ EOT)/(Mean TV Compound 101 @ Day 0)×100. The same calculations were used for end of study (day 42). The results are shown in FIG. **4**. These results demonstrate deep and sustained TGI, including regressions, at well tolerated doses, in a variety of tumor types. Dose-dependent transcriptional responses in xenograft tissue were observed within 4 hours of dosing and were sustained for 24 hours. Similar TGI was seen when the same total dose was administered either QD or BID in the TNBC PDX model, suggesting that the effect was AUC or C_{min} driven. Moreover, the TGI observed in SCLC (in the LU5178 PDX model) had not been observed in previous studies with a covalent CDK7 inhibitor (data not shown). Regarding the model of PDAC, we found Compound 101 induced 100% TGI over the time examined (~28 days) at a dose well below the MTD: at day 21, tumor volume was ~1,250 mm^3 in vehicle-treated mice but only about 250 mm^3 in Compound 1-treated mice (6 mg/kg QD, PO). While Compound 101 could achieve 100% TGI at sub-MTD doses in the tested PDAC PDX tumors, a covalent CDK7 inhibitor achieved only modest TGI at its MTD (40 mg/kg BIW, by IV administration, with evident body weight loss (8.4%) and necrosis at the injection site; data not shown).

Example 11. In Vitro Studies of Compound 101 in Combination with Various Second Agents

[0158] In the studies described here, cancer cell lines from HR+ breast cancers (lines T47D; PIK3CA p.H1047R, MCF7; PIK3CA p.E545K), SCLC, (NCI-H1048) and CRCs (lines RKO; BRAF p.V600E, SW480; KRAS p.G12V) were grown to 70% confluency in their media of preferences based on the manufacturer recommendations. In the SCLC cell line (NCI-H1048), Compound 101 was tested in combination with SOC chemotherapy agents gemcitabine (a DNA synthesis inhibitor) and carboplatin (a DNA damage agent). In a CRC cell line (RKO; BRAF p.V600E), Compound 101 was tested in combination with SOC chemotherapy agent oxaliplatin (a DNA damage agent). Additionally, in CRC, Compound 101 was tested in combination with the selective MAPK pathway inhibitor trametinib in two CRC cell lines harboring MAPK pathway alterations; RKO (BRAF p.V600E mutant) and SW480 (KRAS p.G12V mutant). Compound 101 was tested in combination with the SOC agent capecitabine (an antimetabolite) in HR+MCF-7 cells. In the HR+ cell lines MCF7 and T47D, which have activating mutations in the PIK3CA kinases, Compound 101 was tested in combination with the PIK3CA selective inhibitor alpelisib.

[0159] On the day of assay, cells were lifted and counted using the Countess II FL (Life Technologies). Using an automated dispenser (here, Multidrop™ Combi Reagent Dispenser), 50 μL of preferred cell media containing 20,000-50,000 cells/ml was distributed into black 384-well Nunc plates (Thermo) and allowed to adhere overnight prior to compound addition. Compound arrays were distributed to 384 well assay plates using Synergy Plate Format with an HP D300e Digital Dispenser (HP). Compound 101 and other TEST agents were dissolved in DMSO to make a stock solution that allowed for more accurate dispensing. However, due to solubility and reactivity, platinum agents were dissolved in water with an addition of 0.03% Tween-20 to allow for dispensing with digital printer. Compounds were plated in each quadrant of a 384-well plate in quadruplicate. Each quadrant contained test wells with combination of SY-1365 and carboplatin or oxaliplatin (TEST/test agent) as well as single agent columns, and vehicle wells.

[0160] Compound 101 was plated in across from left to right in a high to low concentration (8 columns), and the varying concentrations of carboplatin or oxaliplatin (TEST) plated in synergy wells from top to bottom (7 rows).

Concentrations were selected to cover the full isobologram of activity based on activity of single agents. Single agents were plated in dose in two columns, with a third separate column of just DMSO/vehicle treated wells. A separate plate for each cell line was seeded to allow for determination of a "Time Zero"/"Day Zero" number of cells to parse the differential cytostatic vs cytotoxic effects. On the day compounds were added, viability of the time zero plate was determined to identify growth inhibition from cell killing effects.

