BACKGROUND

Hyaluronan-degrading enzymes have been used therapeutically, typically as dispersing and spreading agents in combination with other therapeutic agents. Hyaluronan-degrading enzymes also can be used in single-agent therapy for the treatment of hyaluronan-associated diseases and disorders. For example, tumors and cancers are associated with accumulation of hyaluronan and treatment with a hyaluronan-degrading enzyme inhibits the growth of tumor and increases vascular perfusion and improves delivery of chemotherapeutic agents to the tumor. There exists a need for methods and reagents for improving treatment of patients who are treated with hyaluronan-degrading enzymes.

SUMMARY

The present disclosure provides a TSG-6-LM multimer, comprising:

a first polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a first multimerization domain; and

a second polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a second multimerization domain, wherein:

the first and second multimerization domains interact to form a multimer containing two or more TSG-6 link modules;

the first and second polypeptide do not comprise the full-length sequence of TSG-6; and

the TSG-6 multimer has a binding affinity to hyaluronan (HA) with an association constant of at least 10^7 M^{-1}.

The present disclosure provides a nucleic acid molecule encoding the TSG-6 multimer as described herein, wherein:

the nucleic acid molecule encodes a fusion polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a multimerization domain; and

the multimerization domain is a polypeptide that interacts with itself to form a stable protein-protein interaction, whereby the encoded protein forms a multimer containing at least two TSG-6 link modules.

The present disclosure provides a vector, comprising the nucleic acid molecule as described herein.

The present disclosure provides an isolated cell or cell culture, comprising the nucleic acid molecule as described herein.

The present disclosure provides a method of producing a TSG-6 multimer, comprising:

introducing the nucleic acid molecule as described herein into a cell;

culturing the cell under conditions whereby the fusion polypeptide is expressed by the cell; and

recovering the TSG-6 multimer.

The present disclosure provides a kit or combination, comprising:

the TSG-6 multimer as described herein; and

an anti-hyaluronan agent.

The present disclosure provides a method for selecting a subject for treatment of a tumor with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample previously obtained from a subject who has a tumor or cancer with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage; and

detecting binding of the hyaluronan binding protein to the sample, thereby determining the amount of hyaluronan in the sample, wherein if the amount of hyaluronan in the sample is at or above a predetermined threshold level, selecting the subject for treatment with an anti-hyaluronan agent.

The present disclosure provides a method of diagnosis of a hyaluronan-associated disease or condition, comprising:

contacting a tissue or body fluid sample previously obtained from a subject with TSG-6 or a hyaluronan binding region thereof; and

detecting binding of the TSG-6 or a hyaluronan-binding region thereof that binds to the sample, thereby determining the amount of hyaluronan in the sample, wherein the subject is diagnosed with a hyaluronan-associated disease or condition if the amount of hyaluronan is above a predetermined level or above the hyaluronan level of a reference sample.

The present disclosure provides a method for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample previously obtained from a subject who is or has been treated with an anti-hyaluronan agent with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage;

detecting binding of the hyaluronan binding protein to the sample, thereby determining the amount of hyaluronan in the sample, wherein detection of a decrease in hyaluronan compared to before treatment with anti-hyaluronan agent or the last dose of anti-hyaluronan agent indicates that the treatment is effective.

The present disclosure provides use of a hyaluronan binding protein (HABP) for manufacture of a composition for selecting a subject for treatment of a tumor with anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

The present disclosure provides a pharmaceutical composition comprising a hyaluronan binding protein (HABP) for use in selecting a subject for treatment of a tumor with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

The present disclosure provides use of a hyaluronan binding protein (HABP) for manufacture of a composition for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

The present disclosure provides a pharmaceutical composition comprising a hyaluronan binding protein (HABP) for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

The present disclosure provides use of a hyaluronan binding protein (HABP) for manufacture of a composition for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

The present disclosure provides use of an anti-hyaluronan agent for manufacture of a medicament for treatment of a hyaluronan-associated disease or condition in a subject selected for treatment based on the level or amount of hyaluronan (HA) in a sample from the subject as detected with a hyaluronan binding protein (HABP), wherein:

the subject selected for treatment exhibits a level or amount of hyaluronan (HA) in the sample that is at or above a predetermined level;

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

The present disclosure provides a pharmaceutical composition comprising an anti-hyaluronan agent for use in treatment of a hyaluronan-associated disease or condition in a subject selected for treatment based on the level or amount of hyaluronan (HA) in a sample from the subject as detected with a hyaluronan binding protein (HABP), wherein:

the subject selected for treatment exhibits a level or amount of hyaluronan (HA) in the sample that is at or above a predetermined level;

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

Provided herein is a method for selecting a subject for treatment of a tumor with an anti-hyaluronan agent, for example, a hyaluronan-degrading enzyme. In the provided method, a tissue or body fluid sample from a subject with a tumor or cancer is contacted with a hyaluronan binding protein (HABP) that has not been prepared from or isolated from animal cartilage. Binding of the hyaluronan binding protein to the sample is detected, thereby determining the amount of hyaluronan in the sample, wherein if the amount of hyaluronan in the sample is at or above a predetermined threshold level, selecting the subject for treatment with an anti-hyaluronan agent, for example, a hyaluronan degrading enzyme.

Provided herein is a method for selecting a subject for treatment of a tumor with an anti-hyaluronan agent, for example a hyaluronan-degrading enzyme, wherein a body fluid from a

subject with a tumor or cancer is contacted with a hyaluronan binding protein (HABP) that has not been prepared from or isolated from animal cartilage and binding of the HABP to the sample is effected by a solid-phase binding assay with a colorimetric or fluorescent signal, thereby determining the amount of hyaluronan in the sample, wherein a subject is selected for treatment with am anti-hyaluronan agent, for example a hyaluronan degrading enzyme, when the predetermined threshold level is high HA. In some examples of the method, the predetermined threshold level is at least or above 0.025 μg HA/ml of sample, 0.030 μg/ml, 0.035 μg/ml, 0.040 μg/ml, 0.045 μg/ml, 0.050 μg/ml, 0.055 μg/ml, 0.060

CLAIMS:

1. A method for selecting a subject for treatment of a tumor with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample from a subject who has a tumor or cancer with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage; and

detecting binding of the hyaluronan binding protein to the sample, thereby determining the amount of hyaluronan in the sample, wherein if the amount of hyaluronan in the sample is at or above a predetermined threshold level, selecting the subject for treatment with an anti-hyaluronan agent.

2. The method of any of claim 1, wherein the predetermined threshold level is high HA.

3. The method of claim 1 or claim 2, wherein the HABP molecule comprises a link module.

4. The method of claim 3, wherein the link module or modules is the only HABP portion of the molecule.

5. The method of any of claims 1-4, wherein the HABP molecule comprises a link module selected from among CD44, LYVE-1, HAPLN1/link protein, HAPLN2, HAPLN3, HAPLN4, aggrecan, versican, neurocan, brevican, phosphacan, TSG-6, Stabilin-1, Stabilin-2, CAB61358 and KIA0527 or a portion thereof comprising the link module or a sufficient portion of a link module to bind HA.

6. The method of any of claims 1-5, wherein the HABP molecule comprises a TSG-6 link module (LM) or a sufficient portion thereof that specifically binds HA.

7. A method for selecting a subject for treatment of a hyaluronan-associated disease or condition with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample from a subject with a hyaluronan-associated disease or condition with TSG-6 or a hyaluronan binding region thereof; and

detecting binding of the TSG-6 or a hyaluronan-binding region thereof to the sample, thereby determining the amount of hyaluronan in the sample, wherein if the amount of hyaluronan in the sample is at or above a predetermined threshold level, selecting the subject for treatment with an anti-hyaluronan agent.

