(S,E)-N-(1-CYCLOPROPYL-3-(METHYLSULFONYL)ALLYL)-2-(1,1-DIFLUOROETHYL)-4-PHENOXYPYRIMIDINE-5-CARBOXAMIDE FOR TREATING CANCER

FIELD

[0001] The present disclosure describes a covalent, irreversible inhibitor of WRN ATP-Dependent Helicase (WRNi), pharmaceutical compositions comprising the WRNi, and methods of using the WRNi to treat diseases associated with microsatellite instability (MSI-H).

BACKGROUND

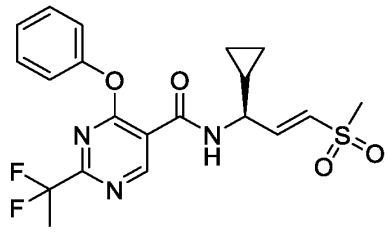
[0002] Microsatellite instability (MSI-H) is a genetic feature of a subset of tumors harboring deficits of the Mismatch repair pathway. MSI-H status is frequently observed across diverse indication including endometrial, stomach cancer, and colorectal cancer. In recent years, the development of immunotherapies revolutionized the management of several oncological indications and particularly high effectiveness of these therapies has been demonstrated in tumors presenting MSI-H status leading to clinical approval of these agents for this subset of patients. Despite the high effectiveness of Checkpoint Inhibitors (CPI) in MSI-H tumors approximately 50% of these patients do not respond to CPI supporting the development of novel therapeutic approaches for these patients.

[0003] Treatment standards for MSI and/or dMMR tumors have shifted in recent years with the approval of several checkpoint inhibitors. While responses to immune checkpoint blockade in MSI and/or dMMR solid tumors are often durable, 40% to 70% of such cancers do not respond to immune checkpoint blockade or develop secondary resistance, and the use of checkpoint inhibitors can be limited by toxicity. Thus, novel therapies are needed for patients with MSI and/or dMMR tumors.

[0004] Different checkpoint inhibitors have been approved for the treatment of refractory dMMR/MSI solid tumors, such as pembrolizumab, dostarlimab and nivolumab with or without ipilimumab. Despite high response rates and durable responses observed with these checkpoint inhibitors, a significant proportion of dMMR/MSI tumors do not respond or evolve resistance mechanisms (Schoenfeld, AJ and Hellmann, MD, “Acquired Resistance to Immune Checkpoint Inhibitors,” *Cancer Cell*, **2020**, *37*, 443-455). Thus, a need exists for methods for treating tumors that have developed resistance to or are unresponsive to, e.g., checkpoint inhibitor therapy.

SUMMARY

[0005] Provided herein is a compound having a structure of Formula (I):



(I)

or a pharmaceutically acceptable salt thereof, for use in the treatment of cancer, wherein the patient suffering from cancer was previously under treatment with a checkpoint inhibitor.

[0006] Also provided are methods for use in the treatment of cancer in a subject, comprising administering to the subject a therapeutically effective amount of a compound having a structure of Formula (I), or a pharmaceutically acceptable salt thereof.

[0007] Further aspects and advantages will be apparent to those of ordinary skill in the art from a review of the following detailed description, taken in conjunction with the drawings. While the compounds and methods disclosed herein are susceptible of cases in various forms, the description hereafter includes specific cases with the understanding that the disclosure is illustrative and is not intended to limit the invention to the specific cases described herein.

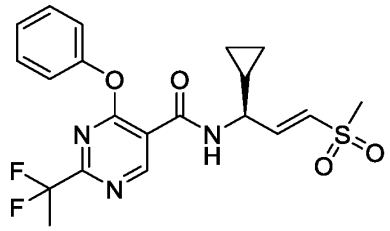
BRIEF DESCRIPTION OF THE DRAWINGS

[0008] FIGURE 1 shows (1) tumor volume and (2) Kaplan-Meyer curves of mice implanted with PDX2 and treated daily with the compound of formula (I) at the indicated doses.

[0009] FIGURE 2 shows (1) tumor volume and (2) Kaplan-Meyer curves of mice implanted with PDX3 and treated daily with the compound of formula (I) at the indicated doses.

DETAILED DESCRIPTION

[0010] Provided herein are methods and uses of an WRN inhibitor for the treatment of cancer, e.g., microsatellite instability (MSI-H) tumors, and/or in patients after relapse from a prior therapy such as an immunotherapy, wherein the WRN inhibitor is a compound having a structure of Formula (I):



(I)

or a pharmaceutically acceptable salt thereof. The compound of Formula (I) has an IUPAC name of (S,E)-N-(1-cyclopropyl-3-(methylsulfonyl)allyl)-2-(1,1-difluoroethyl)-4-phenoxypyrimidine-5-carboxamide. WO 2024/010782 provides a discussion of the compound of Formula (I) and how to synthesize it, the disclosure of which is incorporated by reference in its entirety. In some embodiments, the compound of Formula (I), or a pharmaceutically acceptable salt or solvate thereof, comprises at least one radioisotope. Particular examples of radioisotopes are ^{2}H, ^{3}H, ^{13}C, ^{14}C and ^{18}F.

[0011] It has been discovered as disclosed herein that a WRN inhibitor may be useful in treating certain types of cancers, and more specifically tumors. In some cases, the tumor is an MSI-H tumor.

[0012] A collection of Patient Derived Xenografts (PDX) from several patients including Colorectal cancer (CRC) presenting MSI-H were developed. For these patients, the clinical history is annotated and select 2 PDX models derived from the same patient. One of these models was derived from surgical material prior that the patient received immune therapy while a second PDX was derived after the tumor progressed after several lines of treatment including immune checkpoint inhibitors.

[0013] The aim of this study is to investigate whether a novel WRN inhibitor would present antitumor activity in both patient immunotherapy naïve and immunotherapy experienced.

[0014] In case activity in both models could be confirmed, it would imply that the WRN inhibitor would have utility in both patients progressing after immunotherapy and as a first line treatment (i.e., treatment naïve patients), in combination with immune checkpoint inhibitors, in the latter case to delay or reduce the onset of resistance acquisition.

