

Cancer Research Annual Meeting 2008, 99th:April 14, Abstract LB-146; Friedman et al, American Association for Cancer Research Annual Meeting 2008, 99th:April 14, Abstract LB-110) and shows synergistic activity *in vitro* and *in vivo* in combination with certain

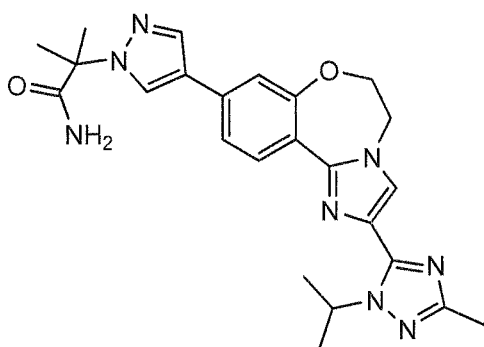
5 chemotherapeutic agents against solid tumor cell lines (US 2009/0098135).

GDC-0032 (Roche RG7604, CAS Reg. No. 1282512-48-4), named as 2-(4-(2-(1-isopropyl-3-methyl-1H-1,2,4-triazol-5-yl)-5,6-dihydrobenzo[f]imidazo[1,2-d][1,4]oxazepin-9-yl)-1H-pyrazol-1-yl)-2-methylpropanamide, has potent PI3K activity (WO 2011/036280; US 8242104; US 8343955) and is being studied in patients with locally advanced or metastatic solid
10 tumors.

SUMMARY OF THE INVENTION

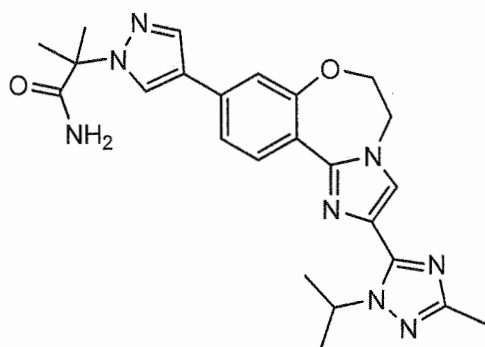
This application also includes any combination of the aspects and embodiments of the ~~invention disclosure~~ listed below.

It has been determined that additive or synergistic effects in inhibiting the growth of
15 cancer cells *in vitro* and *in vivo* can be achieved by administering the compound GDC-0032, or a pharmaceutically acceptable salt thereof, in combination with certain other specific chemotherapeutic agents. The combinations and methods may be useful in the treatment of hyperproliferative disorders such as cancer.



GDC-0032

20 ~~In one aspect, the invention disclosure~~ includes a method for the treatment of a hyperproliferative disorder comprising administering a therapeutic combination as a combined formulation or by alternation to a mammal, wherein the therapeutic combination comprises a therapeutically effective amount of GDC-0032 having the structure:



and a therapeutically effective amount of a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole.

- 5 In one ~~aspect~~ embodiment ~~the invention~~ disclosure provides a method for treating the hyperproliferative disorder wherein administration of GDC-0032 and one or more chemotherapeutic agents selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole provides a synergistic effect in treating the
- 10 hyperproliferative disorder. In a further ~~embodiment~~ aspect, the synergistic effect has a Combination Index value of less than about 0.8.

In one ~~embodiment~~ aspect ~~the invention~~ disclosure provides the therapeutic combination further including carboplatin.

- In one ~~embodiment~~ aspect ~~the invention~~ disclosure includes the therapeutic combination
- 15 further including an anti-VEGF antibody.

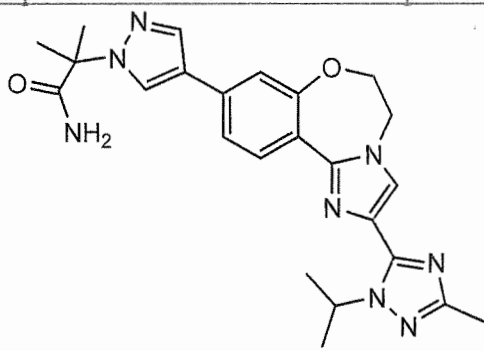
In one ~~embodiment~~ aspect ~~of the invention~~ disclosure includes the anti-VEGF antibody is bevacizumab.

- In one ~~embodiment~~ aspect ~~of the invention~~ disclosure includes the pharmaceutically acceptable salt of GDC-0032 is selected from a salt formed with hydrochloric acid, hydrobromic acid, hydroiodic acid, sulfuric acid, nitric acid, phosphoric acid, methanesulfonic acid,
- 20 benzenesulfonic acid, formic acid, acetic acid, trifluoroacetic acid, propionic acid, oxalic acid, malonic acid, succinic acid, fumaric acid, maleic acid, lactic acid, malic acid, tartaric acid, citric acid, ethanesulfonic acid, aspartic acid and glutamic acid.

- In one ~~embodiment~~ aspect ~~the invention~~ disclosure includes a method of treating a
- 25 hyperproliferative disorder in a patient comprising administering a therapeutically effective amount of GDC-0032, or a combination of GDC-0032 and a chemotherapeutic agent, to the patient, wherein a biological

sample obtained from the patient, prior to administration of the combination to the patient, has been tested for status of a biomarker, and wherein the biomarker status is indicative of therapeutic responsiveness by the patient to GDC-0032, or a combination of GDC-0032 and a chemotherapeutic agent. In one embodiment, the biological sample has been tested by measuring functional biomarker protein level, wherein an increased level of functional biomarker indicates that the patient will be resistant to GDC-0032 or the combination. In another embodiment, the biological sample has been tested by measuring functional biomarker level, wherein an increased or decreased level of functional biomarker indicates that the patient will be resistant to GDC-0032 or the combination.