8. The method of any of claims 1-7, wherein the predetermined threshold level is at least or above 0.025 μg HA/ml of sample, 0.030 μg/ml, 0.035 μg/ml, 0.040 μg/ml, 0.045 μg/ml, 0.050 μg/ml, 0.055 μg/ml, 0.060 μg/ml, 0.065 μg/ml, 0.070 μg/ml, 0.08 μg/ml, 0.09 μg/ml. 0.1 μg/ml, 0.2 μg/ml, 0.3 μg/ml or higher.

9. The method of any of claims 1-8, wherein the sample is a tumor tissue sample, and detection is effected by histochemistry.

10. The method of claim 9, wherein the predetermined threshold is an HA score of at least +2 (HA^{+2}) or at least +3 (HA^{+3}); or

(cells and stroma) to total stain in tumor tissue of at least 10%, 10% to 25% or greater than 25%.

11. The method of claim 9 or 10, wherein the predetermined threshold level is an HA score of at least +3 (HA^{+3}) (high levels).

12. (Amended) A method of diagnosis or prognosis of a hyaluronan-associated disease or condition, comprising:

contacting a tissue or body fluid sample from a subject with TSG-6 or a hyaluronan binding region thereof; and

detecting binding of the TSG-6 or a hyaluronan-binding region thereof that binds to the sample, thereby determining the amount of hyaluronan in the sample, wherein the subject is diagnosed with a hyaluronan-associated disease or condition if the amount of hyaluronan is above a predetermined level or above the hyaluronan level of a reference sample.

13. The method of any of claims 7-12, wherein the hyaluronan-associated disease or condition is a tumor or cancer.

14. The method of claim 12 or claim 13, wherein the predetermined or reference level is the median level of hyaluronan present in healthy or normal tissue or fluid samples.

15. The method of any of claims 12-14, wherein the predetermined threshold level is greater than 0.015 μg HA/ml of sample.

16. The method of any of claims 12-14, wherein the predetermined threshold level is an HA score of HA^{+1}, HA^{+2} or HA^{+3} as determined by histochemistry.

17. The method of any of claims 7-16, wherein the TSG-6 or a hyaluronan-binding region comprises a TSG-6 link module (LM) or a sufficient portion thereof that specifically binds HA.

18. The method of any of claims 6-17, wherein the TSG-6-LM has the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418, or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 and specifically binds HA.

19. The method of claim 18, wherein the HABP comprises a link module that exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO:207, 360, 417 or 418, whereby the HABP specifically binds HA.

20. The method of any of claims 6-19, wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

21. The method of claim 20, wherein binding to heparin is reduced at least 1.2-fold, 1.5-fold, 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, 10-fold, 20-fold, 30-fold, 40-fold, 50-fold, 100-fold or more.

22. The method of any of claims 6-21, wherein the TSG-6 link module comprises an amino acid replacement at an amino acid position corresponding to amino acid residue 20, 34, 41, 54, 56, 72 or 84 set forth in SEQ ID NO:360, whereby a corresponding amino acid residue is identified by alignment to a TSG-6-LM set forth in SEQ ID NO:360.

amino acid residue selected from among Asp (D), Glu (E), Ser (S), Thr (T), Asn (N), Gln (Q), Ala (A), Val (V), Ile (I), Leu (L), Met (M), Phe (F), Tyr (Y) and Trp (W).

24. The method of claim 22 or claim 23, wherein TSG-6 link module comprises an amino acid replacement corresponding to amino acid replacement K20A, K34A or K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

25. The method of any of claims 22-24, wherein TSG-6 link module comprises amino acid replacements corresponding to amino acid replacements K20A, K34A and K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

26. The method of any of claims 1-25, wherein the HABP or TSG-6 comprises a link module having the sequence of amino acids set forth in SEQ ID NO:361 or 416 or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO:361 or 416 that specifically binds HA.

27. The method of claim 26, wherein the HABP or TSG-6 comprises a link module that exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO:361 or 416, whereby the HABP specifically binds HA.

28. The method of any of claims 6-27, wherein the link module is the only TSG-6 portion of the HABP molecule.

29. The method of any of claims 1-5, wherein the HABP comprises a G1 domain of a type C hyaluronan binding protein.

30. The method of claim 29, wherein the HABP comprises a G1 domain selected from among Aggrecan G1, Versican G1, Neurocan G1 and Brevican G1.

31. The method of claim 29 or claim 30 wherein the G1 domain is the only HABP portion of the molecule.

32. The method of any of claims 1-31, wherein the HABP specifically binds to HA with a binding affinity that has a dissociation constant (Kd) of at least less than or less than 1×10^{-7} M, 9×10^{-8} M, 8×10^{-8} M, 7×10^{-8} M, 6×10^{-8} M, 5×10^{-8} M, 4×10^{-8} M, 3×10^{-8} M, 2×10^{-8} M, 1×10^{-8} M, 9×10^{-9} M, 8×10^{-9} M, 7×10^{-9} M, 6×10^{-9} M, 5×10^{-9} M, 4×10^{-9} M, 3×10^{-9} M, 2×10^{-9} M, 1×10^{-9} M or lower Kd.

33. The method of any of claims 1-32, wherein the HABP or TSG-6 is a multimer comprising a first HA-binding domain linked directly or indirectly via a linker to a multimerization domain and a second HA-binding domain linked directly or indirectly via a linker to a multimerization domain.

34. The method of claim 33, wherein the HA-binding domain is a link module or a G1 domain.

35. The method of claim 33 or claim 34, wherein the first and second HA-binding domain is the same or different.

36. The method of any of claims 33-35, wherein the first and second HA-binding domain is a TSG-6 link module, a variant thereof or a sufficient portion thereof that specifically binds to HA.

amino acids set forth in SEQ ID NO: 207, 360, 361, 416, 417 or 418 or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 361, 416, 417 or 418 that specifically binds HA.

38. The method of claim 37, wherein the link module exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 361, 416, 417 or 418, whereby the HABP specifically binds HA.

39. The method of any of claims 33-38, wherein the multimerization domain is selected from among an immunoglobulin constant region (Fc), a leucine zipper, complementary hydrophobic regions, complementary hydrophilic regions, compatible protein-protein interaction domains, free thiols that forms an intermolecular disulfide bond between two molecules, and a protuberance-into-cavity and a compensatory cavity of identical or similar size that form stable multimers.

40. The method of any of claims 33-39, wherein the multimerization domain is an Fc domain or a variant thereof that effects multimerization.

41. The method of claim 40, wherein the Fc domain is from an IgG, IgM or an IgE.

42. The method of claim 40 or claim 41, wherein the Fc domain has the sequence of amino acids set forth in SEQ ID NO:359.

43. The method of any of claims 1-42, wherein the HABP or TSG-6 is a fusion protein that contains a TSG-6 link module and an immunoglobulin Fc domain.

44. The method of any of claims 1-43, wherein the HABP or TSG-6 is TSG-6-LM-Fc that has the sequence of amino acids set forth in SEQ ID NO: 212 or 215 or a sequence of amino acids that exhibits at least 65% amino acid sequence identity to SEQ ID NO:212 and specifically binds HA.

45. The method of claim 44, wherein the HABP or TSG-6 has a sequence of amino acids that exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98% or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO:212 or 215, whereby the HABP or TSG-6 specifically binds HA.

46. The method of any of claim 1-45, wherein the step of contacting the sample with a HABP or TSG-6 is effected at between or about between pH 5.6 to 6.4.

47. The method of any of claims 1-46, further comprising treating the patient with an anti-hyaluronan agent.

48. The method of claim 47, wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

49. The method of claim 48, further comprising treating the subject with a hyaluronan-degrading enzyme.

50. The method of claim 49, wherein the hyaluronan-degrading enzyme is administered in a dosage range amount of between or about between 0.01 μg/kg (of the subject) to 50 μg/kg, 0.01 μg/kg to 20 μg/kg, 0.01 μg/kg to 15 μg/kg, 0.05 μg/kg to 10 μg/kg, 0.75 μg/kg to 7.5 μg/kg or 1.0 μg/kg to 3.0 μg/kg.

twice weekly, once weekly, once every 14 days, once every 21 days or once every month.

52. The method of any of claims 47-51, comprising administering a corticosteroid prior to administration with the anti-hyaluronan agent, with the anti-hyaluronan agent, or after administration with the anti-hyaluronan agent, wherein the corticosteroid is administered in an amount sufficient to ameliorate an adverse effect in the subject from the administered anti-hyaluronan agent.

53. The method of claim 52, wherein the amount of corticosteroid administered is between at or about 0.1 to 20 mgs, 0.1 to 15 mgs, 0.1 to 10 mgs, 0.1 to 5 mgs, 0.2 to 20 mgs, 0.2 to 15 mgs, 0.2 to 10 mgs, 0.2 to 5 mgs, 0.4 to 20 mgs, 0.4 to 15 mgs, 0.4 to 10 mgs, 0.4 to 5 mgs, 0.4 to 4 mgs, 1 to 20 mgs, 1 to 15 mgs or 1 to 10 mgs.

54. The method of claim 52 or claim 53, wherein the corticosteroid is a glucocorticoid.

55. The method of claim 54, wherein the glucocorticoid is selected from among cortisones, dexamethasones, hydrocortisones, methylprednisolones, prednisolones and prednisones.

56. A method for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample from a subject who is or has been treated with an anti-hyaluronan agent with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage;

detecting binding of the hyaluronan binding protein to the sample, thereby determining the amount of hyaluronan in the sample, wherein detection of a decrease in hyaluronan compared to before treatment with anti-hyaluronan agent or the last dose of anti-hyaluronan agent indicates that the treatment is effective.

57. A method for monitoring treatment of a subject with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample from a subject with a tumor or cancer with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage;

detecting the amount of hyaluronan binding protein that binds to the sample, thereby determining the amount of hyaluronan in the sample; and

comparing the level of hyaluronan to a control or reference sample to thereby determine the amount of hyaluronan in the sample relative to the control or reference sample, wherein the amount of hyaluronan is an indicator of the progress of treatment.

58. The method of claim 56 or 57, further comprising altering treatment based on the determined amount of hyaluronan in the sample relative to the control or reference sample, wherein:

if the amount of hyaluronan in the sample is at or above the amount in the control or reference sample, continuing treatment or escalating treatment by increasing the dosage and/or dose schedule of the anti-hyaluronan agent; or

or reference sample, continuing the treatment, reducing treatment by decreasing the dosage and/or dose schedule or terminating treatment of the anti-hyaluronan agent.

59. The method of claim 58, wherein the control or reference sample is a sample from a healthy subject, is a baseline sample from the subject prior to treatment with a an anti-hyaluronan agent or is a sample from a subject prior to the last dose of anti-hyaluronan agent.

60. The method of any of claims 56-59, wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

61. The method of claim 60, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme.

62. The method of any of claims 56-61, wherein the subject has a tumor or cancer.

63. The method of any of claims 56-62, wherein the HABP comprises a link module.

64. The method of claim 63, wherein the link module or modules is the only HABP portion of the molecule.

65. The method of any of claims 56-64 wherein the HABP comprises a link module selected from among CD44, LYVE-1, HAPLN1/link protein, HAPLN2, HAPLN3, HAPLN4, aggrecan, versican, neurocan, brevican, phosphacan, TSG-6, Stabilin-1, Stabilin-2, CAB61358 and KIA0527 or a portion thereof comprising the link module or a sufficient portion of a link module to bind HA.

66. The method of any of claims 56-65, wherein the HABP comprises a TSG-6 link module (LM) or a sufficient portion thereof that specifically binds HA.

67. The method of any of claims 56-66, wherein the HABP is a multimer comprising a first HA-binding domain linked directly or indirectly via a linker to a multimerization domain and a second HA-binding domain linked directly or indirectly via a linker to a multimerization domain.

68. The method of claim 67, wherein the first and second HA-binding domain is a TSG-6 link module, a variant thereof or a sufficient portion thereof that specifically binds to HA.

69. The method of claim 67 or claim 68, wherein the multimerization domain is selected from among an immunoglobulin constant region (Fc), a leucine zipper, complementary hydrophobic regions, complementary hydrophilic regions, compatible protein-protein interaction domains, free thiols that forms an intermolecular disulfide bond between two molecules, and a protuberance-into-cavity and a compensatory cavity of identical or similar size that form stable multimers.

70. The method of any of claims 67-69, wherein the multimerization domain is an Fc domain or a variant thereof that effects multimerization.

71. The method of any of claims 67-70, wherein the HABP is a fusion protein that contains a TSG-6 link module and an immunoglobulin Fc domain.

72. The method of any of claims 1-71, wherein the HABP or TSG-6 is conjugated to a detectable moiety that is detectably labeled or that can be detected.

73. The method of claim 72, wherein the HABP or TSG-6 is biotinylated.

sample.

75. The method of any of claims 1-74, wherein the sample is a stromal tissue sample from a tumor.

76. The method of any of claims 1-75, wherein the tissue is fixed, paraffin-embedded, fresh, or frozen.

77. The method of any of claims 1-76, wherein the sample is from a biopsy from a solid tumor.

78. The method of any of claims 1-77, wherein the sample is obtained by needle biopsy, CT-guided needle biopsy, aspiration biopsy, endoscopic biopsy, bronchoscopic biopsy, bronchial lavage, incisional biopsy, excisional biopsy, punch biopsy, shave biopsy, skin biopsy, bone marrow biopsy, and the Loop Electrosurgical Excision Procedure (LEEP).

79. The method of any of claims 1-73, wherein the sample is a fluid sample that is a blood, serum, urine, sweat, semen, saliva, cerebral spinal fluid, or lymph sample.

80. The method of any of claims 1-79, wherein the sample is obtained from a mammal.

81. The method of claim 80, wherein the mammal is a human.

82. The method of any of claims 1-81, wherein the tumor is of a cancer selected from among breast cancer, pancreatic cancer, ovarian cancer, colon cancer, lung cancer, non-small cell lung cancer, in situ carcinoma (ISC), squamous cell carcinoma (SCC), thyroid cancer, cervical cancer, uterine cancer, prostate cancer, testicular cancer, brain cancer, bladder cancer, stomach cancer, hepatoma, melanoma, glioma, retinoblastoma, mesothelioma, myeloma, lymphoma, and leukemia.

83. The method of any of claims 1-82, wherein the tumor is of a cancer that is a late-stage cancer, a metastatic cancer and an undifferentiated cancer.

84. The method of any of claims 1-83, wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme that is a hyaluronidase.

85. The method of claim 84, wherein the hyaluronan-degrading enzyme is a PH20 hyaluronidase or truncated form thereof lacking a C-terminal glycosylphosphatidylinositol (GPI) attachment site or a portion of the GPI attachment site.

86. The method of claim 84 or claim 85, wherein the hyaluronidase is a PH20 selected from a human, monkey, bovine, ovine, rat, mouse or guinea pig PH20.

87. The method of any of claims 84-86, wherein the hyaluronan-degrading enzyme is a human PH20 hyaluronidase that is neutral active and N-glycosylated and is selected from among:

(a) a hyaluronidase polypeptide that is a full-length PH20 or is a C-terminal truncated form of the PH20, wherein the truncated form includes at least amino acid residues 36-464 of SEQ ID NO:1, wherein the full-length PH20 comprises the sequence of amino acids set forth in SEQ ID NO:2; or

(b) a hyaluronidase polypeptide comprising a sequence of amino acids having at least 85 %, 86 %, 87 %, 88 %, 89 %, 90 %, 91 %, 92 %, 93 %, 94 %, 95 %, 96 %, 97 %, 98 %, 99 % or more sequence identity with the polypeptide or truncated form of sequence of amino acids set forth in SEQ ID NO:2; or

substitutions, whereby the hyaluronidase polypeptide has a sequence of amino acids having at least 85 %, 86 %, 87 %, 88 %, 89 %, 90 %, 91 %, 92 %, 93 %, 94 %, 95 %, 96 %, 97 %, 98 %, 99 % or more sequence identity with the polypeptide set forth in SEQ ID NO:2 or the with the corresponding truncated forms thereof.

88. The method of any of claims 84-87, wherein the PH20 or truncated form thereof comprises the sequence of amino acids set forth in any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189 or a sequence of amino acids that exhibits at least 85% sequence identity to any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189.

89. The method of any of claims 1-88, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme that is modified by conjugation to a polymer.

90. The method of claim 89, wherein the polymer is PEG and the hyaluronan degrading enzyme is PEGylated.

91. The method of claim 90, wherein the hyaluronan-degrading enzyme is a PEGylated PH20 enzyme (PEGPH20).

92. The method of any of claims 1-91, wherein hyaluronan is detected by a solid phase binding assay.

93. The method of any of claims 1-91, wherein hyaluronan is detected by histochemistry.

94. The method of any of claims 1-91, wherein hyaluronan is detected via *in vivo* imaging.

95. The method of claim 94, wherein the *in vivo* imaging method is selected from among magnetic resonance imaging (MRI), single-photon emission computed tomography (SPECT), computed tomography (CT), computed axial tomography (CAT), electron beam computed tomography (EBCT), high resolution computed tomography (HRCT), hypocycloidal tomography, positron emission tomography (PET), scintigraphy, gamma camera, a β+ detector, a γ detector, fluorescence imaging, low-light imaging, X-rays, and bioluminescence imaging.

96. The method of claim 94 or claim 95, wherein the HABP or TSG-6 is conjugated to a moiety that provides a signal or induces a signal that is detectable *in vivo*.

97. The method of claim 96, wherein the moiety is a radioisotope, bioluminescent compound, chemiluminescent compound, fluorescent compound, metal chelate or enzyme.

98. A TSG-6-LM multimer, comprising:

a first polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a first multimerization domain; and

a second polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a second multimerization domain, wherein:

the first and second multimerization domains interact to form a multimer containing two or more TSG-6 link modules;

the first and second polypeptide do not comprise the full-length sequence of TSG-6; and

association constant of at least 10^7 M^{-1}.

99. The TSG-6-LM multimer of claim 98, wherein the full-length sequence of TSG-6 is set forth in SEQ ID NO:206 or SEQ ID NO:222.

100. The TSG-6-LM multimer of claim 98 or claim 99, wherein the link module is the only TSG-6 portion of the first polypeptide and the second polypeptide.

101. The TSG-6 multimer of any of claims 98-100, wherein the first and second link module are the same or different.

102. The TSG-6 multimer of any of claims 98-101, wherein the link module comprises the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 that specifically binds HA.

103. The TSG-6 multimer of claim 102, wherein the link module exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 that specifically binds HA.

104. The TSG-6 multimer of any of claims 98-103, wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

105. The TSG-6 multimer of claim 104, wherein binding to heparin is reduced at least 1.2-fold, 1.5-fold, 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, 10-fold, 20-fold, 30-fold, 40-fold, 50-fold, 100-fold or more.

106. The TSG-6 multimer of claim 104 or claim 105, wherein the TSG-6 link module comprises an amino acid replacement at an amino acid position corresponding to amino acid residue 20, 34, 41, 54, 56, 72 or 84 set forth in SEQ ID NO:360, whereby a corresponding amino acid residue is identified by alignment to a TSG-6-LM set forth in SEQ ID NO:360.

107. The TSG-6 multimer of claim 106, wherein the amino acid replacement is to a non-basic amino acid residue selected from among Asp (D), Glu (E), Ser (S), Thr (T), Asn (N), Gln (Q), Ala (A), Val (V), Ile (I), Leu (L), Met (M), Phe (F), Tyr (Y) and Trp (W).

108. The TSG-6 multimer of any of claims 104-107, wherein the TSG-6 link module comprises an amino acid replacement corresponding to amino acid replacement K20A, K34A or K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

109. The TSG-6 multimer of any of claims 104-108, wherein the TSG-6 link module comprises amino acid replacements corresponding to amino acid replacements K20A, K34A and K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

110. The TSG-6 multimer of any of claims 104-109, wherein the link module comprises the sequence of amino acids set forth in SEQ ID NO:361 or 416 or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 361 or 416 that specifically binds HA.

111. The TSG-6 multimer of claim 110, wherein the link module exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO:361 or 416 and specifically binds HA.

112. The TSG-6 multimer of any of claims 98-111, wherein the multimerization domain is selected from among an immunoglobulin constant region (Fc), a leucine zipper, complementary hydrophobic regions, complementary hydrophilic regions, compatible protein-protein interaction domains, free thiols that forms an intermolecular disulfide bond between two molecules, and a protuberance-into-cavity and a compensatory cavity of identical or similar size that form stable multimers.

113. The TSG-6 multimer of any of claims 98-112, wherein the multimerization domain is an Fc domain or a variant thereof that effects multimerization.

114. The TSG-6 multimer of claim 113, wherein the Fc domain is from an IgG, IgM or an IgE.

115. The TSG-6 multimer of claim 113 or claim 114, wherein the Fc domain has the sequence of amino acids set forth in SEQ ID NO:359.

116. The TSG-6 multimer of any of claims 98-115, wherein the first and second polypeptide comprise a TSG-6 link module and an immunoglobulin Fc domain.

117. The TSG-6 multimer of any of claims 98-116, comprising the sequence of amino acids set forth in SEQ ID NO: 212 or 215 or a sequence of amino acids that exhibits at least 65% amino acid sequence identity to SEQ ID NO:212 or 215.

118. The TSG-6 multimer of claim 117, wherein the TSG-6 multimer comprises a sequence of amino acids that exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO:212 or 215 and that specifically binds HA.

119. The TSG-6 multimer of any of claims 98-118 that has a binding affinity to HA with an association constant (Ka) of at least $1 \times 10^8\ M^{-1}$, $2 \times 10^8\ M^{-1}$, $3 \times 10^8\ M^{-1}$, $4 \times 10^8\ M^{-1}$, $5 \times 10^8\ M^{-1}$, $6 \times 10^8\ M^{-1}$, $7 \times 10^8\ M^{-1}$, $8 \times 10^8\ M^{-1}$, $9 \times 10^8\ M^{-1}$, $1 \times 10^9\ M^{-1}$ or higher.

120. A kit or combination, comprising:

the TSG-6 multimer of any of claims 98-119 ; and

an anti-hyaluronan agent.

121. A kit or combination, comprising:

a hyaluronan binding agent (HABP) for detecting the amount of hyaluronan in a sample, wherein:

the HABP has not been prepared from animal cartilage; and

the HABP comprises a link module selected from among CD44, LYVE-1, HAPLN1/link protein, HAPLN2, HAPLN3, HAPLN4, aggrecan, versican, neurocan, brevican, phosphacan, TSG-6, Stabilin-1, Stabilin-2, CAB61358 and KIA0527 or a portion thereof comprising the link module or a sufficient portion of the link module to bind HA; and

an anti-hyaluronan agent.

122. The kit or combination of claim 121 , wherein the link module or modules is the only HABP portion of the molecule.

123. The kit or combination of claim 121 or claim 122, wherein the HABP comprises a TSG-6 link module (LM), a variant thereof or a sufficient portion thereof that binds HA.

124. The kit or combination of claim 123 , wherein the HABP comprises a link module set forth in SEQ ID NO: 207, 360, 417 or 418, or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 and specifically binds HA.

125. The kit or combination of claim 124 , wherein the HABP comprises a link module that exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418, whereby the HABP binds HA.

126. The kit or combination of any of claims 123-125 , wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

127. The kit or combination of claim 126 , wherein binding to heparin is reduced at least 1.2-fold, 1.5-fold, 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, 10-fold, 20-fold, 30-fold, 40-fold, 50-fold, 100-fold or more.

128. The kit or combination of claim 126 or claim 127, wherein the TSG-6 link module comprises an amino acid replacement at an amino acid position corresponding to amino acid residue 20, 34, 41, 54, 56, 72 or 84 set forth in SEQ ID NO:360, whereby a corresponding amino acid residue is identified by alignment to a TSG-6-LM set forth in SEQ ID NO:360.

129. The kit or combination of claim 128 , wherein the amino acid replacement is to a non-basic amino acid residue selected from among Asp (D), Glu (E), Ser (S), Thr (T), Asn (N), Gln (Q), Ala (A), Val (V), Ile (I), Leu (L), Met (M), Phe (F), Tyr (Y) and Trp (W).

130. The kit or combination of any of claims 126-129 , wherein TSG-6 link module comprises an amino acid replacement corresponding to amino acid replacement K20A, K34A or K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

131. The kit or combination of any of claims 126-130 , wherein TSG-6 link module comprises amino acid replacements corresponding to amino acid replacements K20A, K34A and K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

132. The kit or combination of any of claims 126-131 , wherein the HABP comprises a link module set forth in SEQ ID NO:361 or 416 or a sequence of amino acids comprising at least 65% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO:361 or 416 that specifically binds HA.

133. The kit or combination of claim 132 , wherein the HABP comprises a link module that exhibits at least 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the sequence of amino acids set forth in SEQ ID NO:361 or 416, whereby the HABP specifically binds HA.

134. The kit or combination of any of claims 123-133 , wherein the link module is the only TSG-6 portion of the HABP.

135. The kit or combination of any of claim 121 , wherein the HABP comprises a G1 domain of a type C hyaluronan binding protein.

136. The kit or combination of claim 135 , wherein the HABP comprises a G1 domain selected from among Aggrecan G1, Versican G1, Neurocan G1 and Brevican G1.

137. The kit or combination of claim 135 or claim 136 , wherein the G1 domain is the only HABP portion of the molecule.

138. The kit or combination of any of claims 121-137 , wherein the HABP is a multimer comprising a first HA-binding domain linked directly or indirectly via a linker to a multimerization domain and a second HA-binding domain linked directly or indirectly via a linker to a multimerization domain.

139. The kit or combination of claim 138, wherein the HA-binding domain is a link module or a G1 domain.

140. The kit or combination of claim 138 or claim 139 , wherein the first and second HA-binding domain is the same or different.

141. The kit or combination of any of claims 138-140 , wherein the multimerization domain is selected from among an immunoglobulin constant region (Fc), a leucine zipper, complementary hydrophobic regions, complementary hydrophilic regions, compatible protein-protein interaction domains, free thiols that forms an intermolecular disulfide bond between two molecules, and a protuberance-into-cavity and a compensatory cavity of identical or similar size that form stable multimers.

142. The kit or combination of any of claims 138-141 , wherein the multimerization domain is an Fc domain or a variant thereof that effects multimerization.

143. The kit or combination of any of claims 120-142 , wherein the HABP has a binding affinity to HA of at least 10^7 M^{-1}.

144. The kit or combination of any of claims 120-143 , wherein the HABP has a binding affinity to HA with an association constant (Ka) of at least $1 \times 10^8 M^{-1}$, $2 \times 10^8 M^{-1}$, $3 \times 10^8 M^{-1}$, $4 \times 10^8 M^{-1}$, $5 \times 10^8 M^{-1}$, 6×10^8 M^{-1}, $7 \times 10^8 M^{-1}$, $8 \times 10^8 M^{-1}$, $9 \times 10^8 M^{-1}$, 1×10^9 M^{-1} or higher.

145. The kit or combination of any of claims 120-144 , wherein the HABP or TSG-6 multimer is conjugated to a detectable moiety that is detectably labeled or that can be detected.

146. The kit or combination of claim 145 , wherein the HABP or TSG-6 multimer is biotinylated.

147. The kit or combination of any of claims 120-146 , further comprising reagents for detection of the HABP or TSG-6 multimer.

148. The kit or combination of any of claims 120-147 , wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

149. The kit or combination of claim 148 , wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme.

150. The kit or combination of claim 149 , wherein the hyaluronan degrading enzyme is a hyaluronidase.

151. The kit or combination of claim 150 , wherein the hyaluronan-degrading enzyme is a PH20 hyaluronidase or truncated form thereof to lacking a C-terminal glycosylphosphatidylinositol (GPI) attachment site or a portion of the GPI attachment site.

selected from a human, monkey, bovine, ovine, rat, mouse or guinea pig PH20.

153. The kit or combination of any of claims 150-152 , wherein he hyaluronan-degrading enzyme is a human PH20 hyaluronidase that is neutral active and N-glycosylated and is selected from among:

(a) a hyaluronidase polypeptide that is a full-length PH20 or is a C-terminal truncated form of the PH20, wherein the truncated form includes at least amino acid residues 36-464 of SEQ ID NO:1, wherein the full-length PH20 comprises the sequence of amino acids set forth in SEQ ID NO:2; or

(b) a hyaluronidase polypeptide comprising a sequence of amino acids having at least 85 %, 86 %, 87 %, 88 %, 89 %, 90 %, 91 %, 92 %, 93 %, 94 %, 95 %, 96 %, 97 %, 98 %, 99 % or more sequence identity with the polypeptide or truncated form of sequence of amino acids set forth in SEQ ID NO:2; or

(c) a hyaluronidase polypeptide of (a) or (b) comprising amino acid substitutions, whereby the hyaluronidase polypeptide has a sequence of amino acids having at least 85 %, 86 %, 87 %, 88 %, 89 %, 90 %, 91 %, 92 %, 93 %, 94 %, 95 %, 96 %, 97 %, 98 %, 99 % or more sequence identity with the polypeptide set forth in SEQ ID NO:2 or the with the corresponding truncated forms thereof.

154. The kit or combination of any of claims 150-153 , wherein the PH20 or truncated form thereof comprises the sequence of amino acids set forth in any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189 or a sequence of amino acids that exhibits at least 85% sequence identity to any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189.

155. The kit or combination of any of claims 120-154 , wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme that is modified by conjugation to a polymer.

156. The kit or combination of claim 155 , wherein the polymer is PEG and the hyaluronan degrading enzyme is PEGylated.

157. The kit or combination of any of claims 120-156 , further comprising a corticosteroid.

158. The kit or combination of any of claims 120-157 , further comprising a label or package insert for use of its components.

159. Use of a hyaluronan binding protein (HABP) for manufacture of a medicament for selecting a subject for treatment of a tumor with anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

160. A pharmaceutical composition comprising a hyaluronan binding protein (HABP) for use in selecting a subject for treatment of a tumor with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

161. Use of a hyaluronan binding protein (HABP) for formulation of a medicament for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP has not been prepared or isolated from animal cartilage.

162. A pharmaceutical composition comprising a hyaluronan binding protein (HABP) for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

163. Use of an anti-hyaluronan agent for manufacture of a medicament for treatment of a hyaluronan-associated disease or condition in a subject selected for treatment based on the level or amount of hyaluronan (HA) in a sample from the subject as detected with a hyaluronan binding protein (HABP), wherein:

the subject selected for treatment exhibits a level or amount of hyaluronan (HA) in the sample that is at or above a predetermined level; and

the HABP has not been prepared or isolated from animal cartilage.

164. A pharmaceutical composition comprising an anti-hyaluronan agent for use in treatment of a hyaluronan-associated disease or condition in a subject selected for treatment based on the level or amount of hyaluronan (HA) in a sample from the subject as detected with a hyaluronan binding protein (HABP), wherein:

the subject selected for treatment exhibits a level or amount of hyaluronan (HA) in the sample that is at or above a predetermined level; and

the HABP has not been prepared or isolated from animal cartilage.

165. The use of claim 163 or pharmaceutical composition of claim 164 , wherein the subject is a subject previously selected for treatment based on the level or amount of hyaluronan in a sample from the subject.

166. The use or pharmaceutical composition of any of claims 163-165 , wherein the sample from the subject contains high HA.

167. The use or pharmaceutical composition of any of claims 163-166 , wherein the predetermined level or amount of HA in the sample is at least or above 0.025 μg HA/ml of sample, 0.030 μg/ml, 0.035 μg/ml, 0.040 μg/ml, 0.045 μg/ml, 0.050 μg/ml, 0.055 μg/ml, 0.060 μg/ml, 0.065 μg/ml, 0.070 μg/ml, 0.08 μg/ml, 0.09 μg/ml. 0.1 μg/ml, 0.2 μg/ml, 0.3 μg/ml or higher.

168. The use or pharmaceutical composition of any of claims 163-167 , wherein the sample is a tumor tissue sample, and detection is effected by histochemistry.

169. The use or pharmaceutical composition of claim 168 , wherein the predetermined level is an HA score of at least +2 (HA^{+2}) or at least +3 (HA^{+3}); or

the predetermined level is at least a percent HA positive pixels in tumor (cells and stroma) to total stain in tumor tissue of at least 10%, 10% to 25% or greater than 25%.

170. The use or pharmaceutical composition of claim 168 or claim 169 , wherein the predetermined threshold level is an HA score of at least +3 (HA^{+3}) (high levels).

171. The use or pharmaceutical composition of any of claims 163-170 , wherein the hyaluronan-associated disease or condition is a tumor or a cancer.

HABP comprises a link module or modules or a G1 domain.

173. The use or pharmaceutical composition of any of claims 159-172 , wherein the HABP comprises a TSG-6 link module (LM), a variant thereof or a sufficient portion thereof that binds HA.

174. The use or pharmaceutical composition of claim 173 , wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

175. The use or pharmaceutical composition of claim 173 or claim 174 , comprising a first TSG-6 link module linked directly or indirectly via a linker to a multimerization domain; and a second TSG-6 link module linked directly or indirectly via a linker to a multimerization domain, wherein the first and second polypeptide do not comprise the full-length sequence of TSG-6.

176. The use or pharmaceutical composition of any of claims 159-175 , wherein the HABP comprises a sequence of amino acids set forth in any of SEQ ID NOS:207, 212, 215, 222, 360, 361, 371-394 and 416-418, and 423-426 or a sequence of amino acids that exhibits at least 65%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 95%, 95%, 96%, 97%, 98%, 99% or more sequence identity to a sequence of amino acids set forth in any of SEQ ID NOS: 207, 212, 215, 222, 360, 361, 371-394 and 416-418, and 423-426 and specifically binds HA, or an HA-binding domain thereof or a sufficient portion thereof to specifically bind to HA.

177. The use or pharmaceutical composition of any of claims 159-176 , wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

178. The use of pharmaceutical composition of claim 177, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme.

CLAIMS:

1. A TSG-6-LM multimer, comprising:

a first polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a first multimerization domain; and

a second polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a second multimerization domain, wherein:

the first and second multimerization domains interact to form a multimer containing two or more TSG-6 link modules;

the first and second polypeptide do not comprise the full-length sequence of TSG-6; and

the TSG-6 multimer has a binding affinity to hyaluronan (HA) with an association constant of at least 10^7 M^{-1}.

2. The TSG-6-LM multimer of claim 1, wherein the link module is the only TSG-6 portion of the first polypeptide and the second polypeptide.

3. The TSG-6 multimer of claim 1 or claim 2, wherein the first and second link module are the same or different.

4. The TSG-6 multimer of any of claims 1-3, wherein the link module comprises the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 or a sequence of amino acids comprising at least 85% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 207, 360, 417 or 418 that specifically binds HA.

5. The TSG-6 multimer of any of claims 1-4, wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

6. The TSG-6 multimer of claim 5, wherein binding to heparin is reduced at least 1.2-fold, 1.5-fold, 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, 10-fold, 20-fold, 30-fold, 40-fold, 50-fold, 100-fold or more.

7. The TSG-6 multimer of claim 5 or claim 6, wherein the TSG-6 link module comprises an amino acid replacement at an amino acid position corresponding to amino acid residue 20, 34, 41, 54, 56, 72 or 84 set forth in SEQ ID NO:360, whereby a corresponding amino acid residue is identified by alignment to a TSG-6-LM set forth in SEQ ID NO:360.

8. The TSG-6 multimer of claim 7, wherein the amino acid replacement is to a non-basic amino acid residue selected from among Asp (D), Glu (E), Ser (S), Thr (T), Asn (N), Gln (Q), Ala (A), Val (V), Ile (I), Leu (L), Met (M), Phe (F), Tyr (Y) and Trp (W).

9. The TSG-6 multimer of any of claims 5-8, wherein the TSG-6 link module comprises an amino acid replacement corresponding to amino acid replacement K20A, K34A or K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

10. The TSG-6 multimer of any of claims 5-9, wherein the TSG-6 link module comprises amino acid replacements corresponding to amino acid replacements K20A, K34A and K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

11. The TSG-6 multimer of any of claims 5-10, wherein the link module comprises the sequence of amino acids set forth in SEQ ID NO:361 or 416 or a sequence of amino acids comprising at least 85% amino acid sequence identity to the sequence of amino acids set forth in SEQ ID NO: 361 or 416 that specifically binds HA.

12. The TSG-6 multimer of any of claims 1-11, wherein the multimerization domain is selected from among an immunoglobulin constant region (Fc), a leucine zipper, complementary hydrophobic regions, complementary hydrophilic regions, compatible protein-protein interaction domains, free thiols that forms an intermolecular disulfide bond between two molecules, and a protuberance-into-cavity and a compensatory cavity of identical or similar size that form stable multimers.

13. The TSG-6 multimer of any of claims 1-12, wherein the multimerization domain is an Fc domain or a variant thereof that effects multimerization.

14. The TSG-6 multimer of any of claims 1-13, wherein the first and second polypeptide comprise a TSG-6 link module and an immunoglobulin Fc domain.

15. The TSG-6 multimer of any of claims 1-14, comprising the sequence of amino acids set forth as amino acids 24-349 of SEQ ID NO: 212 or 215 or a sequence of amino acids that exhibits at least 85% amino acid sequence identity to amino acids 24-349 of SEQ ID NO:212 or 215.

16. The TSG-6 multimer of any of claims 1-14, wherein the TSG-6 multimer comprises a sequence of amino acids encoded by the sequence of nucleotides set forth in any of SEQ ID NOS: 211, 214 or 217, or a sequence of nucleotides that exhibits at least 85% sequence identity to the sequence of nucleotides set forth in any of SEQ ID NOS: 211, 214 or 217.

17. The TSG-6 multimer of any of claims 1-16 that has a binding affinity to HA with an association constant (Ka) of at least 1 x 10^8 M^{-1}.

18. A nucleic acid molecule encoding the TSG-6 multimer of any of claims 1-17, wherein:

the nucleic acid molecule encodes a fusion polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a multimerization domain; and

the multimerization domain is a polypeptide that interacts with itself to form a stable protein-protein interaction, whereby the encoded protein forms a multimer containing at least two TSG-6 link modules.

19. The nucleic acid molecule of claim 18, comprising the sequence of nucleotides set forth in any of SEQ ID NO:211, 214 or 217, or a sequence of nucleotides that exhibits at least 85% sequence identity to the sequence of nucleotides set forth in any of SEQ ID NO: 211, 214 or 217.

20. A vector, comprising the nucleic acid molecule of claim 18 or 19.

21. An isolated cell or cell culture, comprising the nucleic acid molecule of claim 18 or 19.

22. A method of producing a TSG-6 multimer, comprising:

introducing the nucleic acid molecule of claim 18 or claim 19 into a cell;

culturing the cell under conditions whereby the fusion polypeptide is expressed by the cell; and

recovering the TSG-6 multimer.

23. A kit or combination, comprising:

the TSG-6 multimer of any of claims 1-17; and

an anti-hyaluronan agent.

24. The kit or combination of claim 23, further comprising reagents for detection of the HABP or TSG-6 multimer.

25. The kit or combination of claim 23 or claim 24, wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

26. The kit or combination of claim 25, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme.

27. The kit or combination of claim 26, wherein the hyaluronan degrading enzyme is a hyaluronidase.

28. The kit or combination of claim 27, wherein the hyaluronan-degrading enzyme is a PH20 hyaluronidase or truncated form thereof to lacking a C-terminal glycosylphosphatidylinositol (GPI) attachment site or a portion of the GPI attachment site.

29. The kit or combination of claim 28, wherein the PH20 or truncated form thereof comprises the sequence of amino acids set forth in any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189 or a sequence of amino acids that exhibits at least 85% sequence identity to any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189.

30. The kit or combination of any of claims 23-29, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme that is modified by conjugation to a polymer.

31. The kit or combination of claim 30, wherein the polymer is PEG and the hyaluronan degrading enzyme is PEGylated.

32. A method for selecting a subject for treatment of a tumor with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample previously obtained from a subject who has a tumor or cancer with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage; and

detecting binding of the hyaluronan binding protein to the sample, thereby determining the amount of hyaluronan in the sample, wherein if the amount of hyaluronan in the sample is at or above a predetermined threshold level, selecting the subject for treatment with an anti-hyaluronan agent.

33. The method of any of claim 32, wherein the predetermined threshold level is high HA.

34. The method of claim 32 or claim 33, wherein:

the predetermined threshold level is at least or above 0.025 μg HA/ml of sample, 0.030 μg/ml, 0.035 μg/ml, 0.040 μg/ml, 0.045 μg/ml, 0.050 μg/ml, 0.055 μg/ml, 0.060 μg/ml, 0.065 μg/ml, 0.070 μg/ml, 0.08 μg/ml, 0.09 μg/ml. 0.1 μg/ml, 0.2 μg/ml, 0.3 μg/ml or higher; or

the sample is a tumor tissue sample, and the predetermined threshold is an HA score of at least +2 (HA^{+2}) or at least +3 (HA^{+3}); or

the sample is a tumor tissue sample, and the predetermined threshold level is at least a percent HA positive pixels in tumor (cells and stroma) to total stain in tumor tissue of at least 10%, 10% to 25% or greater than 25%.

35. The method of claim 34, wherein the predetermined threshold level is an HA score of at least +3 (HA^{+3}) (high levels).

36. A method for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, comprising:

contacting a tissue or body fluid sample previously obtained from a subject who is or has been treated with an anti-hyaluronan agent with a hyaluronan binding protein (HABP) molecule, wherein the HABP has not been prepared from or isolated from animal cartilage;

detecting binding of the hyaluronan binding protein to the sample, thereby determining the amount of hyaluronan in the sample, wherein detection of a decrease in hyaluronan compared to before

treatment with anti-hyaluronan agent or the last dose of anti-hyaluronan agent indicates that the treatment is effective.

37 The method of claim 36, wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

38. The method of claim 37, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme.

39. The method of claim 38, wherein the hyaluronan degrading enzyme is a hyaluronidase.

40. The method of claim 39, wherein the hyaluronan-degrading enzyme is a PH20 hyaluronidase or truncated form thereof lacking a C-terminal glycosylphosphatidylinositol (GPI) attachment site or a portion of the GPI attachment site.

41. The method of claim 40, wherein the PH20 or truncated form thereof comprises the sequence of amino acids set forth in any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189 or a sequence of amino acids that exhibits at least 85% sequence identity to any of SEQ ID NOS: 4-9, 47, 48, 150-170 and 183-189.

42. The method of any of claims 37-41, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme that is modified by conjugation to a polymer.

43. The method of claim 42, wherein the polymer is PEG and the hyaluronan degrading enzyme is PEGylated.

44. The method of any of claims 36-43, wherein the subject has a tumor or cancer.

45. The method of any of claims 32-44, wherein the HABP comprises a link module.

46. The method of claim 45, wherein the link module or modules is the only HABP portion of the molecule.

47. The method of any of claims 32-46, wherein the HABP comprises a link module selected from among CD44, LYVE-1, HAPLN1/link protein, HAPLN2, HAPLN3, HAPLN4, aggrecan, versican, neurocan, brevican, phosphacan, TSG-6, Stabilin-1, Stabilin-2, CAB61358 and KIA0527 or a portion thereof comprising the link module or a sufficient portion of a link module to bind HA.

48. A method of diagnosis of a hyaluronan-associated disease or condition, comprising:

contacting a tissue or body fluid sample previously obtained from a subject with TSG-6 or a hyaluronan binding region thereof; and

detecting binding of the TSG-6 or a hyaluronan-binding region thereof that binds to the sample, thereby determining the amount of hyaluronan in the sample, wherein the subject is diagnosed with a hyaluronan-associated disease or condition if the amount of hyaluronan is above a predetermined level or above the hyaluronan level of a reference sample.

49. The method of claim 48, wherein the hyaluronan-associated disease or condition is a tumor or cancer.

50. The method of claim 48 or claim 49, wherein the predetermined or reference level is the median level of hyaluronan present in healthy or normal tissue or fluid samples from a population of control subjects.

51. The method of any of claims 48-50, wherein:

the predetermined threshold level is greater than 0.015 μg HA/ml of sample; or

the sample is a tumor or tissue sample, and the predetermined threshold level is an HA score of HA^{+1}, HA^{+2} or HA^{+3}.

52. The method of any of claims 32-51, wherein the HABP or TSG-6 comprises a TSG-6 link module (LM) or a sufficient portion thereof that specifically binds HA.

53. The method of any of claims 52, wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

54. The method of claim 53, wherein binding to heparin is reduced at least 1.2-fold, 1.5-fold, 2-fold, 3-fold, 4-fold, 5-fold, 6-fold, 7-fold, 8-fold, 9-fold, 10-fold, 20-fold, 30-fold, 40-fold, 50-fold, 100-fold or more.

55. The method of claim 53 or claim 54, wherein TSG-6 link module comprises an amino acid replacement selected from among an amino acid replacement corresponding to amino acid replacement K20A, K34A and K41A in a TSG-6-LM set forth in SEQ ID NO:360 or the replacement at the corresponding residue in another TSG-6-LM.

56. The method of any of claims 32-55, wherein the HABP or TSG-6 is a multimer comprising a first HA-binding domain linked directly or indirectly via a linker to a multimerization domain and a second HA-binding domain linked directly or indirectly via a linker to a multimerization domain, wherein the first and second multimerization domains interact to form a multimer containing two or more HA-binding domains.

