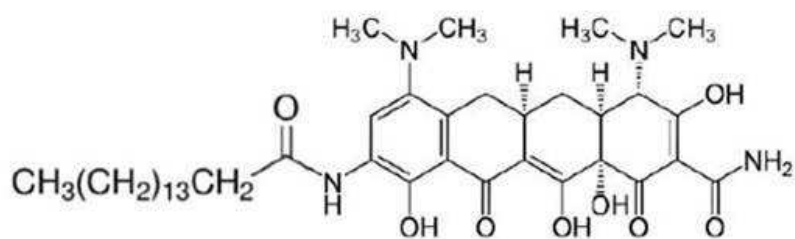


similarities between aerobic bacteria and mitochondria, certain classes of antibiotics or compounds having antibiotic activity can inhibit mitochondrial protein translation as an off-target side-effect.

SUMMARY

[0005] In view of the foregoing background, it is an object of this disclosure to demonstrate that existing antibiotics having intrinsic anti-mitochondrial properties can be chemically modified to target the mitochondria and thus can be used to eradicate CSCs. In one aspect, there is provided an antimitoscin comprising an antibiotic having intrinsic anti-mitochondrial properties and a membrane targeting signal comprising a fatty acid. In another aspect, there is provided use of a pharmaceutically effective amount of an antimitoscin and a pharmaceutically acceptable carrier in the manufacture of a medicament for treating cancer. In another aspect, there is provided use of a pharmaceutical composition in the manufacture of a medicament for treating cancer, said pharmaceutical composition containing, as the active ingredient, at least one antimitoscin. In a further aspect, there is provided an antimitoscin synthesis method comprising: chemically modifying an antibiotic having anti-cancer properties with a membrane targeting signal comprising a fatty acid. In a further aspect, there is provided a method for improving the anti-cancer properties of an antibiotic, the method comprising: chemically modifying an antibiotic having anti-cancer properties with a membrane targeting signal comprising a fatty acid. In a further aspect, there is provided a method for monitoring the effectiveness of an antimitoscin therapy, the method comprising: obtaining a tissue sample from a patient; determining the level of at least one CSC marker in the sample; and classifying the antimitoscin therapy as effective if the sample is determined to have a decreased level of at least one CSC marker relative to a threshold level. In

another aspect, there is provided an antimitoscin comprising the formula:



. Described herein are examples

of existing antibiotics having intrinsic anti-mitochondrial properties that have been chemically modified with one or more mitochondria-targeting signals that, as a result, have enhanced anti-cancer properties. The term “antimitoscin” used herein broadly refers to an antibiotic having intrinsic anti-mitochondrial properties that is chemically modified to target the antibiotic to mitochondria. The contemporary art considers intrinsic anti-mitochondrial activity in antibiotics to be an unwanted side-effect. Indeed, some potential antibiotics have been excluded from trials due to excessive anti-mitochondrial properties, and researchers have viewed anti-mitochondrial activity as a potential drawback. However, under the present approach, an antibiotic’s intrinsic anti-mitochondrial activity can become the basis for an entirely new therapeutic. The inventors have determined that these anti-mitochondrial properties may be harnessed and enhanced through chemical modification. As a result, antibiotics with intrinsic anti-mitochondrial activity may be repurposed as novel therapeutics for, among other potential therapies, anti-cancer treatments. These compounds may bind to either the large sub-unit or the small sub-unit of the mitochondrial ribosome (or in some instances, both) and inhibit mitochondrial biogenesis. Alternatively, these compounds may bind to the inner mitochondrial membrane to block the OXPHOS pathway and thus inhibit mitochondrial metabolism. The present disclosure further describes methods of

synthesizing antimitoscins, methods of using antimitoscins to target cancer stem cells, and pharmaceutical compositions for both treating cancer and for reducing drug resistance, the pharmaceutical compositions containing one or more antimitoscins as the active ingredient. The present disclosure further describes methods for monitoring the effectiveness of an antimitoscin therapy. The methods may include obtaining a tissue sample from a patient, determining the level of at least one CSC marker in the sample, and classifying the antimitoscin therapy as effective if the sample is determined to have a decreased level of at least one CSC marker relative to a threshold level. The CSC marker may be at least one of CD44, Sox2, Nanog, Oct 4, MYC, and ALDH.