[0015] The agent investigated, (S,E)-N-(1-cyclopropyl-3-(methylsulfonyl)allyl)-2-(1,1-difluoroethyl)-4-phenoxypyrimidine-5-carboxamide, is a novel covalent, irreversible inhibitor of WRN helicase that causes DNA damage accumulation and has selective antitumor activity in MSI cell lines and in vivo mouse models, including patient-derived models. Compound and synthesis are detailed in WO2024/010782.

[0016] The following definitions of the general terms used in the present description apply irrespectively of whether the terms in question appear alone or in combination with other groups.

[0017] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of the invention, suitable methods and materials are described below.

[0018] The term “inhibitor” denotes a compound which competes with, reduces, or prevents the binding of a particular ligand to particular receptor, or which reduces or prevents the function of a particular protein.

[0019] The terms “pharmaceutically acceptable carrier” and “pharmaceutically acceptable auxiliary substance” refer to carriers and auxiliary substances such as diluents or excipients that are compatible with the other ingredients of the formulation. Such a formulation is a formulation in pharmaceutically acceptable form.

[0020] The term "pharmaceutical composition" encompasses a product comprising specified ingredients in pre-determined amounts or proportions, as well as any product that results, directly or indirectly, from combining specified ingredients in specified amounts. In particular, it encompasses a product comprising one or more active ingredients, and an optional carrier comprising inert ingredients, as well as any product that results, directly or indirectly, from combination, complexation, or aggregation of any two or more of the ingredients, or from dissociation of one or more of the ingredients, or from other types of reactions or interactions of one or more of the ingredients.

[0021] “Therapeutically effective amount” means an amount of a compound that, when administered to a subject for treating a disease state, is sufficient to effect such treatment for the disease state. The “therapeutically effective amount” will vary depending on the compound, disease state being treated, the severity or the disease treated, the age and relative health of the subject, the route and form of administration, the judgment of the attending medical or veterinary practitioner, and other factors.

[0022] The term “as defined herein” and “as described herein” when referring to a variable incorporates by reference the broad definition of the variable as well as in particular, more particularly and most particular definitions, if any.

[0023] The terms “treating,” “contacting” and “reacting” when referring to a chemical reaction mean adding or mixing two or more reagents under appropriate conditions to produce the

indicated and/or the desired product. It should be appreciated that the reaction which produces the indicated and/or the desired product may not necessarily result directly from the combination of two reagents which were initially added, i.e., there may be one or more intermediates which are produced in the mixture which ultimately leads to the formation of the indicated and/or the desired product. Treatments include prophylactic treatment as well as the acute alleviation of symptoms.

[0024] The term “pharmaceutically acceptable excipient” denotes any ingredient having no therapeutic activity and being non-toxic such as disintegrators, binders, fillers, solvents, buffers, tonicity agents, stabilizers, antioxidants, surfactants, or lubricants used in formulating pharmaceutical products. As used herein, a "pharmaceutically acceptable carrier" or "pharmaceutically acceptable excipient" is intended to include any and all material compatible with pharmaceutical administration including solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying agents, and other materials and compounds compatible with pharmaceutical administration. Except insofar as any conventional media or agent is incompatible with the active compound, use thereof in the compositions of the invention is contemplated. Supplementary active compounds can also be incorporated into the compositions.

[0025] The corresponding pharmaceutically acceptable salts with acids can be obtained by standard methods known to the person skilled in the art, e.g., by dissolving the compound of Formula (I) in a suitable solvent such as e.g., dioxan or THF and adding an appropriate amount of the corresponding acid. The products can usually be isolated by filtration or by chromatography. The conversion of a compound of Formula (I) into a pharmaceutically acceptable salt with a base can be carried out by treatment of such a compound with such a base. One possible method to form such a salt is e.g. by addition of 1/n equivalents of a basic salt such as e.g. $M(OH)_n$, wherein M = metal or ammonium cation and n = number of hydroxide anions, to a solution of the compound in a suitable solvent (e.g. ethanol, ethanol-water mixture, tetrahydrofuran-water mixture) and to remove the solvent by evaporation or lyophilization.

[0026] In various embodiments, a patient has undergone a previous treatment before beginning treatment with a compound of Formula (I) or a pharmaceutically acceptable salt thereof, as disclosed herein. In various cases, that prior therapeutic regime is an immunotherapy. In some embodiments, the immunotherapy of the previous treatment was a checkpoint inhibitor. In some embodiments, the checkpoint inhibitor is a CTLA-4 inhibitor, a PD-1 inhibitor or a PD-L1 inhibitor. In some embodiments, the CTLA-4 inhibitor is ipilimumab (Yervoy®) or tremelimumab (GP-675,206). In some embodiments, the PD-1 inhibitor is selected from pembrolizumab (Keytruda®), nivolumab (Opdivo®) and RN888. In some

embodiments, the PD-L1 inhibitor is selected from atezolizumab (Tecentriq®), avelumab (Bavencio®) and durvalumab (Imfinzi™). In some embodiments, the PD-L1 inhibitor is atezolizumab (Tecentriq®).

[0027] In various embodiments, where the patient relapsed, the relapse occurred under a previous treatment with an immunotherapy.

[0028] In various embodiments, the cancer is thyroid cancer, colorectal cancer, melanoma, brain cancer or non-small cell lung cancer. In some embodiments, the cancer is colorectal cancer.

[0029] In some embodiments, the patient suffers from brain metastases.

[0030] Furthermore, the disclosure includes all optical isomers, i.e., diastereoisomers, diastereomeric mixtures, racemic mixtures, all their corresponding enantiomers and/or tautomers as well as their solvates, wherever applicable, of the compound of Formula (I).

[0031] If desired, racemic mixtures of the compound of Formula (I) may be separated so that the individual enantiomers are isolated. The separation can be carried out by methods well known in the art, such as the coupling of a racemic mixture of compounds to an enantiomerically pure compound to form a diastereomeric mixture, followed by separation of the individual diastereomers by standard methods, such as fractional crystallization or chromatography.

[0032] In embodiments where an optically pure enantiomer is provided, optically pure enantiomer means that the compound contains > 90 % of the desired isomer by weight, particularly > 95 % of the desired isomer by weight, or more particularly > 99 % of the desired isomer by weight, said weight percent based upon the total weight of the isomer of the compound. A chirally pure or chirally enriched compound may be prepared by chirally selective synthesis or by separation of enantiomers. The separation of enantiomers may be carried out on the final product or alternatively on a suitable intermediate.

[0033] Another embodiment provides a pharmaceutical composition containing the compound of Formula (I), or pharmaceutically acceptable salt thereof, for use according to the disclosure and one or more therapeutically inert carriers, diluents, or excipients, as well as a method to prepare such a pharmaceutical compositions. In one example, the compound of formula (I) may be formulated by mixing at ambient temperature at the appropriate pH, and at the desired degree of purity, with physiologically acceptable carriers, i.e., carriers that are non-toxic to recipients at the dosages and concentrations employed into a galenical administration form. The pH of the formulation depends mainly on the particular use and the concentration of compound, but preferably ranges anywhere from about 3 to about 8. In one

example, a compound of formula (I) is formulated in an acetate buffer, at pH 5. In another embodiment, the compound of formula (I) is sterile. The compound may be stored, for example, as a solid or amorphous composition, as a lyophilized formulation or as an aqueous solution.

[0034] Compositions are formulated, dosed, and administered in a fashion consistent with good medical practice. Factors for consideration in this context include the particular disorder being treated, the particular mammal being treated, the clinical condition of the individual patient, the cause of the disorder, the site of delivery of the agent, the method of administration, the scheduling of administration, and other factors known to medical practitioners.

[0035] Pharmaceutical compositions can be obtained by processing the compound of Formula (I) as described herein with pharmaceutically acceptable, inorganic, or organic carriers or excipients. Lactose, corn starch or derivatives thereof, talc, stearic acids or its salts and the like can be used, for example, as such carriers for tablets, coated tablets, dragées and hard gelatine capsules. Suitable carriers for soft gelatine capsules are, for example, vegetable oils, waxes, fats, semi-solid and liquid polyols, and the like. Depending on the nature of the active substance no carriers are, however, usually required in the case of soft gelatine capsules. Suitable carriers for the production of solutions and syrups are, for example, water, polyols, glycerol, vegetable oil and the like. Suitable carriers for suppositories are, for example, natural or hardened oils, waxes, fats, semi-liquid or liquid polyols and the like.

[0036] The pharmaceutical compositions can, moreover, contain preservatives, solubilizers, stabilizers, wetting agents, emulsifiers, sweeteners, colorants, flavorants, salts for varying the osmotic pressure, buffers, masking agents or antioxidants. They can also contain still other therapeutically valuable substances.

[0037] Pharmaceutical compositions of the compound of Formula (I), alone or in combination with a second anticancer agent, can be prepared for storage by mixing the active ingredients having the desired degree of purity with optional pharmaceutically acceptable carriers, excipients, or stabilizers (Remington's Pharmaceutical Sciences 16th edition, Osol, A. (ed.) (1980)), in the form of lyophilized formulations or aqueous solutions. Acceptable carriers, excipients, or stabilizers are nontoxic to recipients at the dosages and concentrations employed, and include buffers such as phosphate, citrate, and other organic acids; antioxidants including ascorbic acid and methionine; preservatives (such as octadecyldimethylbenzyl ammonium chloride; hexamethonium chloride; benzalkonium chloride, benzethonium chloride; phenol, butyl or benzyl alcohol; alkyl parabens such as

methyl or propyl paraben; catechol; resorcinol; cyclohexanol; 3-pentanol; and m-cresol); low molecular weight (less than about 10 residues) polypeptides; proteins, such as serum albumin, gelatin, or immunoglobulins; hydrophilic polymers such as polyvinylpyrrolidone; amino acids such as glycine, glutamine, asparagine, histidine, arginine, or lysine; monosaccharides, disaccharides, and other carbohydrates including glucose, mannose, or dextrins; chelating agents such as EDTA; sugars such as sucrose, mannitol, trehalose or sorbitol; salt-forming counter-ions such as sodium; metal complexes (e.g. Zn-protein complexes); and/or non-ionic surfactants such as TWEEN™, PLURONICS™ or polyethylene glycol (PEG).

[0038] Pharmaceutical compositions of the compound of Formula (I) include those suitable for oral, nasal, topical (including buccal and sublingual), rectal, vaginal and/or parenteral administration. The compositions may conveniently be presented in unit dosage form and may be prepared by any methods well known in the art of pharmacy. The amount of active ingredient which can be combined with a carrier material to produce a single dosage form will vary depending upon the host being treated, as well as the particular mode of administration. The amount of active ingredient which can be combined with a carrier material to produce a single dosage form will generally be that amount of the compound of Formula (I) which produces a therapeutic effect. Generally, out of one hundred percent, this amount will range from about 1 percent to about 90 percent of active ingredient, preferably from about 5 percent to about 70 percent, most preferably from about 10 percent to about 30 percent. Methods of preparing these compositions include the step of bringing into association the compound of Formula (I) with the carrier and, optionally, one or more accessory ingredients. In general, the pharmaceutical compositions can be prepared by uniformly and intimately bringing into association the compound of formula (I) with liquid carriers, or finely divided solid carriers, or both, and then, if necessary, shaping the product. Pharmaceutical compositions suitable for oral administration may be in the form of capsules, cachets, sachets, pills, tablets, lozenges (using a flavored basis, usually sucrose and acacia or tragacanth), powders, granules, or as a solution or a suspension in an aqueous or non-aqueous liquid, or as an oil-in-water or water-in-oil liquid emulsion, or as an elixir or syrup, or as pastilles (using an inert base, such as gelatin and glycerin, or sucrose and acacia) and/or as mouth washes and the like, each containing a predetermined amount of the compound of Formula (I) as an active ingredient.

[0039] The active ingredients may also be entrapped in microcapsules prepared, for example, by coacervation techniques or by interracial polymerization, for example, hydroxymethylcellulose or gelatin-microcapsules and poly- (methylmethacylate) microcapsules, respectively, in colloidal drug delivery systems (for example, liposomes,

albumin microspheres, microemulsions, nanoparticles and nanocapsules) or in macroemulsions. Such techniques are disclosed in Remington's Pharmaceutical Sciences, 16th edition, Osol, A. (ed.) (1980).

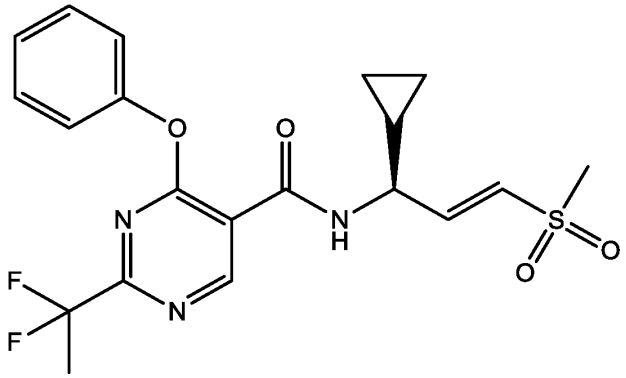
[0040] The formulations to be used for *in vivo* administration must be sterile. This can be readily accomplished by filtration through sterile filtration membranes.

[0041] The dosage can vary within wide limits and will, of course, have to be adjusted to the individual requirements in each particular case. In the case of oral administration the dosage for adults can vary from about 0.01 mg to about 1000 mg per day of a compound of general Formula (I) or of the corresponding amount of a pharmaceutically acceptable solvate thereof. The daily dosage may be administered as single dose or in divided doses and, in addition, the upper limit can also be exceeded when this is found to be indicated. In the case of oral administration the administration can be after a high-fat meal or following a 10-hour fast.

Embodiments of the Invention

[0042] All separate embodiments described below can be combined.

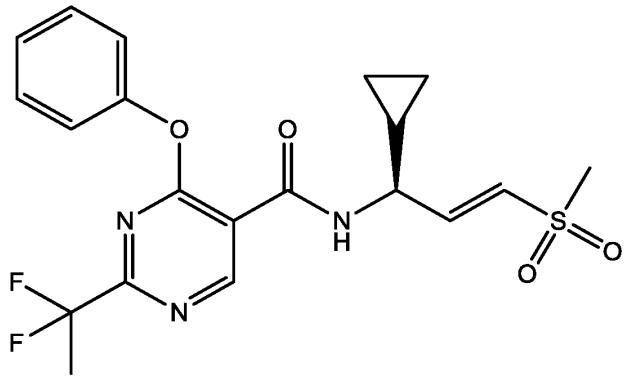
1. A compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof, for use in the treatment of cancer in a patient, wherein the patient suffering from said cancer was previously under treatment with a checkpoint inhibitor.

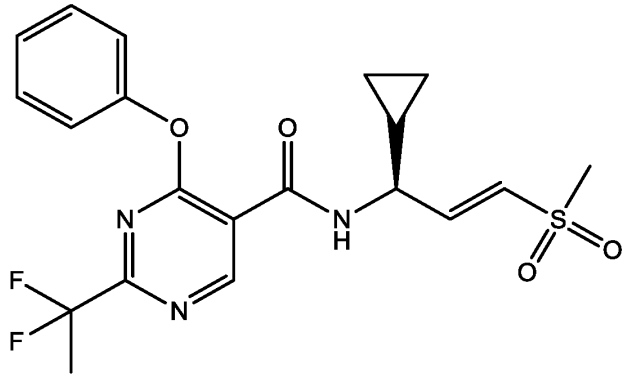
2. A compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof, for use in the treatment of relapsed cancer patients who have been previously treated with a checkpoint inhibitor.

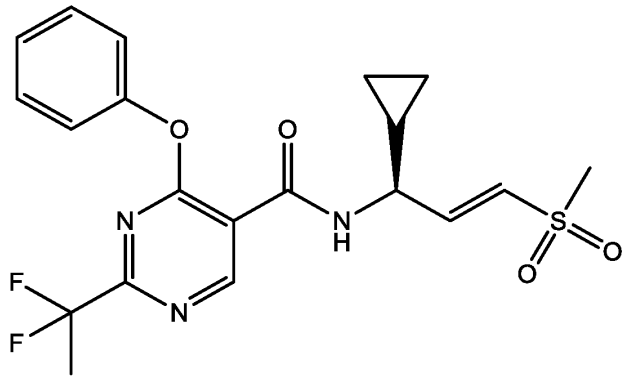
3. A compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof, for preventing formation of treatment resistance, in particular wherein the formation has been observed in patients having been treated with a checkpoint inhibitor.

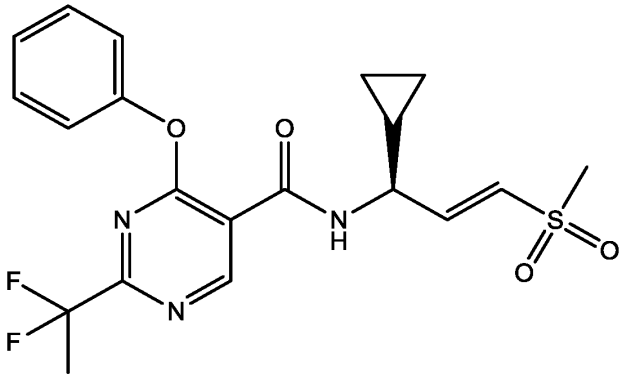
4. A compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof, for use in reducing treatment resistance of a cancer.

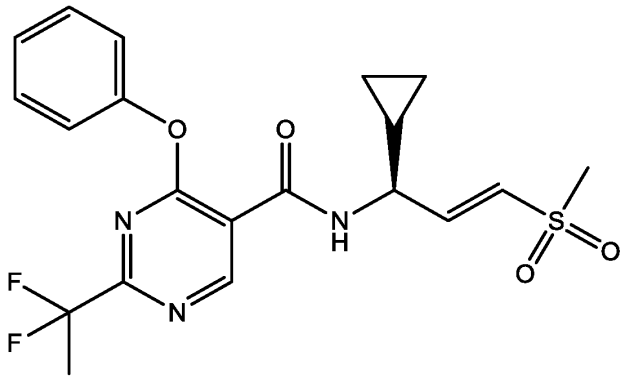
5. A compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof, for use in treating relapsing cancer with a checkpoint inhibitor.

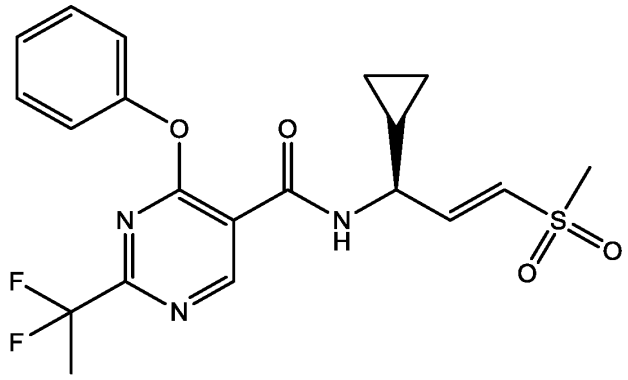
6. A method for the treatment of cancer, which method comprises administering an effective amount of the compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof to a patient in need thereof, wherein the patient suffering from said cancer was previously under treatment with a checkpoint inhibitor.

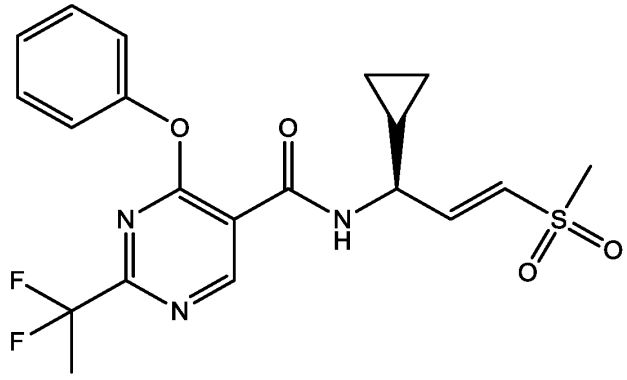
7. A method for reducing relapse growth of a cancer cell comprising administering a therapeutically effective amount of compound of formula (I),



(I)

or a pharmaceutically acceptable salt thereof, in particular wherein said administration reduces relapse growth of the cancer cell, more particularly wherein the relapse has been observed after prior administration of at least one checkpoint inhibitor.

8. The use of a compound of formula (I)



(I)

or a pharmaceutically acceptable salt thereof for the preparation of a medicament for the treatment of cancer, wherein the patient suffering from said cancer was previously treated with a checkpoint inhibitor.

9. The compound for use, method or use according to any one of embodiments 1 to 8, wherein the checkpoint inhibitor of the previous treatment is selected from a CTLA-4 inhibitor, a PD-1 inhibitor or a PD-L1 inhibitor, optionally wherein the CTLA-4 inhibitor is ipilimumab (Yervoy®) or tremelimumab (GP-675,206), optionally wherein the PD-1 inhibitor is selected from pembrolizumab (Keytruda®), nivolumab (Opdivo®) and RN888, optionally wherein the PD-L1 inhibitor is selected from atezolizumab (Tecentriq®), avelumab (Bavencio®) and durvalumab (Imfinzi™), or optionally wherein the PD-L1 inhibitor is atezolizumab (Tecentriq®).

10. The compound for use, method or use according to any one of embodiments 1 to 9, wherein tumor relapse occurred under the previous treatment.

11. The compound for use, method or use according to any one of embodiments 1 to 10, wherein the cancer is thyroid cancer, colorectal cancer, melanoma, brain cancer or non-small cell lung cancer.

12. The compound for use, method or use according to any one of embodiments 1 to 11, wherein the cancer is colorectal cancer.

13. The compound for use, method or use according to any one of embodiments 1 to 12, wherein the cancer is non-small cell lung cancer.

14. The compound for use, method or use according to any one of embodiments 1 to 13, wherein the patient is suffering from brain metastases.

15. The compound for use, method or use according to any one of embodiments 1 to 14, wherein the cancer is associated with microsatellite instability (MSI).

16. The compound of formula (I) in combination with a checkpoint inhibitor for use according to any one of embodiments 1-15.

17. The method according to any one of embodiments 1-16, wherein the patient is further administered a checkpoint inhibitor.

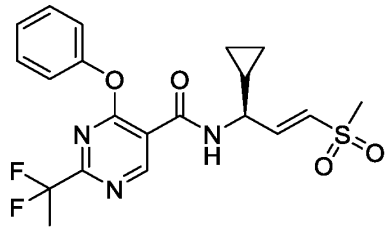
18. The method of claim 17, wherein the checkpoint inhibitor is a CTLA-4 inhibitor, a PD-1 inhibitor or a PD-L1 inhibitor, optionally wherein the CTLA-4 inhibitor is ipilimumab (Yervoy®) or tremelimumab (GP-675,206), or optionally wherein the PD-1 inhibitor is selected from pembrolizumab (Keytruda®), nivolumab (Opdivo®) and RN888, or optionally wherein the PD-L1 inhibitor is selected from atezolizumab (Tecentriq®), avelumab (Bavencio®) and durvalumab (Imfinzi™), or optionally wherein the PD-L1 inhibitor is atezolizumab (Tecentriq®).

19. The method according to embodiment 18, wherein the checkpoint inhibitor is atezolizumab (Tecentriq®).

20. The use of a compound of formula (I) or a pharmaceutically acceptable salt thereof according to any one of embodiments 1 to 19 in combination with a checkpoint inhibitor.

21. The use of embodiment 20 wherein the checkpoint inhibitor is atezolizumab (Tecentriq®).

22. A method of treating a patient suffering from a microsatellite instability (MSI-H) cancer, comprising administering to the patient a therapeutically effective amount of a compound of Formula (I):

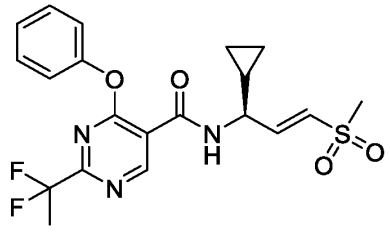


(I)

or pharmaceutically acceptable salt thereof.

23. The method of embodiment 22, wherein the patient was previously administered an immunotherapy prior to start of the administration of the compound of Formula (I).

24. A method of treating a cancer in a patient comprising administering to the patient a therapeutically effective amount of a compound of Formula (I):



(I)

or pharmaceutically acceptable salt thereof,
wherein the patient was previously administered an immunotherapy prior to start of the administration of the compound of Formula (I).

25. The method of embodiment 24, wherein the cancer is a microsatellite instability (MSI-H) cancer.

26. The method of any one of embodiments 23 to 25, wherein the immunotherapy was a checkpoint inhibitor.

27. The method of embodiment 26, wherein the checkpoint inhibitor was a CTLA-4 inhibitor, a PD-1 inhibitor, or a PD-L1 inhibitor.

28. The method of embodiment 27, wherein the CTLA-4 inhibitor was ipilimumab (Yervoy®) or tremelimumab (GP-675,206).

29. The method of embodiment 27, wherein the PD-1 inhibitor was pembrolizumab (Keytruda®), nivolumab (Opdivo®) or RN888.

30. The method of embodiment 27, wherein the PD-L1 inhibitor was atezolizumab (Tecentriq®), avelumab (Bavencio®) or durvalumab (Imfinzi™).

31. The method of embodiment 30, wherein the PD-L1 inhibitor was atezolizumab.

32. The method of any one of embodiments 23 to 31, wherein the patient relapsed from the immunotherapy.

33. The method of any one of embodiments 22 to 32, wherein the cancer is thyroid cancer, colorectal cancer, melanoma, brain cancer or non-small cell lung cancer.

34. The method of embodiment 33, wherein the cancer is colorectal cancer.

35. The method of any one of embodiments 22 to 33, wherein the patient is suffering from brain metastases.

36. The method of any one of embodiments 22 to 35, further comprising administering an immunotherapy to the patient.

37. The method of embodiment 36, wherein the wherein the immunotherapy is a checkpoint inhibitor.

38. The method of embodiment 37, wherein the checkpoint inhibitor is a CTLA-4 inhibitor, a PD-1 inhibitor, or a PD-L1 inhibitor.

39. The method of embodiment 37, wherein the CTLA-4 inhibitor is ipilimumab (Yervoy®) or tremelimumab (GP-675,206).

40. The method of embodiment 37, wherein the PD-1 inhibitor is pembrolizumab (Keytruda®), nivolumab (Opdivo®) or RN888.

41. The method of embodiment 37, wherein the PD-L1 inhibitor is atezolizumab (Tecentriq®), avelumab (Bavencio®) or durvalumab (Imfinzi™).

42. The method of embodiment 41, wherein the PD-L1 inhibitor is atezolizumab.

EXAMPLES

[0043] The following examples illustrate the present disclosure without limiting it but serve merely as representative thereof.

Material and Methods

[0044] A collection of Patient Derived Xenografts (PDX) from several patients including CRC presenting MSI-H was developed.

[0045] For these patients the clinical history is annotated and 2 PDX models were selected that were derived from the same patient. One of these models was derived from surgical material prior that the patient received immune therapy while, the second PDX was derived after the tumour progressed by several line of treatment including immune checkpoint inhibitors.

[0046] The aim of this study was to investigate whether a novel WRN inhibitor would present antitumor activity in both patient immunotherapy naïve and immunotherapy experienced.

[0047] In case activity in both models could be confirmed, it would imply that WRN inhibitor would have utility in both patients progressing immunotherapy and in first line, in combination with immune checkpoint inhibitors.

Experimental setup:

[0048] 2 mm x 2 mm tumor fragments of PDX2 and PDX3 were implanted subcutaneously in the right flank of NSG mice (NSG mouse (NOD scid gamma mouse) is a brand of immunodeficient laboratory mice). When tumor had grown to 100-300 mm^3, mice were randomized according to their tumor volume (mm^3). Four groups were established as described below:

[0049] For each PDX, a total of 52 NSG mice were distributed in four groups of 13 animals/group. The treatments administered orally to each group are:

(1) - Vehicle group: 13 NSG mice

(2) - Compound of formula (I) 5mg/Kg: 13 NSG mice

(3) - Compound of formula (I) 20mg/Kg: 13 NSG mice

(4) - Compound of formula (I) 75mg/Kg: 13 NSG mice

[0050] The Compound of formula (I) was administered daily through oral gavage at the doses of 5, 20 and 75 mg/kg to mice in groups 2, 3, and 4, respectively.

[0051] The animals were operated on and subcutaneously inoculating 100,000 cells/mouse. Once tumor growth started, measurements were taken. Tumor volume was measured twice weekly via caliper and mice euthanized when tumor reached a maximum of 1500 mm in diameter.

[0052] After the randomization, performed four weeks after operation on the animals, treatments were started. Each mouse was treated daily with 0.1 mL of the corresponding dose of compound or vehicle.

[0053] 24 hours post-first dose, three animals/group were sacrificed, collecting the tumors in two tubes, and immediately freezing in liquid nitrogen and stored at -80ºC.

[0054] Preparation of the vehicle (500mL) in drinking water treated by reverse osmosis as solvent:

HPMC: 1.25% (w/v) 6.25g in 500mL water

DOSS: 0.1% (w/v) 0.5g in 500mL water

Parabens: 0.2%(w/v) (9/1): Methylhydroxparabenzoate: 900mg in 500mL water; Propyhydroxyparabenzoate: 100mg in 500mL water

[0055] Preparation of the Compound Formulations:

75mg/kg: 750mg of compound (I) in 40mL of vehicle (0.1mL/mouse containing 1.875mg of compound).

20mg/kg: diluted from the 75mg/kg tube (dil 1:3,75)

5mg/kg: diluted from the 20mg/kg tube (dil 1:4)

Shaking overnight at 4ºC.

Results

[0056] Figure 1 top graph shows tumor volume and lower graph shows Kaplan-Meyer curves of mice implanted with PDX2 and treated daily with the compound of formula (I) at the indicated doses.

[0057] Figure 2 top graph shows tumor volume and lower graph shows Kaplan-Meyer curves of mice implanted with PDX3 and treated daily with the compound of formula (I) at the indicated doses.

[0100] Conclusion, the compound of Formula (I) showed activity in both PDXs. This datum suggests the use of the compound of formula (I) after relapse from immunotherapy or at first line in combo with immunotherapy for MSI-H patients.

[0101] The pharmaceutical compositions conveniently contain about 1-500 mg, particularly 5-250 mg, of a compound of Formula (I), e.g., 5, 25, 100, or 500 mg of the compound of Formula (I). In certain embodiments, the pharmaceutical compositions containing a compound of Formula (I) contains in addition about 1-500 mg, particularly 5-80 mg, of a MEK inhibitor in a fixed-dose combination.

[0102] Non-limiting examples of compositions according to the disclosure are:

EXAMPLE A-1

[0103] Tablets of the following composition are manufactured in the usual manner:

Table 1 – Tablet Compositions

ingredient	mg/tablet			
	5	25	100	500
(1) Compound of Formula (I)	5	25	100	500
(2) Lactose Anhydrous DTG	125	105	30	150
(3) Sta-Rx 1500	6	6	6	60
(4) Microcrystalline Cellulose	30	30	30	450
(5) Magnesium Stearate	1	1	1	1
Total weight of tablet	167	167	167	831

[0104] Manufacturing Procedure for Tablets of Table 1

1. Mix ingredients 1, 2, 3 and 4 and granulate with purified water.
2. Dry the granules at 50°C.
3. Pass the granules through suitable milling equipment.
4. Add ingredient 5 and mix for three minutes; and compress on a suitable press.

Example A-2

[0105] A film-coated tablet for oral administration may comprise the compound of formula (I) and the inactive ingredients sodium lauryl sulfate, microcrystalline cellulose, lactose monohydrate, magnesium oxide, colloidal silicon dioxide, sodium croscarmellose and magnesium stearate. The coating mixture may comprise polyvinyl alcohol, macrogol, talc, titanium dioxide, red iron oxide, and yellow iron oxide. These tablets can be administered QD or BID and conveniently contain about 5-500 mg, in particular 25 mg or 200 mg of the compound of Formula (I).

Example B-1

[0106] Capsules of the following composition are manufactured:

Table 2 – Capsules of the Compound of Formula (I)

ingredient	mg/capsule			
	5	25	100	500
(1) Compound of Formula (I)	5	25	100	500
(2) Hydrous Lactose	159	123	148	-
(3) Corn Starch	25	35	40	70
(4) Talc	10	15	10	25
(5) Magnesium Stearate	1	2	2	5
Total Weight in Capsule	200	200	300	600

[0107] Manufacturing Procedure for the Capsules of Table 2

1. Mix ingredients 1, 2 and 3 in a suitable mixer for 30 minutes.
2. Add ingredients 4 and 5 and mix for 3 minutes.
3. Fill into a suitable capsule.

[0108] The compound of Formula (I), lactose and corn starch are firstly mixed in a mixer and then in a comminuting machine. The mixture is returned to the mixer; the talc is added thereto and mixed thoroughly. The mixture is filled by machine into suitable capsules, e.g., hard gelatin capsules.

EXAMPLE B-2

[0109] Soft Gelatin Capsules of the following composition are manufactured with the components as listed in Table 3 or Table 4:

Table 3 – Soft Gelatin Capsules

Ingredient	mg/capsule
Compound of Formula (I)	5
Yellow wax	8
Hydrogenated Soya bean oil	8
Partially hydrogenated plant oils	34
Soya bean oil	110
Total weight in capsule	165

Table 4 – Soft Gelatin Capsules

Ingredient	mg/capsule
Gelatin	75
Glycerol 85 %	32
Karion 83	8 (dry matter)
Titanium dioxide	0.4
Iron oxide yellow	1.1
Total weight in capsule	116.5

[0110] Manufacturing Procedure

[0111] The compound of Formula (I) is dissolved in a warm melting of the other ingredients as listed in Table 3 or 4, and the mixture is filled into soft gelatin capsules of appropriate size. The filled soft gelatin capsules are treated according to the usual procedures.

EXAMPLE C

[0112] Suppositories of the composition as shown in Table 5 are manufactured:

Table 5 – Suppository Composition

ingredient	mg/supp.
Compound of Formula (I)	15
Suppository mass	1285

Total	1300

[0113] Manufacturing Procedure

[0114] The suppository mass is melted in a glass or steel vessel, mixed thoroughly, and cooled to 45°C. Thereupon, the finely powdered compound of Formula (I) is added thereto and stirred until it has dispersed completely. The mixture is poured into suppository molds of suitable size, left to cool; the suppositories are then removed from the molds and packed individually in wax paper or metal foil.

EXAMPLE D

[0115] Injection solutions of the composition as shown in Table 6 are manufactured:

Table 6 – Injection Solution Composition

ingredient	mg/injection solution.
Compound of Formula (I)	3
Polyethylene Glycol 400	150
acetic acid	q.s. ad pH 5.0
water for injection solutions	ad 1.0 ml

[0116] Manufacturing Procedure

[0117] The compound of Formula (I) is dissolved in a mixture of Polyethylene Glycol 400 and water for injection (part). The pH is adjusted to 5.0 by acetic acid. The volume is adjusted to 1.0 ml by addition of the residual amount of water. The solution is filtered, filled into vials using an appropriate overage and sterilized.

EXAMPLE E

[0118] Sachets of the following composition are manufactured:

Table 7 – Sachet Composition

ingredient	mg/sachet
Compound of Formula (I)	50
Lactose, fine powder	1015
Microcrystalline cellulose (AVICEL PH 102)	1400
Sodium carboxymethyl cellulose	14
Polyvinylpyrrolidone K 30	10
Magnesium stearate	10
Flavoring additives	1
Total	2500

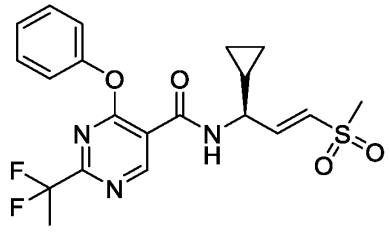
[0119] Manufacturing Procedure

[0120] The compound of Formula (I) is mixed with lactose, microcrystalline cellulose, and sodium carboxymethyl cellulose and granulated with a mixture of polyvinylpyrrolidone in water. The granulate is mixed with magnesium stearate and the flavoring additives and filled into sachets.

