effective interventions that overcome the immune suppressive tumor microenvironment and for treating cancers associated with hypersialylated cancer cells.

SUMMARY OF THE INVENTION

[0006] The invention is based, in part, upon the discovery that it is possible to treat a sialic acid-mediated disorder by administering a sialidase enzyme or a sialidase enzyme conjugated to a serum half-life enhancer. Surprisingly, it has been discovered that a sialidase or a sialidase enzyme conjugated to a serum half-life enhancer that lacks a targeting moiety (*e.g.*, an antibody binding domain directed to a tumor antigen) can effectively treat a sialic acid-mediated disorder (*e.g.*, cancer, *e.g.*, a solid tumor) *in vivo*.

[0007] The invention further relates to recombinant forms of sialidase enzymes, sialidase enzymes conjugated to a serum half-life enhancer, and pharmaceutical compositions thereof, that have suitable substrate specificities and activities to be useful in removing sialic acid and/or sialic acid containing molecules from the surface of cancer cells and/or removing sialic acid and/or sialic acid containing molecules from the tumor microenvironment, and/or reducing the concentration of sialic acid and/or sialic acid containing molecules in the tumor microenvironment.

[0008] Thus, in certain aspects, the invention provides a pharmaceutical composition comprising or consisting essentially of a sialidase conjugated to a serum half-life enhancer that increases the serum half-life of the sialidase when administered to a subject.

[0008a] In one aspect, provided herein is a pharmaceutical composition comprising a sialidase conjugate comprising a sialidase conjugated to an Fc domain, wherein the sialidase is a recombinant mutant human Neu2 sialidase comprising the amino acid sequence of SEQ ID NO: 48.

[0008b] In another aspect, provided herein is the use of a pharmaceutical composition in the preparation of a medicament for the treatment of cancer in a subject in need thereof, wherein the pharmaceutical composition comprises an effective amount of a sialidase conjugate comprising a sialidase conjugated to an Fc domain,

wherein the sialidase is a recombinant mutant human Neu2 sialidase comprising the amino acid sequence of SEQ ID NO: 48.

[0008c] In another aspect, provided herein is the use of the pharmaceutical composition as described herein in the preparation of a medicament for the treatment of cancer in a subject.

[0008d] In another aspect, provided herein is the use of the pharmaceutical composition as described herein in the preparation of a medicament for increasing phagocytosis of a tumor cell in a subject, so as to increase phagocytosis of the tumor cell.

[0008e] In another aspect, provided herein is the use of the pharmaceutical composition as described herein in the preparation of a medicament for reducing Siglec-15 binding activity in a subject, so as to increase anti-tumor activity in the subject.

[0008f] In another aspect, provided herein is the pharmaceutical composition as described herein for use in therapy.

[0009] In another aspect, the invention provides a method of treating a sialic acid-related disorder in a subject in need thereof. The method includes administering to the subject an effective amount of a pharmaceutical composition comprising or consisting essentially of a sialidase and a serum half-life enhancer that increases the serum half-life of the sialidase when administered to the subject, thereby to treat the disorder.

[0010] In certain embodiments, the sialidase is not conjugated to a cancer antigen targeting agent that binds a cancer antigen associated with a cancerous cell.

[0011] In certain embodiments, the sialidase is a functional fragment of a full-length sialidase or a variant that exhibits at least 50% of the activity of the full-length sialidase.

[0012] In certain embodiments, the sialidase and the serum half-life enhancer are covalently linked together in a fusion protein or are chemically conjugated together.

WHAT IS CLAIMED IS:

1. A pharmaceutical composition comprising a sialidase conjugate comprising a sialidase conjugated to an Fc domain, wherein the sialidase is a recombinant mutant human Neu2 sialidase comprising the amino acid sequence of SEQ ID NO: 48.

2. The pharmaceutical composition of claim 1, wherein the sialidase is not conjugated to a cancer antigen targeting agent that binds a cancer antigen associated with a cancerous cell.

3. The pharmaceutical composition of claim 1 or 2, wherein the sialidase and the Fc domain are covalently linked together in a fusion protein.

4. The pharmaceutical composition of claim 1 or 2, wherein the sialidase and the Fc domain are chemically conjugated together.

5. The pharmaceutical composition of any one of claims 1-3, wherein the sialidase conjugate comprises the amino acid sequence of SEQ ID NO: 152.

6. The pharmaceutical composition of any one of claims 1-5, comprising from 0.01 mg/kg to 100 mg/kg of the sialidase.

7. The pharmaceutical composition of any one of claims 1-6, wherein the pharmaceutical composition comprises a second therapeutic agent.

8. The pharmaceutical composition of claim 7, wherein the second therapeutic agent is selected from the group consisting of an anti-inflammatory agent, anti-angiogenic agent, anti-fibrotic agent, or an anti-proliferative compound.