10 In one aspect the disclosure includes a product comprising GDC-0032 having the structure:

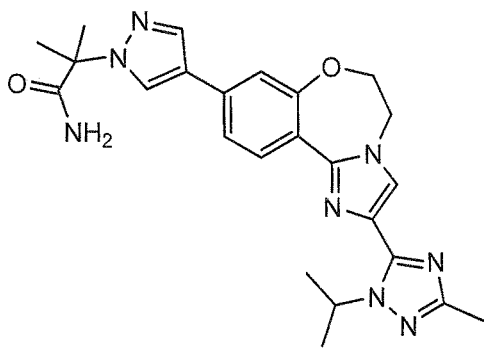


or a pharmaceutically acceptable salt thereof, and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine and trastuzumab; as a combined formulation for separate, simultaneous or sequential use in the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a mammal.

~~In one aspect the invention~~ The invention disclosure includes a pharmaceutical formulation comprising GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole. In another aspect the disclosure includes a pharmaceutical formulation comprising GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab.

~~In one aspect the invention~~ The invention disclosure includes a use of a therapeutic combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole in the manufacture of a medicament for the treatment of hyperproliferative disorder.

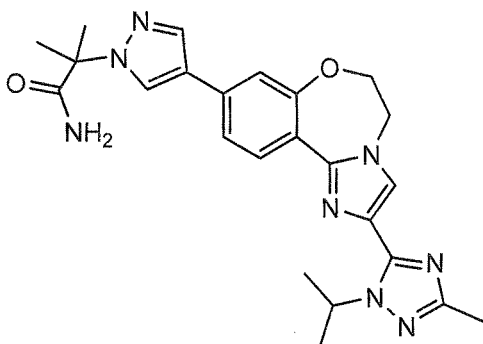
~~In one aspect~~ The invention disclosure includes a use of a therapeutic combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate. In another aspect the disclosure includes the use of a therapeutic combination of GDC-0032 having the structure:



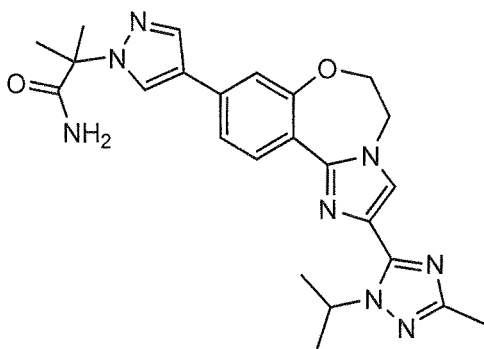
and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

~~In one aspect~~ The invention disclosure includes a therapeutic combination of GDC-0032 having the structure:

~~In one aspect (~~The invention-disclosure~~)~~ includes a therapeutic combination of GDC-0032 having the structure:

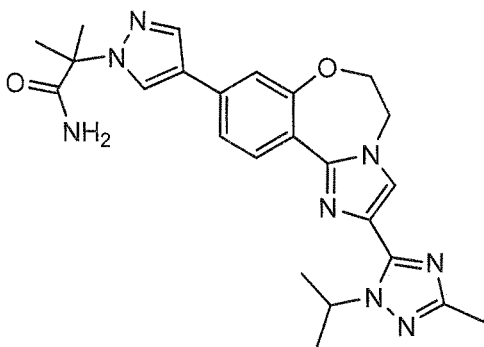


~~In one aspect~~ The invention disclosure includes the use of a therapeutic combination of
15 GDC-0032 having the structure:



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole in the treatment of a hyperproliferative disorder.

- 5 ~~In one aspect the invention~~ The invention disclosure includes the use of a therapeutic combination of GDC-0032 having the structure:



- and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab
10 emtansine, trastuzumab and letrozole in the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

~~In one aspect the invention includes the invention as hereinbefore described.~~

~~In one aspect the invention~~ The invention disclosure includes an article of manufacture for treating a hyperproliferative disorder comprising:

- 15 a) a therapeutic combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole; and
b) instructions for use.

In another aspect the disclosure includes an article of manufacture for treating cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate comprising:

a) a combined formulation as defined herein; and

5

b) instructions for use.

~~In one aspect the invention~~ disclosure includes a product comprising GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole; as a combined formulation for separate, simultaneous or sequential
5 use in the treatment of a hyperproliferative disorder.

~~In one embodiment aspect the invention~~ disclosure includes a method wherein the therapeutically effective amount of GDC-0032, and the therapeutically effective amount of the chemotherapeutic agent are administered as a combined formulation.

~~In one aspect embodiment the invention~~ disclosure includes the therapeutically effective
10 amount of GDC-0032, and the therapeutically effective amount of the chemotherapeutic agent are administered to a mammal by alternation.

~~In one embodiment aspect the invention~~ disclosure includes the mammal is administered the chemotherapeutic agent and subsequently administered GDC-0032.

~~In one embodiment aspect the invention~~ disclosure includes the therapeutic combination
15 is administered by a dosing regimen where the therapeutically effective amount of GDC-0032 is administered in a range from twice daily to once every three weeks, and the therapeutically effective amount of the chemotherapeutic agent is administered in a range from twice daily to once every three weeks.

~~In one embodiment aspect the invention~~ disclosure includes the dosing regimen is
20 repeated one or more times.

~~In one embodiment aspect the invention~~ disclosure includes administration of the therapeutic combination results in a synergistic effect.

~~In one embodiment aspect the invention~~ disclosure includes administration of the therapeutic combination results in a Combination Index value of less than about 0.7.

~~In one embodiment aspect the invention~~ disclosure includes the hyperproliferative
25 disorder is cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

~~In one embodiment aspect the invention~~ disclosure includes the cancer expresses a PIK3CA mutant selected from E542K, E545K, Q546R, H1047L and H1047R.

~~In one embodiment aspect the invention~~ disclosure includes the cancer expresses a PTEN
30 mutant.

In one ~~embodiment aspect the disclosure invention~~ includes the cancer is HER2 positive.

In one ~~embodiment aspect the disclosure invention~~ includes the mammal is a breast cancer patient wherein the patient is HER2 negative, ER (estrogen receptor) negative, and PR (progesterone receptor) negative.

5 In one ~~embodiment aspect the disclosure invention~~ includes the breast cancer subtype is Basal or Luminal.

In one ~~embodiment aspect the disclosure invention~~ includes the patient is administered GDC-0032 and eribulin.

10 In one ~~embodiment aspect the disclosure invention~~ includes GDC-0032 and the chemotherapeutic agent are each administered in an amount from about 1 mg to about 1000 mg per unit dosage form.

In one ~~embodiment aspect the disclosure invention~~ includes GDC-0032 and the chemotherapeutic agent are administered in a ratio of about 1:50 to about 50:1 by weight.

15 In one ~~embodiment aspect the disclosure invention~~ includes a pharmaceutical formulation comprising GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole.

20 In one ~~embodiment aspect the disclosure invention~~ includes the pharmaceutical formulation further comprising a pharmaceutically acceptable glidant selected from silicon dioxide, powdered cellulose, microcrystalline cellulose, metallic stearates, sodium aluminosilicate, sodium benzoate, calcium carbonate, calcium silicate, corn starch, magnesium carbonate, asbestos free talc, stearowet C, starch, starch 1500, magnesium lauryl sulfate, magnesium oxide, and combinations thereof.

25 In one ~~embodiment aspect the disclosure invention~~ includes the pharmaceutical formulation wherein GDC-0032 and the chemotherapeutic agent each comprise an amount from about 1 mg to about 1000 mg per unit dosage form.

In one ~~embodiment aspect the disclosure invention~~ includes the pharmaceutical formulation for use in a method of treatment of cancer.

30 ~~In one aspect the disclosure invention~~ includes a method for determining compounds to be used in combination for the treatment of cancer comprising:

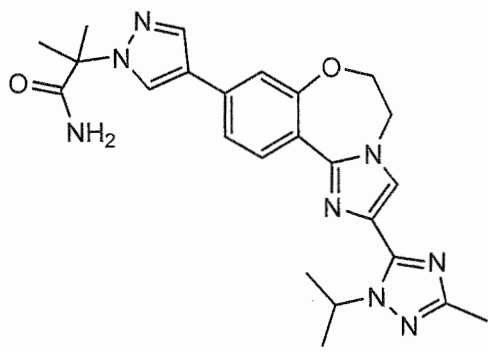
a) treating an in vitro tumor cell line with a K-ras mutation with a therapeutic combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole, and

5 b) measuring a synergistic or non-synergistic effect;

whereby a synergistic therapeutic combination for the treatment of cancer is determined.

In another aspect, the disclosure includes a method for determining compounds to be used in combination for the treatment of cancer comprising:

10 a) treating an in vitro tumor cell line with a K-ras mutation with a therapeutic combination of GDC-0032 having the structure



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab, and

15 b) measuring a synergistic or non-synergistic effect;

whereby a synergistic therapeutic combination for the treatment of cancer is determined.

~~In one aspect, the disclosure invention~~ The disclosure includes a method for selecting compounds to be used in combination for the treatment of cancer comprising:

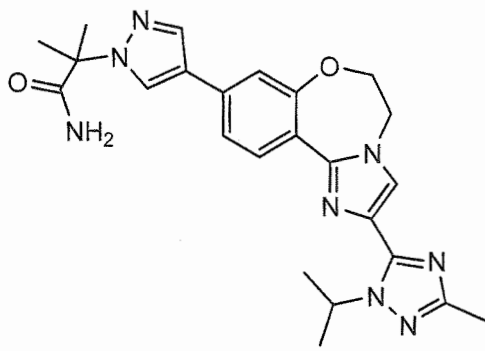
20 a) administering a therapeutic combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole to tumor cells;

b) measuring a change in pAkt level; and

c) selecting a synergistic therapeutic combination which shows an increase in pAkt levels.

In another aspect the disclosure includes a method for selecting compounds to be used in combination for the treatment of cancer comprising:

5 a) measuring a change in pAkt level in a tumour cell that had been administered a therapeutic combination of GDC-0032 having the structure



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and
10 trastuzumab; and

b) selecting a synergistic therapeutic combination which shows an increase in pAkt levels

~~In one aspect the~~ The disclosure invention includes a method of treating a hyperproliferative disorder in a patient comprising administering a therapeutically effective amount of GDC-0032
15 or a combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole to the patient, wherein a biological sample obtained from the patient, prior to administration of the combination to the patient, has been tested for PIK3CA or PTEN mutation status, and wherein PIK3CA or PTEN
20 mutation status is indicative of therapeutic responsiveness by the patient to the combination. In another aspect the disclosure includes a use of a combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a patient,
25 wherein a biological sample obtained from the patient, prior to administration of the combination to the patient, has been tested for PIK3CA or PTEN mutation status, and wherein PIK3CA or PTEN mutation status is indicative of therapeutic responsiveness by the patient to the

combination. The patient is administered a therapeutically effective amount of GDC-0032 as a single agent, or a combination of GDC-0032 and a chemotherapeutic agent. The biological sample may be tested by measuring functional PI3K protein level after administration of GDC-0032 or the combination of GDC-0032 and the chemotherapeutic agent, wherein a change in the
5 level of functional PI3K protein indicates that the patient will be resistant or responsive to GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent.