[0006] The present disclosure may, in some examples, take the form of an antimitoscin. Exemplar antimitoscins are disclosed herein. In some examples, the antimitoscin comprises an antibiotic having intrinsic anti-mitochondrial properties and a mitochondria-targeting compound. In some examples, the antibiotic is a member of the tetracycline family, the erythromycin family, chloramphenicol, pyriminidyl pamoate, atovaquone, and bedaquiline. The mitochondria-targeting compound may be a chemical modification to the antibiotic. In some examples, the mitochondria-targeting compound is at least one compound selected from the group comprising a membrane targeting signal and a mitochondrial ribosome-targeting signal. In some examples, the membrane targeting signal is a compound selected from the group comprising palmitic acid, stearic acid, myristic acid, and oleic acid. In some examples, the mitochondrial targeting signal is selected from the group comprising tri-phenyl-phosphonium and guanidinium. In some examples, the antimitoscin possesses anti-cancer activity. In some examples, the antimitoscin binds to the large sub-unit or the small sub-unit of the mitochondrial ribosome. In some examples, the antimitoscin binds to at least one of the large

sub-units of the mitochondrial ribosome and the small sub-unit of the mitochondrial ribosome. In some examples, the antimitoscin binds to the inner mitochondrial membrane. In some examples, an antimitoscin possesses radiosensitizing activity, photosensitizing activity, sensitizes cancer cells to chemotherapeutic agents, sensitizes cancer cells to natural substances, and/or sensitizes cancer cells to caloric restriction. In some examples, the present disclosure relates to methods of treating cancer comprising administering to a patient in need thereof of a pharmaceutically effective amount of an antimitoscin and a pharmaceutically acceptable carrier. In some examples, the present disclosure relates to a pharmaceutical composition for treating cancer containing, as the active ingredient, at least one antimitoscin. In some examples, the pharmaceutical composition comprises a plurality of antimitoscins. Examples of the present approach may take the form of methods of synthesizing antimitoscins. Examples of the present approach may also take the form of improving the anti-cancer properties of an antibiotic.

[0007] The inventors analyzed phenotypic properties of CSCs that could be targeted across a wide range of cancer types, and identified a strict dependence of CSCs on mitochondrial biogenesis for the clonal expansion and survival of a CSC. Previous work by the inventors demonstrated that different classes of FDA-approved antibiotics, and in particular tetracyclines such as doxycycline and erythromycin, have an off-target effect of inhibiting mitochondrial biogenesis. As a result, such compounds have efficacy for eradicating CSCs. However, these common antibiotics were not designed to target the mitochondria, and therefore their anti-cancer efficacy is limited. Under the present approach, existing antibiotics having intrinsic anti-mitochondrial properties may be chemically modified to form antimitoscins, to target the mitochondria, and inhibit mitochondrial biogenesis and metabolism. Antimitoscins selectively inhibit CSCs because mitochondrial biogenesis is upregulated in CSCs and is required for

CLAIMS

What is claimed is:

1. An antimitoscin comprising an antibiotic having intrinsic anti-mitochondrial properties and a membrane targeting signal comprising a fatty acid.
2. The antimitoscin of claim 1, further comprising a mitochondria-targeting compound.
3. The antimitoscin of claim 1, wherein the antibiotic is selected from the group comprising at least one member of the tetracycline family, at least one member of the erythromycin family, chloramphenicol, pyruvium pamoate, atovaquone, and bedaquiline.
4. The antimitoscin of claim 2, wherein the mitochondria-targeting compound is at least one compound selected from the group comprising a membrane targeting signal and a mitochondrial ribosome-targeting signal.
5. The antimitoscin of claim 1, wherein the membrane targeting signal is a compound selected from the group comprising palmitic acid, stearic acid, myristic acid, and oleic acid.
6. The antimitoscin of claim 4, wherein the mitochondrial targeting signal is a compound selected from the group comprising tri-phenyl-phosphonium and guanidinium.
7. The antimitoscin of claim 1, wherein the antimitoscin possesses anti-cancer activity.
8. The antimitoscin of claim 1, wherein the antimitoscin binds to the large sub-unit of the mitochondrial ribosome.
9. The antimitoscin of claim 1, wherein the antimitoscin binds to the small sub-unit of the mitochondrial ribosome.
10. The antimitoscin of claim 1, wherein the antimitoscin binds to at least one of the large sub-unit of the mitochondrial ribosome or the small sub-unit of the mitochondrial ribosome.
11. The antimitoscin of claim 1, wherein the antimitoscin binds to a mitochondrial membrane.

12. The antimitoscin of claim 1, wherein the antimitoscin possesses radiosensitizing activity.
13. The antimitoscin of claim 1, wherein the antimitoscin possesses photosensitizing activity.
14. The antimitoscin of claim 1, wherein the antimitoscin sensitizes cancer cells to chemotherapeutic agents.
15. The antimitoscin of claim 1, wherein the antimitoscin sensitizes cancer cells to natural substances.
16. The antimitoscin of claim 1, wherein the antimitoscin sensitizes cancer cells to caloric restriction.
17. Use of a pharmaceutically effective amount of an antimitoscin and a pharmaceutically acceptable carrier in the manufacture of a medicament for treating cancer.
18. Use of a pharmaceutical composition in the manufacture of a medicament for treating cancer, said pharmaceutical composition containing, as the active ingredient, at least one antimitoscin.
19. The use of claim 18, wherein the pharmaceutical composition comprises a plurality of antimitoscins.
20. An antimitoscin synthesis method comprising:
chemically modifying an antibiotic having anti-cancer properties with a membrane targeting signal comprising a fatty acid.
21. The method of claim 20, wherein the antibiotic is selected from the group comprising at least one member of the tetracycline family, at least one member of the erythromycin family, chloramphenicol, pyruvium pamoate, atovaquone, and bedaquiline.
22. The method of claim 20, further comprising modifying the antimitoscin with at least one mitochondrial ribosome-targeting signal.

23. The method of claim 20, wherein the membrane targeting signal is at least one compound selected from the group comprising palmitic acid, stearic acid, myristic acid, and oleic acid.

24. The method of claim 22, wherein the mitochondrial targeting signal is at least one compound selected from the group comprising tri-phenyl-phosphonium and guanidinium.

25. A method for improving the anti-cancer properties of an antibiotic, the method comprising:

chemically modifying an antibiotic having anti-cancer properties with a membrane targeting signal comprising a fatty acid.

26. The method of claim 25, wherein the antibiotic is selected from the group comprising at least one member of the tetracycline family, at least one member of the erythromycin family, chloramphenicol, pyrvinium pamoate, atovaquone, and bedaquiline.

27. The method of claim 25, further comprising at least one mitochondrial ribosome-targeting signal.

28. The method of claim 25, wherein the membrane targeting signal is at least one compound selected from the group comprising palmitic acid, stearic acid, myristic acid, and oleic acid.

29. The method of claim 27, wherein the mitochondrial targeting signal is at least one compound selected from the group comprising tri-phenyl-phosphonium and guanidinium.

30. A method for monitoring the effectiveness of an antimitoscin therapy, the method comprising:

obtaining a tissue sample from a patient;

determining the level of at least one CSC marker in the sample;

and classifying the antimitoscin therapy as effective if the sample is determined to have a decreased level of at least one CSC marker relative to a threshold level.

31. The method of claim 30, wherein the CSC marker is at least one of CD44, Sox2, Nanog, Oct 4, MYC, and ALDH.

32. An antimitoscin comprising the formula:

