tissues where direct drug administration is common include the posterior eye chamber via intravitreal injection and articular joints via intra-articular injection.

[0005] However, local tissue administration requires a professional to safely provide the required injection, which makes them more burdensome and costly to administer compared to systemic administration. When the peptide drug is administered as part of a peptide-polymer conjugate, it is possible to substantially reduce the frequency of drug administration, thereby reducing the burden on the patient to receive effective treatment. Furthermore, a reduction in the number of local injections reduces the risk of local tissue injury or adverse effects to the injection. Finally, the need for less frequent administrations can reduce the amount of time that the drug concentration in the target tissue is below the therapeutic concentration, thereby improving the overall efficacy of the drug. Based on these advantages, there is a strong motivation to develop protein-polymer drug products for a variety of diseases.

[0006] To appropriately formulate a peptide-polymer conjugate as a drug product, it is necessary to achieve sufficiently high drug concentrations to enable appropriate dosing in the patient. Achieving sufficient dosing requires achieving both an appropriate concentration of the peptide polymer conjugates in the drug product solution as well as the appropriate drug load of peptide conjugated to each polymer. It is also necessary to filter the peptide-polymer drug substance through a 0.22-micron filter to eliminate any bacteria or pathogens that may be contaminating the solution. Finally, the peptide-polymer drug product must exhibit shelf-stability by remaining in solution for up to two years from the date of manufacture to the date of clinical use. Interactions between the peptide-polymer conjugates can negatively impact the ability to complete any of these drug-enabling properties.

[0007] The linkers used to attach the polymer and the peptides can have a substantial impact on the pharmacological properties of the conjugates, intra-conjugate interactions, as well as conjugate-to-conjugate interactions. Therefore, there is a need to develop peptide-polymer conjugates with the specific linker chemistries that will enable them to achieve the preferred pharmacological properties for a given disease as well as to be successfully formulated into a drug product. The present invention meets this and other needs.

SUMMARY

[0008] In one aspect, the present disclosure provides a conjugate of Formula I:

$(X\text{-}Y)_n\text{-}Z$ Formula (I)

wherein each X is independently a peptide having a molecular weight of from about 5 kDa to about 200 kDa; each Y is independently a hydrophilic linker; Z is a biocompatible polymer having a molecular weight of from about 0.1 MDa to about 3 MDa; and subscript n is an integer from 10 to 1000.

[0008a] In one aspect, the present disclosure refers to a pharmaceutical composition comprising the conjugate as disclosed herein, and a pharmaceutically acceptable carrier.

[0008b] In another aspect, the present disclosure refers to the use of a conjugate as disclosed herein in the manufacture of a medicament for treating an ocular disease or disorder, wherein the medicament is to be intravitreal administered to a subject in need thereof.

[0008c] In another aspect, the present disclosure refers to the use of a conjugate as disclosed herein in the manufacture of a medicament for treating a disease or disorder in an articular joint, wherein the medicament is to be injected into the articular joint.

[0009] In another example, the present disclosure provides a pharmaceutical composition including a conjugate of the present invention and a pharmaceutically acceptable excipient.

[0010] In another example, the present disclosure provides a method of treating an ocular disease or disorder, the method comprising intravitreal administration to a subject in need thereof, a therapeutically effective amount of a conjugate of the present invention, thereby treating the ocular disease or disorder.

[0011] In another example, the present disclosure provides a method of treating a disease or disorder in an articular joint, the method comprising injecting into the articular joint an effective amount of a conjugate of the present invention, thereby treating the disease or disorder in the articular joint.

BRIEF DESCRIPTION OF THE DRAWINGS

[0012] FIG. 1 shows UV spectra of purified 830 kDa hyaluronic acid intermediates synthesized using different thiol reactive linkers. See Table 2 for conjugates.

[0013] FIG. 2 shows reverse phase HPLC analysis of HyA-(Mal PEG2 Hydrazide) intermediate product before and after purification through a Zeba 7 kDa MWCO desalting column.

[0014] FIG. 3 shows SEC trace of conjugate 40 showing the conjugate peak, followed by the smaller DARPin dimer and monomer peaks.

[0015] FIG. 4 shows percent loss after 0.22 um filtration for peptide protein conjugates 1 (EMCH), 3 (BMPH), 4 (MP2H), 16 (MP2A), 5 (MP3A), and 6 (n-AEM) linkers.

[0016] FIG. 5 shows improved log anti-VEGF potency for MVPs with a Mal-PEG2-hydrazide linker and a peptide of DARPin, E1-1 or VHH compared to MVPs with an EMCH linker.

WHAT IS CLAIMED IS:

1. A conjugate of Formula I:

$$(X\text{-}Y)_n\text{-}Z \qquad \text{Formula (I)}$$

wherein

each X is independently a peptide having a molecular weight of from about 5 kDa to about 200 kDa;

each Y is independently a hydrophilic linker;

Z is a biocompatible polymer having a molecular weight of from about 0.1 MDa to about 3 MDa; and

subscript n is an integer from 10 to 1000.

2. The conjugate of claim 1, wherein the peptide is an inhibitor of angiogenesis.

3. The conjugate of claim 2, wherein the peptide is a soluble vascular endothelial growth factor (VEGF) receptor, angiostatin, endostatin, vasostatin, an antibody specific for VEGF, or a DARPin specific for VEGF.

4. The conjugate of claim 2 or 3, wherein the peptide inhibits VEGF-A, VEGF-B, VEGF-C, VEGF-D, Ang-1, Ang-2, PDGF, or PlGF.

5. The conjugate of any one of claims 2 to 4, wherein the peptide is a monoclonal IgG antibody, an IgG antibody fragment, a single-chain variable region antibody, a single-domain heavy chain antibody, an adnectin, an affibody, an anticalin, a DARPin, a Kunitz-type inhibitor, or a receptor decoy.

6. The conjugate of claim 1, wherein the peptide modulates the activity of immune cell function.

7. The conjugate of claim 6, wherein the peptide inhibits tumor necrosis factor-α, interleukin-1β, interleukin-6, or interferon-γ.

8. The conjugate of claims 6 or 7, wherein the peptide inhibits tumor necrosis factor-α.

9. The conjugate of any one of claims 6 to 8, wherein the peptide is a monoclonal IgG antibody, an IgG antibody fragment, a single-chain variable region antibody, a single-domain heavy chain antibody, an adnectin, an affibody, an anticalin, a DARPin, a Kunitz-type inhibitor, or a receptor decoy.

10. The conjugate of any one of claims 6 to 9, wherein the peptide has a molecular weight of from about 5 kDa to about 30 kDa.

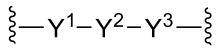
11. The conjugate of any one of claims 6 to 10, wherein the peptide has a molecular weight of from about 10 kDa to about 20 kDa.

12. The conjugate of any one of claims 6 to 11, wherein the peptide has an amino acid sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, and SEQ ID NO:10.

13. The conjugate of any one of claims 6 to 12, wherein the peptide has a sequence:

QVQLQESGGGLVQPGGSLRLSCAASGRTFSDHSGYTYTIGWFRQAPGKEREF VARIYWSSGNTYYADSVKGRFAISRDIAKNTVDLTMNNLEPEDTAVYYCAAR DGIPTSRSVESYNYWGQGTQVTVSS (SEQ ID NO:1).

14. The conjugate of any one of claims 1 to 13, wherein each hydrophilic linker independently has the formula:



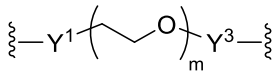
wherein

Y^1 is a thiol reactive group;

Y^2 is a $C_{3\text{-}20}$ heteroalkylene having from 1 to 6 heteroatoms each independently N, O or S, or $-(CH_2CH_2O)_m-$, wherein subscript m is an integer of from 1 to 100; and

Y^3 is a carboxy reactive group.

15. The conjugate of any one of claims 1 to 14, wherein each hydrophilic linker independently has the formula:



wherein

Y^1 is a thiol reactive group;

Y^3 is a carboxy reactive group; and

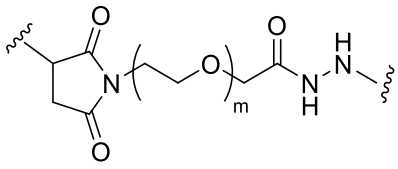
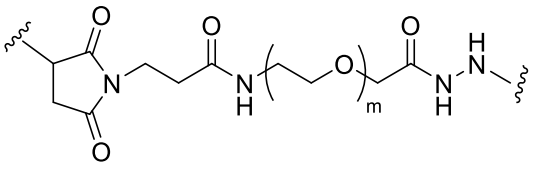
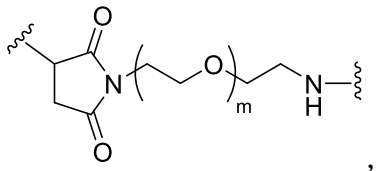
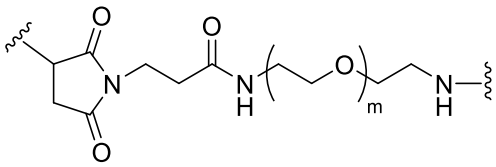
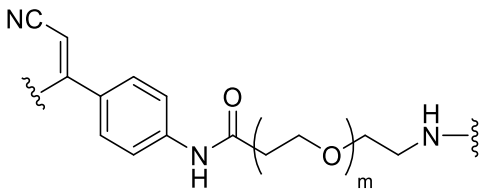
subscript m is an integer of from 1 to 100.

16. The conjugate of claim 15, wherein

Y^1 is a thiol, arylpropiolonitrile or maleimide; and

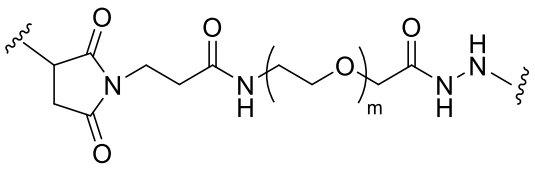
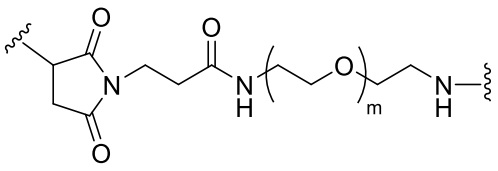
Y^3 is an amine or a N-acylhydrazide.

17. The conjugate of any one of claims 14 to 16, wherein the hydrophilic linker has the formula:


,

,

,

or


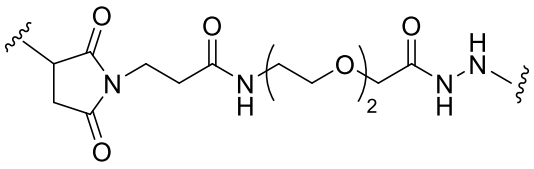
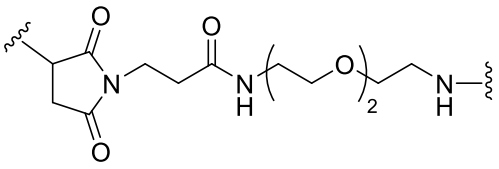
wherein subscript m is an integer of from 1 to 100.

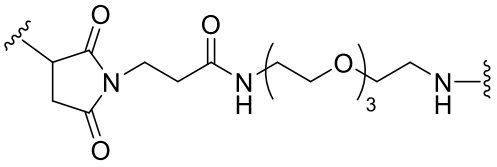
18. The conjugate of any one of claims 14 to 17, wherein the hydrophilic linker has the formula:


or


wherein subscript m is an integer of from 1 to 100.

19. The conjugate of any one of claims 14 to 18, wherein the hydrophilic linker has the formula:



or


.

20. The conjugate of any one of claims 1 to 19, wherein the biocompatible polymer is a polysaccharide.

21. The conjugate of any one of claims 1 to 20, wherein the biocompatible polymer is a glycosaminoglycan.

22. The conjugate of any one of claims 1 to 21, wherein the biocompatible polymer is hyaluronic acid.

23. The conjugate of any one of claims 1 to 22, wherein the biocompatible polymer has a molecular weight of from about 0.2 MDa to about 1.5 MDa.

24. The conjugate of any one of claims 1 to 23, wherein the biocompatible polymer has a molecular weight of about 0.9 MDa.

25. The conjugate of any one of claims 1 to 22, wherein the biocompatible polymer has a molecular weight of from about 0.8 MDa to about 3 MDa.

26. The conjugate of any one of claims 1 to 22, wherein the biocompatible polymer has a molecular weight of about 2 MDa.

27. The conjugate of any one of claims 1 to 26, wherein subscript n is an integer of from 10 to 400.

28. The conjugate of any one of claims 1 to 27, wherein subscript n is an integer of from 10 to 100.

29. The conjugate of any one of claims 1 to 28, wherein subscript n is an integer of from 50 to 100.

30. A pharmaceutical composition comprising the conjugate of any one of claims 1 to 29, and a pharmaceutically acceptable carrier.

31. Use of a conjugate of any one of claims 1 to 29 in the manufacture of a medicament for treating an ocular disease or disorder, wherein the medicament is to be intravitreal administered to a subject in need thereof.

32. The use of claim 31, wherein the ocular disorder is macular degeneration, choroidal neovascularization, retinal neovascularization, proliferative vitreoretinopathy, diabetic retinopathy, glaucoma, or ocular inflammation.

33. The use of claim 31 or 32, wherein the vitreous half-life of the conjugate is at least 2 weeks.

34. The use of any one of claims 31 to 33, wherein the subject is a human.

35. The use of any one of claims 31 to 34, wherein the conjugate is to be administered once every two months, once every three months, once every 6 months, or once a year.

36. The use of any one of claims 31 to 35, wherein the vitreous half-life of the conjugate is at least 2-fold greater than the half-life of the biologically active polypeptide not conjugated to the biocompatible polymer.

37. Use of a conjugate of any one of claims 1 to 29 in the manufacture of a medicament for treating a disease or disorder in an articular joint, wherein the medicament is to be injected into the articular joint.

38. The use of claim 37, wherein the diffusion half-life of the conjugate is at least about 2 times longer than the peptide.

39. The use of claims 37 or 38, wherein the diffusion half-life of the conjugate is from about 2 to about 100 times longer than the peptide.

40. The use of any one of claims 37 to 39, wherein the intra-articular half-life of the conjugate is at least about 20% longer than the peptide.

41. The use of any one of claims 37 to 40, wherein the intra-articular half-life of the conjugate is from about 20% to about 1000% longer than the peptide.

42. The use of any one of claims 37 to 41, wherein the disease or disorder is rheumatoid arthritis, wear-related osteoarthritis, age-related osteoarthritis, post-traumatic osteoarthritis, psoriatic arthritis, and aseptic implant loosening, joint effusion, ankylosing spondylitis, bursitis, gout, reactive arthritis, synovitis, or avascular necrosis.

43. The use of any one of claims 37 to 42, wherein the conjugate is to be injected into the articular joint no more than about once a month.

44. The use of any one of claims 37 to 43, wherein the conjugate is to be injected into the articular joint from about once a month to once every 6 months.

45. The use of any one of claims 37 to 44, wherein the conjugate is to be injected into the articular joint once every 2 months or once every 3 months.