57. The method of claim 56, wherein the first and second HA-binding domain is a TSG-6 link module, a variant thereof or a sufficient portion thereof that specifically binds to HA.

58. The method of claim 56 or claim 57, wherein the multimerization domain is selected from among an immunoglobulin constant region (Fc), a leucine zipper, complementary hydrophobic regions, complementary hydrophilic regions, compatible protein-protein interaction domains, free thiols that forms an intermolecular disulfide bond between two molecules, and a protuberance-into-cavity and a compensatory cavity of identical or similar size that form stable multimers.

59. The method of any of claims 56-58, wherein the multimerization domain is an Fc domain or a variant thereof that effects multimerization.

60. The method of any of claims 56-59, wherein the HABP is a fusion protein that contains a TSG-6 link module and an immunoglobulin Fc domain.

61. The method of claim 60 , wherein the TSG-6-LM-Fc has the sequence of amino acids set forth as amino acids 24-349 of SEQ ID NO: 212 or 215 or a sequence of amino acids that exhibits at least 85% amino acid sequence identity to amino acids 24-349 of SEQ ID NO:212 or 215 and specifically binds HA.

62. The method of claim 60 or claim 61, wherein the TSG-6-LM-Fc comprises the sequence of amino acids encoded by the sequence of nucleotides set forth in any of SEQ ID NOS: 211, 214 or 217, or a sequence of nucleotides that exhibits at least 85% sequence identity to the sequence of nucleotides set forth in any of SEQ ID NOS: 211, 214 or 217.

63. The method of any of claims 32-62, wherein the HABP or TSG-6 specifically binds to HA with a binding affinity that has a dissociation constant (Kd) of at least less than or less than 1×10^{-7} M.

64. The method of any of claims 32-63 , wherein the HABP or TSG-6 is conjugated to a detectable moiety that is detectably labeled or that can be detected.

65. The method of any of claims 32-64, wherein:

the sample is a stromal tissue sample; or

the sample is a fluid sample that is a blood, serum, urine, sweat, semen, saliva, cerebral spinal fluid, or lymph sample

66. The method of any of claims 32-65, wherein the sample is a stromal tissue sample from a tumor.

67. The method of any of claims 32-66, wherein the tumor is of a cancer selected from among breast cancer, pancreatic cancer, ovarian cancer, colon cancer, lung cancer, non-small cell lung cancer, in situ carcinoma (ISC), squamous cell carcinoma (SCC), thyroid cancer, cervical cancer, uterine cancer, prostate cancer, testicular cancer, brain cancer, bladder cancer, stomach cancer, hepatoma, melanoma, glioma, retinoblastoma, mesothelioma, myeloma, lymphoma, and leukemia.

68. The method of any of claims 32-67, wherein the tumor is of a cancer that is a late-stage cancer, a metastatic cancer and an undifferentiated cancer.

69. The method of any of claims 32-68, wherein hyaluronan is detected by a solid phase binding assay by histochemistry , or by *in vivo* imaging.

70. Use of a hyaluronan binding protein (HABP) for manufacture of a composition for selecting a subject for treatment of a tumor with anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

71. A pharmaceutical composition comprising a hyaluronan binding protein (HABP) for use in selecting a subject for treatment of a tumor with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

72. Use of a hyaluronan binding protein (HABP) for manufacture of a composition for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

73. A pharmaceutical composition comprising a hyaluronan binding protein (HABP) for predicting efficacy of treatment of a subject with an anti-hyaluronan agent, wherein:

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

74. Use of an anti-hyaluronan agent for manufacture of a medicament for treatment of a hyaluronan-associated disease or condition in a subject selected for treatment based on the level or amount of hyaluronan (HA) in a sample from the subject as detected with a hyaluronan binding protein (HABP), wherein:

the subject selected for treatment exhibits a level or amount of hyaluronan (HA) in the sample that is at or above a predetermined level;

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

75. A pharmaceutical composition comprising an anti-hyaluronan agent for use in treatment of a hyaluronan-associated disease or condition in a subject selected for treatment based on the level or amount of hyaluronan (HA) in a sample from the subject as detected with a hyaluronan binding protein (HABP), wherein:

the subject selected for treatment exhibits a level or amount of hyaluronan (HA) in the sample that is at or above a predetermined level;

the HABP comprises a link module or modules or a G1 domain; and

the HABP has not been prepared or isolated from animal cartilage.

76. The use of claim 74 or pharmaceutical composition of claim 75, wherein the subject is a subject previously selected for treatment based on the level or amount of hyaluronan in a sample from the subject.

77. The use or pharmaceutical composition of any of claims 74-76, wherein the sample from the subject contains high HA.

78. The use or pharmaceutical composition of any of claims 74-77, wherein:

the predetermined level or amount of HA in the sample is at least or above 0.025 μg HA/ml of sample, 0.030 μg/ml, 0.035 μg/ml, 0.040 μg/ml, 0.045 μg/ml, 0.050 μg/ml, 0.055 μg/ml, 0.060 μg/ml, 0.065 μg/ml, 0.070 μg/ml, 0.08 μg/ml, 0.09 μg/ml. 0.1 μg/ml, 0.2 μg/ml, 0.3 μg/ml or higher; or

the sample is a tumor tissue sample, and the predetermined level is an HA score of at least +2 (HA^{+2}) or at least +3 (HA^{+3}); or

the sample is a tumor tissue sample, and the predetermined level is at least a percent HA positive pixels in tumor (cells and stroma) to total stain in tumor tissue of at least 10%, 10% to 25% or greater than 25%.

79. The use or pharmaceutical composition of claim 78, wherein the predetermined threshold level is an HA score of at least +3 (HA^{+3}) (high levels).

80. The use or pharmaceutical composition of any of claims 74-79, wherein the hyaluronan-associated disease or condition is a tumor or a cancer.

81. The use or pharmaceutical composition of any of claims 70-80, wherein the HABP comprises a TSG-6 link module (LM), a variant thereof or a sufficient portion thereof that binds HA.

82. The use or pharmaceutical composition of claim 81, wherein the TSG-6 link module is modified to reduce or eliminate binding to heparin.

83. The use or pharmaceutical composition of claim 81 or claim 82, wherein the TSG-6-LM is provided as a multimer comprising:

a first polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a multimerization domain; and

a second polypeptide containing a TSG-6 link module linked directly or indirectly via a linker to a multimerization domain, wherein:

the first and second multimerization domains interact to form a multimer containing two or more TSG-6 link modules; and

the first and second polypeptide do not comprise the full-length sequence of TSG-6.

84. The use or pharmaceutical composition of any of claims 70-83, wherein the HABP is a TSG-6-LM-Fc that comprises the sequence of amino acids set forth as amino acids 24-349 of SEQ ID NO:211 or 215, or a sequence of amino acids that exhibits at least 85%, sequence identity to the sequence of amino acids 24-349 set forth in SEQ ID NO: 212 or 215 and specifically binds HA, or an HA-binding domain thereof or a sufficient portion thereof to specifically bind to HA.

85. The use of pharmaceutical composition of any of claims 70-84 wherein the TSG-6-LM-Fc comprises the sequence of amino acids encoded by the sequence of nucleotides set forth in any of SEQ ID NOS: 211, 214 or 217, or a sequence of nucleotides that exhibits at least 85% sequence identity to the sequence of nucleotides set forth in any of SEQ ID NOS: 211, 214 or 217.

86. The use or pharmaceutical composition of any of claims 70-84, wherein the anti-hyaluronan agent is a hyaluronan degrading enzyme or is an agent that inhibits hyaluronan synthesis.

87. The use of pharmaceutical composition of claim 86, wherein the anti-hyaluronan agent is a hyaluronan-degrading enzyme.