[0121] All of the features disclosed in this specification on WRN inhibitors (including any accompanying claims, abstract and drawings), and/or all of the steps of any method or process so disclosed, may be combined in any combination, except combinations where at least some of such features and/or steps are mutually exclusive. The invention is not restricted to the details of any embodiments explicitly disclosed herein on WRN inhibitors Any embodiment described in this application on WRN inhibitors can be combined with any other embodiment. The invention extends to any novel one, or any novel combination, of the features disclosed in this specification (including any accompanying claims, and abstract), or to any novel one, or any novel combination, of the any embodiment on WRN inhibitors, or any steps of any method or process so disclosed.

What is claimed is:

1. A compound of Formula (I):

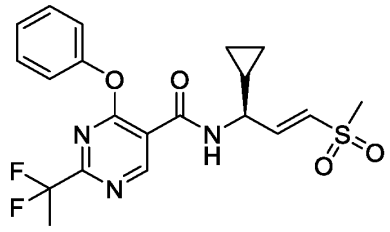


(I)

or a pharmaceutically acceptable salt thereof, for use in treating a patient suffering from a microsatellite instability (MSI-H) cancer.

2. The compound for use of claim 1, wherein the patient was previously administered an immunotherapy prior to start of the administration of the compound of Formula (I).

3. A compound of Formula (I):



(I)

or pharmaceutically acceptable salt thereof, for use in treating a cancer in a patient, wherein the patient was previously administered an immunotherapy prior to start of the administration of the compound of Formula (I).

4. The compound for use of claim 3, wherein the cancer is a microsatellite instability (MSI-H) cancer.

5. The compound for use of any one of claims 2 to 4, wherein the immunotherapy was a checkpoint inhibitor.

6. The compound for use of claim 5, wherein the checkpoint inhibitor was a CTLA-4 inhibitor, a PD-1 inhibitor, or a PD-L1 inhibitor.

7. The compound for use of claim 6, wherein the CTLA-4 inhibitor was ipilimumab (Yervoy®) or tremelimumab (GP-675,206).

8. The compound for use of claim 6, wherein the PD-1 inhibitor was pembrolizumab (Keytruda®), nivolumab (Opdivo®) or RN888.

9. The compound for use of claim 6, wherein the PD-L1 inhibitor was atezolizumab (Tecentriq®), avelumab (Bavencio®) or durvalumab (Imfinzi™).

10. The compound for use of claim 9, wherein the PD-L1 inhibitor was atezolizumab (Tecentriq®).

11. The compound for use of any one of claims 2 to 10, wherein the patient relapsed from the immunotherapy.

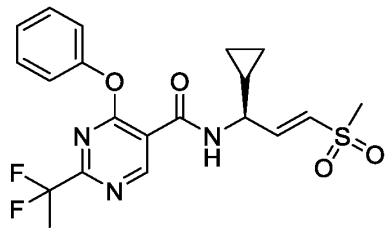
12. The compound for use of any one of claims 1 to 11, wherein the cancer is thyroid cancer, colorectal cancer, melanoma, brain cancer or non-small cell lung cancer.

13. The compound for use of claim 12, wherein the cancer is colorectal cancer.

14. The compound for use of any one of claims 1 to 13, wherein the patient is suffering from brain metastases.

15. The compound for use of any one of claims 1 to 14, wherein the patient is further administered a checkpoint inhibitor, wherein the checkpoint inhibitor is a CTLA-4 inhibitor selected from ipilimumab (Yervoy®) and tremelimumab (GP-675,206), a PD-1 inhibitor selected from pembrolizumab (Keytruda®), nivolumab (Opdivo®) and RN888, or a PD-L1 inhibitor selected from atezolizumab (Tecentriq®), avelumab (Bavencio®) and durvalumab (Imfinzi™), preferably atezolizumab (Tecentriq®).

16. A method of treating a patient suffering from a microsatellite instability (MSI-H) cancer comprising administering to the patient a therapeutically effective amount of a compound of Formula (I):



(I)

or pharmaceutically acceptable salt thereof.

17. The method of claim 16, wherein the MSI-H cancer is thyroid cancer, colorectal cancer, melanoma, brain cancer or non-small cell lung cancer.

18. The method of claim 16 or 17, wherein the patient was previously administered (or relapsed from) an immunotherapy to treat the MSI-H cancer (i.e., the compound of Formula (I) is a second or higher line of therapy).

19. The method of claim 18, wherein the immunotherapy was a checkpoint inhibitor therapy.

20. The method of claim 19, wherein the checkpoint inhibitor was a CTLA-4 inhibitor, a PD-1 inhibitor, or a PD-L1 inhibitor.

21. The method of claim 20, wherein the CTLA-4 inhibitor was ipilimumab (Yervoy®) or tremelimumab (GP-675,206).

22. The method of claim 20, wherein the PD-1 inhibitor was pembrolizumab (Keytruda®), nivolumab (Opdivo®) or RN888.

23. The method of claim 20, wherein the PD-L1 inhibitor was atezolizumab (Tecentriq®), avelumab (Bavencio®) or durvalumab (Imfinzi™).

24. The method of claim 23, wherein the PD-L1 inhibitor was atezolizumab (Tecentriq®).

25. The method of claim 16 or 17, wherein the patient is treatment naïve for the MSI-H cancer (i.e., the compound of Formula (I) is a first line therapy).

26. The method of any one of claims 16 to 25, further comprising administering to the patient a checkpoint inhibitor.

27. The method of claim 26, wherein the checkpoint inhibitor is a CTLA-4 inhibitor selected from ipilimumab (Yervoy®) and tremelimumab (GP-675,206), a PD-1 inhibitor selected from pembrolizumab (Keytruda®), nivolumab (Opdivo®) and RN888, or a PD-L1 inhibitor selected from atezolizumab (Tecentriq®), avelumab (Bavencio®) and durvalumab (Imfinzi™), preferably atezolizumab (Tecentriq®).