[0161] After addition of compound, cell plates were incubated for 5 days in a 37° C. incubator. Cell viability was evaluated using CellTiter-Glo® 2.0 (Promega) following manufacturer protocols. Data was analyzed in CalcuSyn utilizing the median effect principle of presented by Chou-Talalay and visualized using GraphPad Prism Software. Key parameters assessed were combination index and dose reduction index.

[0162] We found the combination of Compound 101 with SOC chemotherapy (gemcitabine or carboplatin in SCLC, oxaliplatin in CRC, or capecitabine in HR+ breast cancer) showed synergy and was superior to either agent alone. The combination of Compound 101 with the targeted agent trametinib, a selective MAPK pathway inhibitor approved for the treatment of BRAF p.V600E mutant melanoma and NSCLC, show significant synergy in BRAF p.V600E mutant CRC as well as in KRAS p.G12V mutant CRC, which harbors a different mutation within the MAPK pathway. The combination of Compound 101 with the targeted agent alpelisib, a selective PIK3CA inhibitor approved for the treatment of PIK3CA mutant HR+BC, showed significant synergy in both HR+ cell lines representing the two most common activating mutation of PIK3CA (p.E545K and p.H1047R). All synergy was determined using CalcuSyn utilizing the median effect principle of presented by Chou-Talalay and visualized using GraphPad Prism Software. Combination effect is reflected by shift in IC50 of Compound 101 with addition of carboplatin or oxaliplatin or increased antiproliferative effect with lower amounts of either single agent. This is visualized in the isobolograms of FIG. **5**, where points below the diagonal line reflect synergy.

Example 12. Deep and Sustained Responses to Compound 101 in TNBC, HGSOC, and SCLC PDX Models

[0163] We evaluated TGI in 12 different PDX models (Crown Biosciences) in various tumor indications with PDXs representing SCLC (n=5; LU5180, LU5178, LU5192, LU5173, LU5210), TNBC (n=4; BR5010, BR1458, BR5399, BR10014) and HGSOC (n=3; OV15398, OV5392, OV15631). Dosing was initiated when tumors were 150-300 mm^3. Mice were treated with either Compound 101, QD (6 or 10 mg/kg once daily, by mouth) or BID (3 or 5 mg/kg twice daily, by mouth) over the course of 21 days, followed by 21 days of observation. TGI was calculated on the last day of dosing using the formula: TGI $(V_{c1}-V_{t1})/(V_{c0}-V_{t0})$, where V_{c1} and V_{t1} are the mean volumes of control and treated groups at the time of tumor extraction, while V_{c0} and V_{t0} are the same groups at the start of dosing.

[0164] To perform whole exome sequencing (WES), we isolated DNA from passage matched tumors using DNeasy® Blood and Tissue Kit via manufacturer protocol and sent it to Wuxi Aptec for WES using Agilent's SureSelectXT Human All Exon V6 kit. Samples were sequenced to a depth of ~300×. Reads were trimmed to remove adapter sequences via Skewer (v0.2.1). Reads were then mapped and further processed using Sentieon tools: BWA, DeDup, Realigner, and QualCal (v201808.03). Variants were called using Sentieon's Haplotyper tool, and initial annotations were performed using Ensembl's Variant Effect Predictor (VEP, release_96.2). FATHMM-MLK was also used to annotate variant effects. Variants that met the following qualifications were included in sample characterizations: (1) variant is located in a protein-coding gene; (2) variant affects protein sequence or results in a frameshift; (3) missense mutations are classified as damaging by SIFT, PolyPhen, or FATHMM-MLK (≥0.75); (4) variant allele frequency is ≥10%. Copy-number (CN) variation across capture regions were called using CNVkit (v0.9.1), and CNs for individual genes were calculated by using the mean CN across its capture regions. For model LU5210 mutation/CNV data was made available from WES data provided by the PDX vendor (Crown Biosciences Inc.).

[0165] At these doses, Compound 101 induced at least 50% TGI at the end of the 21-day dosing period in all models. In a subset of models (58%, 7/12), Compound 101 responses were deep (>95% TGI or regression) and sustained, with no evidence of tumor regrowth for 21 days after treatment discontinuation (see FIG. **6**). Compound 101 was well tolerated, with no evident body weight loss at all once-daily doses tested, indicating that the MTD is above 10 mg/kg once daily in tumor-bearing mice. Deep and sustained responses were observed in each indication tested and were associated with alterations in the RB pathway including RB1 deletion or mutation, CDKN2A deletion, or CCNE1 amplification (FIG. **7**). These results highlight the therapeutic potential of Compound 101 in TNBC, HGSOC, and SCLC, particularly in tumors with RB pathway alterations and aberrant cell cycle control.

[0166] It should it be understood that, in general, where the invention, or aspects of the invention, is/are referred to as comprising particular elements and/or features, certain embodiments of the invention or aspects of the invention consist, or consist essentially of, such elements and/or features. For purposes of simplicity, those embodiments have not been specifically set forth in haec verba herein. Where ranges are given, endpoints are included. Furthermore, unless otherwise indicated or otherwise evident from the context and understanding of one of ordinary skill in the art, values that are expressed as ranges can assume any specific value or sub-range within the stated ranges in different embodiments of the invention, to the tenth of the unit of the lower limit of the range, unless the context clearly dictates otherwise.

[0167] Those skilled in the art will recognize, or be able to ascertain using no more than routine experimentation, that there are many equivalents to the specific embodiments of the disclosure described and claimed herein. Such equivalents are intended to be encompassed by the following claims.

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1 5 10 15

Pro Thr Ser Pro Ser Lys Lys Lys Lys
20 25

<210> SEQ ID NO 2
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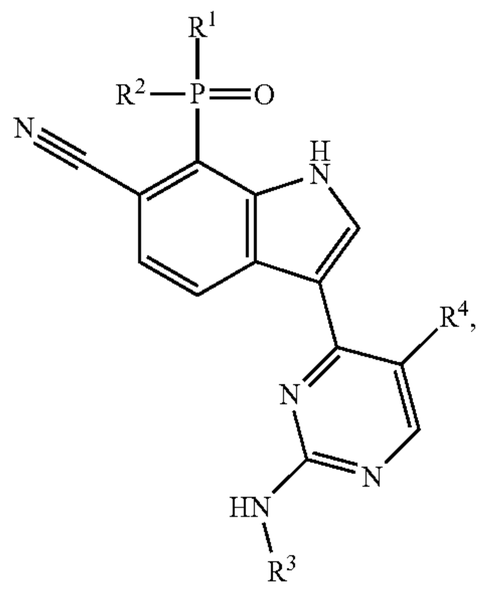
1 5

1. A method of treating a selected patient, the method comprising administering a therapeutically effective amount of a compound of structural Formula (I):

(I)



or a pharmaceutically acceptable salt thereof, wherein the compound or the pharmaceutically acceptable salt thereof is optionally within a pharmaceutical composition;

R^1 is methyl or ethyl;

R^2 is methyl or ethyl;

R^3 is 5-methylpiperidin-3-yl, 5,5-dimethylpiperidin-3-yl, 6-methylpiperdin-3-yl, or 6,6-dimethylpiperidin-3-yl, wherein one or more hydrogen atoms in R^3 is optionally replaced by deuterium;

R^4 is —CF_3 or chloro;

and the selected patient has been determined to have a cancer in which

(a) a gene selected from RB1, RBL1, RBL2, CDKN2A, CDKN2B, CDKN2C, CDKN2D, CDKN1A, CDKN1B, CDKN1C, and FBWX7 is mutated, is genetically deleted, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard;

(b) a gene selected from E2F1, E2F2, E2F3, E2F4, E2F5, E2F6, E2F7, E2F8, CDK1, CDK2, CDK4, CDK6, CCNA1, CCNB1, CCND1, CCND2, CCND3, CCNE1, CCNE2, and BRAF is mutated, is genetically gained or amplified, contains an epigenetic alteration, is translocated, transcribed at a level equal to or above a pre-determined threshold, or encodes a protein that is translated at a level equal to or above a pre-determined threshold or has increased activity relative to a reference standard; or

(c) the gene Bcl2-like 1 is mutated, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a BCL-xL protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard.