9. The pharmaceutical composition of claim 7, wherein the second therapeutic agent is an anti-CD20 antibody.

10. The pharmaceutical composition of any one of claims 1-9, wherein the pharmaceutical composition further comprises a stabilizing amount of a sialidase stabilizing agent.

11. The pharmaceutical composition of claim 10, wherein the sialidase stabilizing agent is a cation.

12. The pharmaceutical composition of claim 11, wherein the cation is selected from the group consisting of calcium and magnesium.

13. The pharmaceutical composition of any one of claims 1-12, wherein the

pharmaceutical composition is disposed in a sterile container.

14. The pharmaceutical composition of claim 13, wherein the pharmaceutical composition is lyophilized in the sterile container.

15. The pharmaceutical composition of claim 13, wherein the pharmaceutical composition is present as a solution in the sterile container.

16. The pharmaceutical composition of any one of claims 13-15, wherein the sterile container is sealed with a septum.

17. The pharmaceutical composition of any one of claims 13-16, wherein the sterile container has a label disposed thereon identifying the pharmaceutical composition contained in the container.

18. Use of a pharmaceutical composition in the preparation of a medicament for the treatment of cancer in a subject in need thereof, wherein the pharmaceutical composition comprises an effective amount of a sialidase conjugate comprising a sialidase conjugated to an Fc domain, wherein the sialidase is a recombinant mutant human Neu2 sialidase comprising the amino acid sequence of SEQ ID NO: 48.

19. The use of claim 18, wherein the sialidase is not conjugated to a cancer antigen targeting agent that binds a cancer antigen associated with a cancerous cell.

20. The use of claim 18 or 19, wherein the sialidase and the Fc domain are covalently linked together in a fusion protein.

21. The use of claim 18 or 19, wherein the sialidase and the Fc domain are chemically conjugated together.

22. The use of any one of claims 18-20, wherein the sialidase conjugate comprises the amino acid sequence of SEQ ID NO: 152.

23. The use of any one of claims 18-22, wherein from 0.01 mg/kg to 100 mg/kg of the sialidase is to be administered to the subject.

24. The use of any one of claims 18-23, wherein the cancer is a solid tumor, soft tissue tumor, hematopoietic tumor or metastatic lesion.

25. The use of claim 24, wherein the solid tumor is a sarcoma, adenocarcinoma, or carcinoma.

26. The use of claim 24 or 25, wherein the solid tumor is a head and neck, thyroid, lung, breast, lymphoid, gastrointestinal, genital or genitourinary tract, CNS, or skin tumor.

27. The use of claim 24, wherein the hematopoietic tumor is a leukemia, acute leukemia, acute lymphoblastic leukemia (ALL), B-cell, T-cell or FAB ALL, acute myeloid leukemia (AML), chronic myelocytic leukemia (CML), chronic lymphocytic leukemia (CLL), diffuse large B-cell lymphoma (DLBCL), follicular lymphoma, hairy cell leukemia, myelodysplastic syndrome (MDS), lymphoma, Hodgkin's disease, malignant lymphoma, non-Hodgkin's lymphoma, Burkitt's lymphoma, multiple myeloma, or Richter's Syndrome (Richter's Transformation).

28. The use of claim 24, wherein the cancer is breast cancer.

29. The use of claim 24, wherein the cancer is lymphoma.

30. The use of any one of claims 18-29, wherein administration of the pharmaceutical composition increases expression of granzyme B, IFNγ, IL-10, IL-6, or IL-17A in the subject.

31. The use of any one of claims 18-30, wherein the pharmaceutical composition is to be administered to the subject in combination with another therapeutic agent.

32. The use of claim 31, wherein the therapeutic agent is selected from the group consisting of an anti-inflammatory agent, anti-angiogenic agent, anti-fibrotic agent, or an anti-proliferative compound.

33. The use of claim 31, wherein the therapeutic agent is an anti-CD20 antibody.

34. The use of any one of claims 18-33, wherein the pharmaceutical composition further comprises a stabilizing amount of a sialidase stabilizing agent.

35. The use of claim 34, wherein the sialidase stabilizing agent is a cation.

36. The use of claim 35, wherein the cation is selected from the group consisting of calcium and magnesium.

37. The use of any one of claims 18-36, wherein the pharmaceutical composition, prior to administration, is disposed in a sterile container.

38. Use of the pharmaceutical composition of any one of claims 1-17 in the preparation of a medicament for the treatment of cancer in a subject.

39. Use of the pharmaceutical composition of any one of claims 1-17 in the preparation of a medicament for increasing phagocytosis of a tumor cell in a subject, so as to increase phagocytosis of the tumor cell.

40. Use of the pharmaceutical composition of any one of claims 1-17 in the preparation of a medicament for reducing Siglec-15 binding activity in a subject, so as to increase anti-tumor activity in the subject.

41. The pharmaceutical composition of any one of claims 1-17 for use in therapy.