In one ~~aspect-embodiment~~ the disclosure invention includes the hyperproliferative disorder is HER2 expressing breast cancer.

In one ~~embodiment aspect~~ the disclosure invention includes a biological sample has been tested by measuring functional PI3K protein level after administration of GDC-0032 or the
5 combination of GDC-0032 and the chemotherapeutic agent, wherein a change in the level of functional PI3K protein indicates that the patient will be resistant or responsive to GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent.

~~In one aspect-embodiment~~ The disclosure invention includes a method of monitoring whether a patient with a hyperproliferative disorder will respond to treatment with GDC-0032 or a
10 combination of GDC-0032 and a chemotherapeutic agent, the method comprising:

(a) detecting a PIK3CA or PTEN mutation in a biological sample obtained from the patient following administration of the at least one dose of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab,
15 trastuzumab emtansine, trastuzumab and letrozole; and

(b) comparing PIK3CA or PTEN mutation status in a biological sample obtained from the patient prior to administration of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent to the patient,

wherein a change or modulation of PIK3CA or PTEN mutation status in the sample
20 obtained following administration of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent identifies a patient who will respond to treatment with GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent.

In another aspect the disclosure includes a method of monitoring whether a patient with cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma,
25 ovarian, pancreatic, and prostate will respond to treatment with a combination of GDC-0032 and a chemotherapeutic agent, the method comprising:

(a) detecting a PIK3CA or PTEN mutation in a biological sample obtained from the patient following administration of the at least one dose of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-
30 0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab; and

(b) comparing PIK3CA or PTEN mutation status in a biological sample obtained from the patient prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to the patient,

- 5 wherein a change or modulation of PIK3CA or PTEN mutation status in the sample obtained following administration of the combination of GDC-0032 and a chemotherapeutic agent identifies a patient who will respond to treatment with the combination of GDC-0032 and a chemotherapeutic agent.

~~In one aspect-embodiment the disclosure invention~~ includes the hyperproliferative disorder is HER2 expressing breast cancer.

- 10 ~~In one aspect-~~The disclosure invention includes a method of optimizing therapeutic efficacy of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent, the method comprising:

- (a) detecting a PIK3CA or PTEN mutation in a biological sample obtained from a patient following administration of at least one dose of GDC-0032 or the combination of GDC-0032 and
15 a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973,

GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole; and

- (b) comparing the PIK3CA or PTEN status in a biological sample obtained from the patient prior to administration of GDC-0032 or the combination of GDC-0032 and a
5 chemotherapeutic agent to the patient,

wherein a change or modulation of PIK3CA or PTEN in the sample obtained following administration of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent identifies a patient who has an increased likelihood of benefit from treatment with GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent.

- 10 In another aspect the disclosure includes a method of optimizing therapeutic efficacy of the combination of GDC-0032 and a chemotherapeutic agent in the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate, the method comprising:

- (a) detecting a PIK3CA or PTEN mutation in a biological sample obtained from a patient
15 following administration of at least one dose of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab; and

- (b) comparing the PIK3CA or PTEN status in a biological sample obtained from the patient prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to
20 the patient,

wherein a change or modulation of PIK3CA or PTEN in the sample obtained following administration of the combination of GDC-0032 and a chemotherapeutic agent identifies a patient who has an increased likelihood of benefit from treatment with the combination of GDC-0032 and a chemotherapeutic agent.

- 25 In one aspect embodiment the disclosure invention includes the hyperproliferative disorder is HER2 expressing breast cancer.

~~In one aspect the disclosure invention~~ The disclosure invention includes a method of identifying a biomarker for monitoring responsiveness to GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent, the method comprising:

- 30 (a) detecting the expression, modulation, or activity of a biomarker selected from a PIK3CA or PTEN mutation in a biological sample obtained from a patient who has received at least one dose of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent

selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole; and

(b) comparing the expression, modulation, or activity of the biomarker to the status of the biomarker in a reference sample wherein the reference sample is a biological sample obtained from the patient prior to administration of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent to the patient;

wherein the modulation of the biomarker changes by at least 2 fold lower or higher compared to the reference sample is identified as a biomarker useful for monitoring responsiveness to GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent. In one embodiment the biomarker is pAkt.

In another aspect the disclosure includes a method of identifying a biomarker for monitoring responsiveness to the combination of GDC-0032 and a chemotherapeutic agent in the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate, the method comprising:

(a) detecting the expression, modulation, or activity of a biomarker selected from a PIK3CA or PTEN mutation in a biological sample obtained from a patient who has received at least one dose of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab; and

(b) comparing the expression, modulation, or activity of the biomarker to the status of the biomarker in a reference sample wherein the reference sample is a biological sample obtained from the patient prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to the patient;

wherein the modulation of the biomarker changes by at least 2 fold lower or higher compared to the reference sample is identified as a biomarker useful for monitoring responsiveness to the combination of GDC-0032 and a chemotherapeutic agent.

In one ~~aspect-embodiment the disclosure invention~~ includes the hyperproliferative disorder is HER2 expressing breast cancer.

~~In one aspect-embodiment the disclosure invention~~ includes a method of treating a hyperproliferative disorder in a patient, comprising administering a therapeutically effective amount of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole to the patient, wherein treatment is based upon a sample from the patient having a PIK3CA or PTEN mutation. The biomarker mutation may be the H1047R, H1047L, E545K, or E542K mutation of PIK3CA.

In another aspect the disclosure includes a use of a combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a patient, wherein treatment is based upon a sample from the patient having a PIK3CA or PTEN mutation.

In one ~~aspect-embodiment the disclosure invention~~ includes the biomarker mutation is the H1047R or H1047L mutation of PIK3CA.

In one ~~embodiment aspect the disclosure invention~~ includes the biomarker mutation is the E542K, E545K or Q546R mutation of PIK3CA.

In one ~~embodiment aspect the disclosure invention~~ includes the hyperproliferative disorder is HER2 expressing breast cancer.

In one ~~embodiment aspect the disclosure invention~~ includes the hyperproliferative disorder is estrogen receptor positive (ER+) breast cancer.

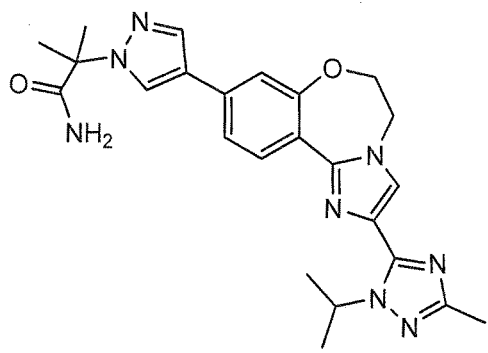
~~In one aspect-embodiment the disclosure invention~~ includes a use of GDC-0032 or a combination of GDC-0032 and a chemotherapeutic agent in treating a hyperproliferative disorder in a patient comprising administering a therapeutically effective amount of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, tamoxifen, fulvestrant, dexamethasone, pertuzumab, trastuzumab emtansine, trastuzumab and letrozole to the patient,

wherein a biological sample obtained from the patient, prior to administration of GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent to the patient, has been tested for PIK3CA or PTEN mutation status, and wherein PIK3CA or PTEN mutation status is indicative of therapeutic responsiveness by the patient to GDC-0032 or the combination of GDC-0032 and a chemotherapeutic agent.

In one embodiment aspect the disclosure invention includes the hyperproliferative disorder is HER2 expressing breast cancer.

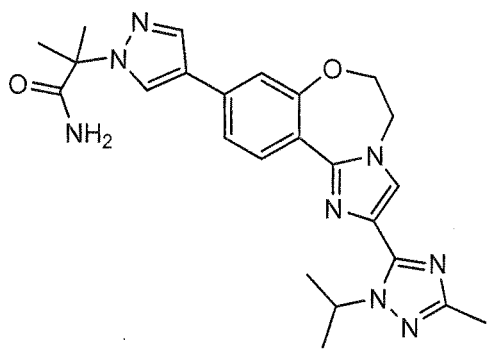
In one embodiment aspect the disclosure invention includes the hyperproliferative disorder is estrogen receptor positive (ER+) breast cancer.

- 5 In another aspect the disclosure includes therapeutic combination of GDC-0032 having the structure:



- and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and
- 10 trastuzumab for use in the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

In another aspect the disclosure includes the use of a therapeutic combination of GDC-0032 having the structure:



- 15 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows two plots of efficacy (EC₅₀ micromolar) of GDC-0032 and GDC-0941 (4-(2-(1H-indazol-4-yl)-6-((4-(methylsulfonyl)piperazin-1-yl)methyl)thieno[3,2-d]pyrimidin-4-yl)morpholine) in a cell proliferation (Cell-Titer Glo®, Promega) assay against PIK3CA wild type (WT) and mutant cell lines. Each dot represents a different cancer cell line.

Figure 2 shows two plots of efficacy (EC₅₀ micromolar) of GDC-0032 and GDC-0941 in cell proliferation assays against PIK3CA wild type (WT), PIK3CA mutant, HER2 expressing, and PI3K mutant/HER2 expressing cell lines. Each dot represents a different cell line.

Figure 3 shows three plots of efficacy (EC₅₀ micromolar) of GDC-0032 against: (3a, top) PIK3CA helical and kinase domain mutant cell lines; (3b, middle) PIK3CA wild type, PIK3CA mutant, PTEN null, and PTEN/PIK3CA mutant cell lines; and (3c, bottom) PIK3CA wild type, PIK3CA mutant, Ras mutant, and Ras/PIK3CA mutant cell lines in 4 day Cell-Titer Glo® viability assays. Each dot represents a different cell line.

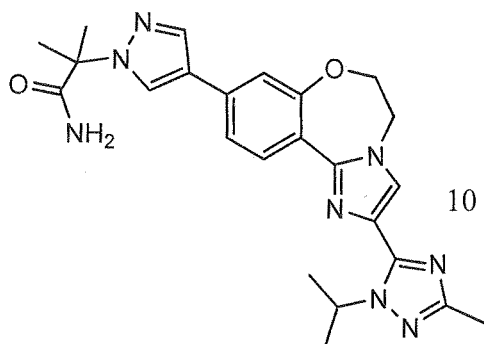
Figure 4a shows the potency of GDC-0032 in an SW48 isogenic cell line set. SW48 parental cells and knock-in mutant subclones harboring common PIK3CA hotspot mutations E545K or H1047R were obtained from Horizon Discovery. GDC-0032 EC₅₀ viability values were determined in these lines using a four day CellTiter-Glo® assay.

Figure 4b shows western blot autoradiograms of gel electrophoresis of cell lysates collected after 18 hours of GDC-0032 exposure at a range of concentrations in the SW48 isogenic cells; parental and PIK3CA knock-in mutants E545K and H1047R.

Figure 5a shows the effect of GDC-0032, paclitaxel (PTX), and the combination of GDC-0032 and paclitaxel on breast cancer cell line MFM223 with PIK3CA H1047R, and D350N. An in vitro assay (Cell-Titer Glo®, Promega) measured viable cells (RLU = Relative Light Units) over varying inhibitor concentrations by 2-fold dose titration of GDC-0032, paclitaxel (PTX), and the combination of GDC-0032 and paclitaxel.

Claims

1. ~~A product comprising method for the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate~~
5 ~~comprising administering a therapeutic combination as a combined formulation or by alternation to a mammal, wherein the therapeutic combination comprises a therapeutically effective amount of GDC-0032 having the structure:~~



~~or a pharmaceutically acceptable salt thereof, and a therapeutically effective amount of a~~
chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-
0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine and trastuzumab; as a
15 combined formulation for separate, simultaneous or sequential use in the treatment of cancer
selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian,
pancreatic, and prostate in a mammal.

2. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is 5-FU.
3. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is docetaxel.
- 20 4. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is eribulin.
5. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is gemcitabine.
6. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is GDC-0973.
7. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is GDC-0623.
- 25 8. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is paclitaxel.
9. The ~~product method~~ of claim 1 wherein the chemotherapeutic agent is fulvestrant.

10. The product method of claim 1 wherein the chemotherapeutic agent is pertuzumab.

11. The product method of claim 1 wherein the chemotherapeutic agent is trastuzumab emtansine.

5 12. The product method of claim 1 wherein the chemotherapeutic agent is trastuzumab.

13. The product method of claim 1 wherein the ~~therapeutic combination~~combined formulation further includes carboplatin.

10 14. The product method of claim 1 wherein the ~~therapeutic combination~~combined formulation further includes an anti-VEGF antibody.

15. The product method of claim 14 wherein the anti-VEGF antibody is bevacizumab.

15 16. The product method of any one of claims 1-15 wherein the pharmaceutically acceptable salt of GDC-0032 is selected from a salt formed with hydrochloric acid, hydrobromic acid, hydroiodic acid, sulfuric acid, nitric acid, phosphoric acid, methanesulfonic acid, benzenesulfonic acid, formic acid, acetic acid, trifluoroacetic acid, propionic acid, oxalic acid, malonic acid, succinic acid, fumaric acid, maleic acid, lactic acid, malic acid, tartaric acid, citric acid, ethanesulfonic acid, aspartic acid and glutamic acid.

20 ~~17. The method of any one of claims 1-16 wherein the therapeutically effective amount of GDC-0032, and the therapeutically effective amount of the chemotherapeutic agent are administered as a combined formulation.~~

~~18~~17. The product method of any one of claims 1-16 wherein the ~~therapeutically effective amount of GDC-0032, and the therapeutically effective amount of the~~ chemotherapeutic agent are to be administered to a mammal by alternation.

25 ~~19~~18. The product method of claim ~~18~~17 wherein the mammal is to be administered the chemotherapeutic agent and subsequently to be administered GDC-0032.

30 ~~20~~19. The product method of claim ~~18~~17 wherein the ~~therapeutic combination combined formulation is to be~~ administered by a dosing regimen where the ~~therapeutically effective amount of GDC-0032 is to be~~ administered in a range from twice daily to once every three weeks, and the ~~therapeutically effective amount of the chemotherapeutic agent is to be~~ administered in a range from twice daily to once every three weeks.

~~2021~~. The product method of claim ~~20-19~~ wherein the dosing regimen is repeated one or more times.

~~2221~~. The product method of any one of claims 1-16 wherein administration of the ~~therapeutic combination~~ combined formulation results in a synergistic effect.

5 ~~2322~~. The product method of claim 22 wherein administration of the ~~therapeutic combination~~ combined formulation results in a Combination Index value of less than about 0.7.

~~2423~~. The product method of claim 1 wherein the cancer expresses a PIK3CA mutant selected from E542K, E545K, Q546R, H1047L and H1047R.

~~2524~~. The product method of claim 1 wherein the cancer expresses a PTEN mutant.

10 ~~2625~~. The product method of claim 1 wherein the cancer is HER2 positive.

~~2726~~. The product method of claim 1 wherein the mammal is a breast cancer patient wherein the patient is HER2 negative, ER (estrogen receptor) negative, and PR (progesterone receptor) negative.

15 ~~2827~~. The product method of claim ~~27-26~~ wherein the breast cancer subtype is Basal or Luminal.

~~2928~~. The product method of claim 26 wherein the patient is to be administered GDC-0032 and eribulin.

20 ~~3029~~. The product method of any one of claims 1-16 wherein GDC-0032 and the chemotherapeutic agent are each to be administered in an amount from about 1 mg to about 1000 mg per unit dosage form.

~~3130~~. The product method of any one of claims 1-16 wherein GDC-0032 and the chemotherapeutic agent are to be administered in a ratio of about 1:50 to about 50:1 by weight.

25 ~~3231~~. A pharmaceutical formulation comprising GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab.