2. The method of claim **1**, wherein (i) R^1 is methyl and R^2 is methyl or (ii) R^1 is methyl and R^2 is ethyl.

3. The method of claim **1**, wherein (i) R^1 is ethyl and R^2 is ethyl or (ii) R^4 is —CF_3.

4. The method of claim **1**, wherein R^4 is chloro.

5. The method of claim **1**, wherein R^3 is 5-methylpiperidin-3-yl.

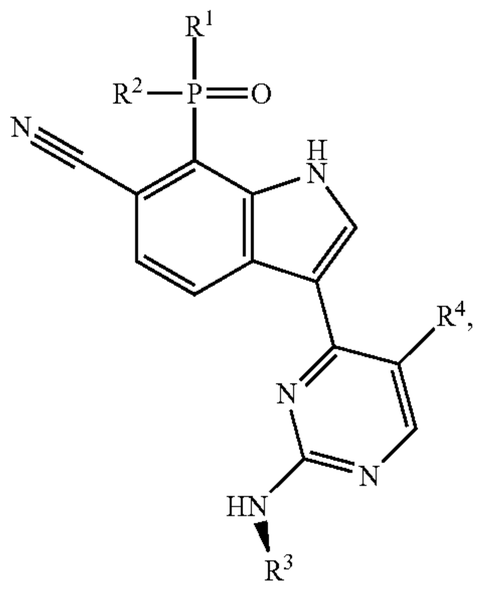
6. The method of claim **1**, wherein R^3 is 5,5-dimethylpiperidin-3-yl.

7. The method of claim **1**, wherein R^3 is 6-methylpiperdin-3-yl.

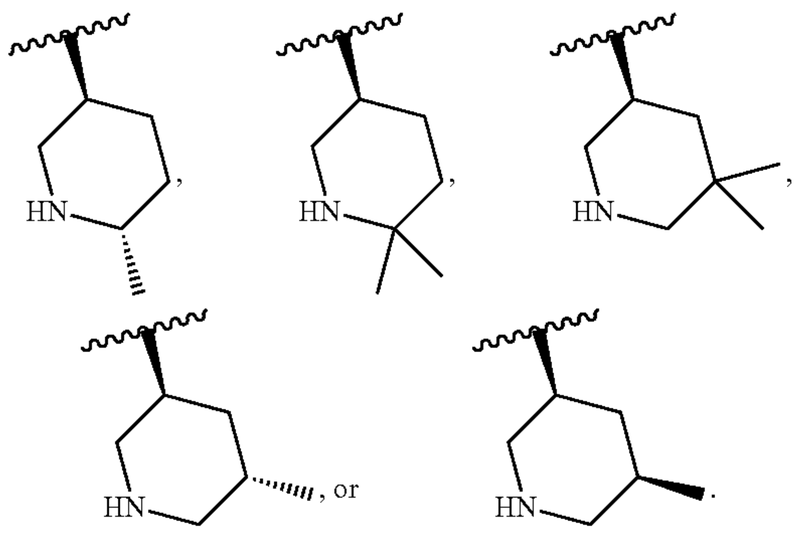
8. The method of claim **1**, wherein R^3 is 6,6-dimethylpiperidin-3-yl.

9. The method of claim **1**, wherein the compound has structural Formula (Ia):

(I)

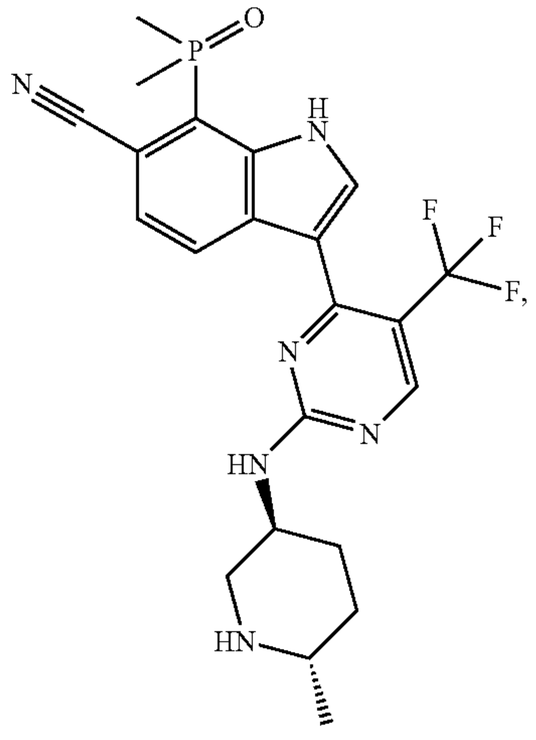


is a pharmaceutically acceptable salt thereof, wherein R^3 is



10.-**12**. (canceled)

13. The method of claim **9**, wherein the compound is:



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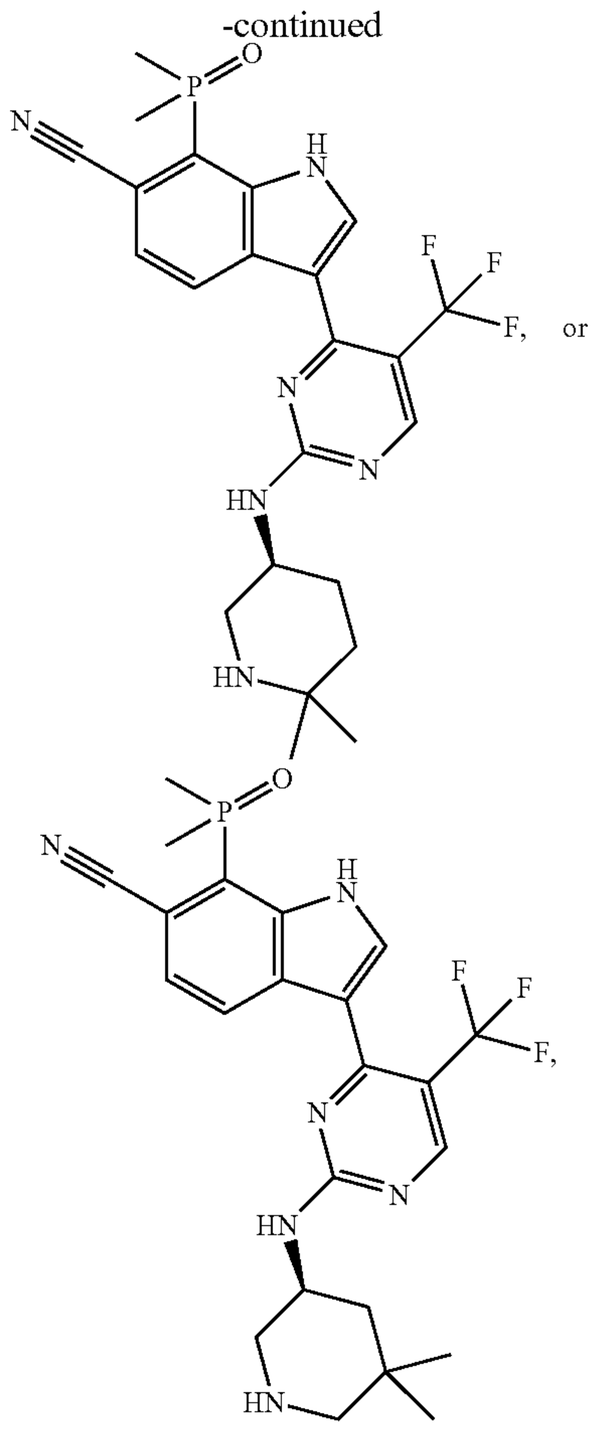
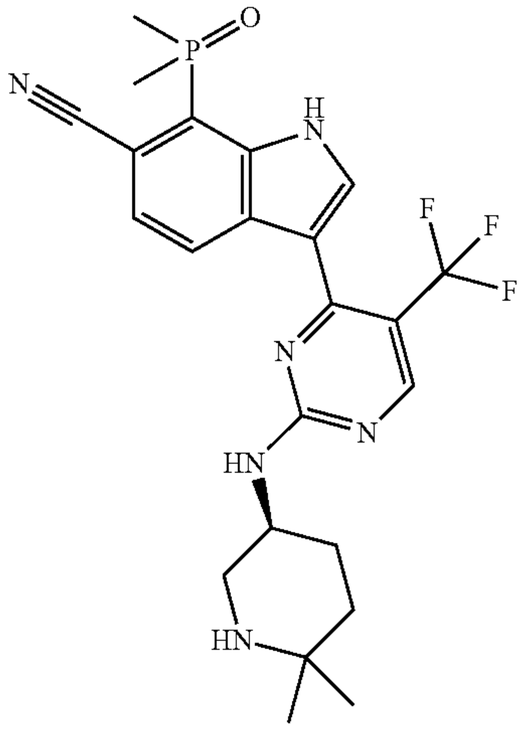


or is a pharmaceutically acceptable salt of any one of the foregoing compounds.

14. The method of claim **13**, wherein the compound is



or a pharmaceutically acceptable salt thereof.

15.-**18**. (canceled)

19. The method of claim **1**, wherein the cancer is a blood cancer, a breast cancer, Ewing's sarcoma, fallopian tube cancer, a GI tract cancer, a glioma, a lung cancer, melanoma,

an osteosarcoma, an ovarian cancer, a pancreatic cancer, a primary peritoneal cancer, prostate cancer, retinoblastoma, or a squamous cell cancer of the head or neck.

20. (canceled)

21. The method of claim **19**, wherein the patient has undergone, is presently undergoing, or is prescribed treatment with a Bcl-2 inhibitor.

22. The method of claim **21**, wherein the Bcl-2 inhibitor is venetoclax and/or wherein the patient has a breast cancer, a blood cancer, an ovarian cancer, or a lung cancer.

23. The method of claim **19**, wherein the patient has been determined to have a cancer in which

(a) RB1 or CDKN2A is mutated, contains an epigenetic alteration, is translocated, is transcribed at a level equal to or below a pre-determined threshold, or encodes a protein that is translated at a level equal to or below a pre-determined threshold or has decreased activity relative to a reference standard; and/or

(b) CDK6, CCND2, or CCNE1 is mutated, has a copy number alteration, contains an epigenetic alteration, is translocated, transcribed at a level equal to or above a pre-determined threshold, or encodes a protein that is translated at a level equal to or above a pre-determined threshold or has increased activity relative to a reference standard.

24. The method of claim **19**, wherein the patient has undergone, is presently undergoing, or is prescribed treatment with a selective estrogen receptor modulator (SERM), a selective estrogen receptor degrader (SERD), a PARP inhibitor, or a platinum-based therapeutic agent.

25. The method of claim **24**, wherein the patient has undergone, is presently undergoing, or is prescribed treatment with a SERM or SERD and has an HR+ breast cancer; the patient has undergone, is presently undergoing, or is prescribed treatment with a PARP inhibitor and has breast cancer, fallopian tube cancer, a glioma, ovarian cancer, or primary peritoneal cancer; or the patient has undergone, is presently undergoing, or is prescribed treatment with a platinum-based therapeutic agent and has an ovarian cancer.

26. The method of claim **19**, wherein the patient has undergone, is presently undergoing, or is prescribed treatment with a BET inhibitor with a CDK4/6 inhibitor; with a FLT3 inhibitor; or with a MEK inhibitor.

27. The method of claim **26**, wherein the patient who has undergone, is presently undergoing, or is prescribed treatment with the CDK4/6 inhibitor has a breast cancer, a pancreatic cancer, or a squamous cell cancer of the head or neck; the patient who has undergone, is presently undergoing, or is prescribed treatment with the FLT3 inhibitor has a blood cancer; or the patient who has undergone, is presently undergoing, or is prescribed treatment with the BET inhibitor has a breast cancer, a blood cancer, Ewing's sarcoma, or an osteosarcoma.

28. The method of claim **1**, wherein the patient has undergone, is presently undergoing, or is prescribed treatment with a second anti-cancer agent.

29. The method of claim **28**, wherein the second anti-cancer agent is a Bcl-2 inhibitor a CDK9 inhibitor; a hormone receptor degradation agent; a Flt3 (FMS-like tyrosine kinase 3) inhibitor; a PARP inhibitor; a BET inhibitor; a platinum-based therapeutic agent; a CDK4/6 inhibitor; a MEK inhibitor; or a phosphoinositide 3-kinase (PI3 kinase) inhibitor.

30. The method of claim **29**, wherein the Bcl-2 inhibitor is venetoclax, the PARP inhibitor is olaparib or niraparib, the platinum-based anti-cancer agent is carboplatin or oxaliplatin, the CDK4/6 inhibitor is palbociclib, ribociclib, abemaciclib, or trilaciclib, and the hormone receptor degradation agent is fulvestrant.

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