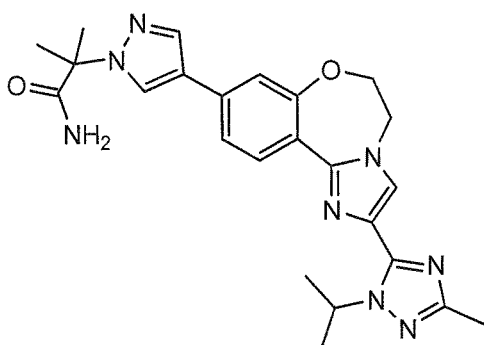
~~3332~~. The pharmaceutical formulation of claim ~~32-31~~ further comprising a pharmaceutically acceptable glidant selected from silicon dioxide, powdered cellulose, microcrystalline cellulose, metallic stearates, sodium aluminosilicate, sodium benzoate, calcium

carbonate, calcium silicate, corn starch, magnesium carbonate, asbestos free talc, stearowet C, starch, starch 1500, magnesium lauryl sulfate, magnesium oxide, and combinations thereof.

5 3433. The pharmaceutical formulation of claim 32-31 wherein GDC-0032 and the chemotherapeutic agent each comprise an amount from about 1 mg to about 1000 mg per unit dosage form.

3534. The pharmaceutical formulation of claim 32-33 for use in a method of treatment of cancer.

3635. The use of a therapeutic combination of GDC-0032 having the structure:

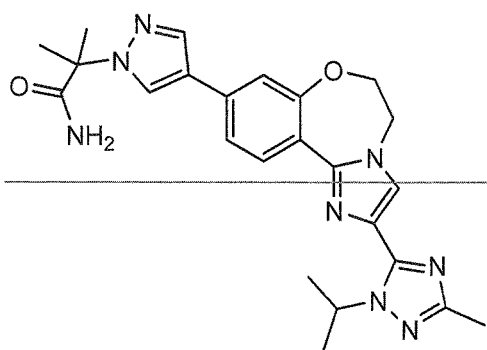


10 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

15 3736. An article of manufacture for treating cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate comprising:

- a) a ~~therapeutic combination~~ combined formulation of any one of claims 1-16; and
- b) instructions for use.

38. ~~A product comprising GDC-0032 having the structure:~~

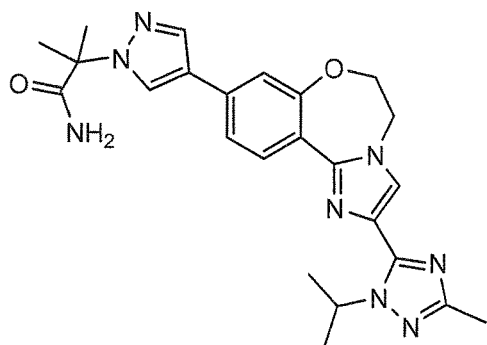


~~and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and~~

5 ~~trastuzumab; as a combined formulation for separate, simultaneous or sequential use in the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate.~~

~~3937.~~ A method for determining compounds to be used in combination for the treatment of cancer comprising:

10 a) treating an in vitro tumor cell line with a K-ras mutation with a therapeutic combination of GDC-0032 having the structure



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and

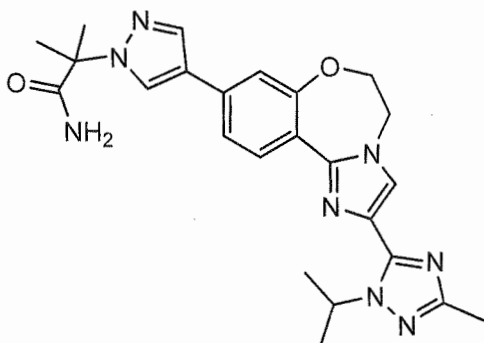
15 trastuzumab, and

b) measuring a synergistic or non-synergistic effect;

whereby a synergistic therapeutic combination for the treatment of cancer is determined.

4038. A method for selecting compounds to be used in combination for the treatment of cancer comprising:

a) measuring a change in pAkt level in a tumour cell that had been administered
~~administering~~ a therapeutic combination of GDC-0032 having the structure



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab to tumor cells; and

~~b) — measuring a change in pAkt level; and~~

eb) selecting a synergistic therapeutic combination which shows an increase in pAkt levels.

4139. ~~A use of a A method of treating cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a patient comprising administering a therapeutically effective amount of combination of GDC-0032 and a~~
chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate to in a the patient, wherein a biological sample obtained from the patient, prior to administration of the combination to the patient, has been tested for PIK3CA or PTEN mutation status, and wherein PIK3CA or PTEN mutation status is indicative of therapeutic responsiveness by the patient to the combination.

4240. The ~~method use~~ of claim 41-39 wherein the patient is to be administered a therapeutically effective amount of the combination of GDC-0032 and a chemotherapeutic agent.

4341. The ~~use method~~ of claim 41-39 wherein the cancer is HER2 expressing breast cancer.

42. The use of claim 39 wherein the cancer is estrogen receptor positive (ER+) breast cancer.

4443. The use method of claim 41-39 wherein a biological sample has been tested by measuring functional PI3K protein level after administration of the combination of GDC-0032 and the chemotherapeutic agent, wherein a change in the level of functional PI3K protein indicates that the patient will be resistant or responsive to the combination of GDC-0032 and a chemotherapeutic agent.

4544. A method of monitoring whether a patient with cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate will respond to treatment with a combination of GDC-0032 and a chemotherapeutic agent, the method comprising:

(a) detecting a PIK3CA or PTEN mutation in a biological sample obtained from the patient following administration of the at least one dose of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab; and

(b) comparing PIK3CA or PTEN mutation status in a biological sample obtained from the patient prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to the patient,

wherein a change or modulation of PIK3CA or PTEN mutation status in the sample obtained following administration of the combination of GDC-0032 and a chemotherapeutic agent identifies a patient who will respond to treatment with the combination of GDC-0032 and a chemotherapeutic agent.

4645. The method of claim 45-44 wherein the cancer is HER2 expressing breast cancer.

4746. A method of optimizing therapeutic efficacy of the combination of GDC-0032 and a chemotherapeutic agent in the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate, the method comprising:

(a) detecting a PIK3CA or PTEN mutation in a biological sample obtained from a patient following administration of at least one dose of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab; and

(b) comparing the PIK3CA or PTEN status in a biological sample obtained from the patient prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to the patient,

wherein a change or modulation of PIK3CA or PTEN in the sample obtained following administration of the combination of GDC-0032 and a chemotherapeutic agent identifies a patient who has an increased likelihood of benefit from treatment with the combination of GDC-0032 and a chemotherapeutic agent.

~~4847~~. The method of claim ~~45-44~~ wherein the cancer is HER2 expressing breast cancer.

~~4948~~. A method of identifying a biomarker for monitoring responsiveness to the combination of GDC-0032 and a chemotherapeutic agent in the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate, the method comprising:

(a) detecting the expression, modulation, or activity of a biomarker selected from a PIK3CA or PTEN mutation in a biological sample obtained from a patient who has received at least one dose of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab; and

(b) comparing the expression, modulation, or activity of the biomarker to the status of the biomarker in a reference sample wherein the reference sample is a biological sample obtained from the patient prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to the patient;

wherein the modulation of the biomarker changes by at least 2 fold lower or higher compared to the reference sample is identified as a biomarker useful for monitoring responsiveness to the combination of GDC-0032 and a chemotherapeutic agent.

~~5049~~. The method of claim ~~49-48~~ wherein the cancer is HER2 expressing breast cancer.

~~5150~~. A use of a method of treating cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a patient, comprising administering a therapeutically effective amount of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-

0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a to the patient, wherein treatment is based upon a sample from the patient having a PIK3CA or PTEN mutation.

5 ~~5251.~~ The ~~method-use~~ of claim ~~51-50~~ wherein the biomarker mutation is the H1047R or H1047L mutation of PIK3CA.

~~5352.~~ The ~~method-use~~ of claim ~~51-50~~ wherein the biomarker mutation is the E542K, E545K or Q546R mutation of PIK3CA.

10 ~~5453.~~ The ~~method-use~~ of claim ~~51-50~~ wherein the cancer is HER2 expressing breast cancer.

~~5554.~~ The ~~method-use~~ of claim ~~51-50~~ wherein the cancer is estrogen receptor positive (ER+) breast cancer.

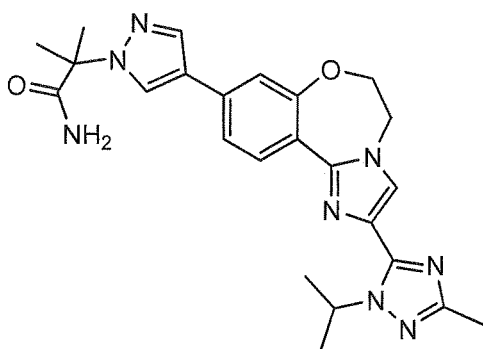
15 ~~56. A use of a combination of GDC-0032 and a chemotherapeutic agent in treating cancer selected from breast, cervical, colon, endometrial, gastric, glioma, lung, melanoma, ovarian, pancreatic, and prostate in a patient comprising administering a therapeutically effective amount of the combination of GDC-0032 and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab to the patient,~~

20 ~~wherein a biological sample obtained from the patient, prior to administration of the combination of GDC-0032 and a chemotherapeutic agent to the patient, has been tested for PIK3CA or PTEN mutation status, and wherein PIK3CA or PTEN mutation status is indicative of therapeutic responsiveness by the patient to the combination of GDC-0032 and a chemotherapeutic agent.~~

~~57. The use of claim 56 wherein the cancer is HER2-expressing breast cancer.~~

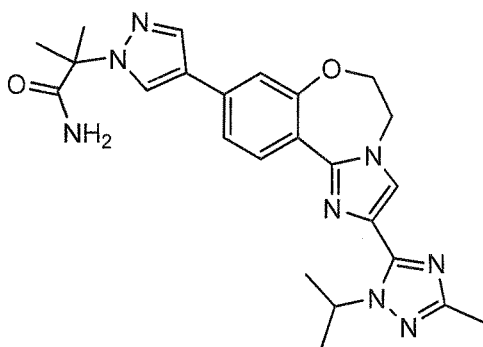
25 ~~58. The use of claim 56 wherein the cancer is estrogen receptor positive (ER+) breast cancer.~~

~~5955.~~ A therapeutic combination of GDC-0032 having the structure:



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab for use in the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.

6056. The use of a therapeutic combination of GDC-0032 having the structure:



and a chemotherapeutic agent selected from 5-FU, docetaxel, eribulin, gemcitabine, GDC-0973, GDC-0623, paclitaxel, fulvestrant, pertuzumab, trastuzumab emtansine, and trastuzumab in the manufacture of a medicament for the treatment of cancer selected from breast, cervical, colon, endometrial, glioma, lung, melanoma, ovarian, pancreatic, and prostate.