CLAIMS

What is claimed is:

1. A composition consisting of an oxyntomodulin, a polyethylene glycol polymer (PEG polymer) and 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS), wherein said PEG polymer is attached to the amino terminus of said oxyntomodulin via Fmoc or FMS.

2. The composition of claim 1, wherein said oxyntomodulin consists of the amino acid sequence set forth in SEQ ID NO: 1.

3. The composition of any one of claims 1 to 2, wherein said PEG polymer is a PEG polymer with a sulfhydryl moiety.

4. The composition of any one of claims 1 to 3, wherein said PEG polymer is PEG_{30}.

5. A pharmaceutical composition comprising the composition of any one of claims 1-4 and a pharmaceutical acceptable carrier.

6. A composition consisting of an oxyntomodulin, a polyethylene glycol polymer (PEG polymer) and 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS), wherein said PEG polymer is attached to a lysine residue on position number twelve (Lys_{12}) of said oxyntomodulin's amino acid sequence via Fmoc or FMS.

7. The composition of claim 6, wherein said oxyntomodulin consists of the amino acid sequence set forth in SEQ ID NO: 1.

8. The composition of any one of claims 6 to 7, wherein said PEG polymer is a PEG polymer with a sulfhydryl moiety.

9. The composition of any one of claims 6 to 8, wherein said PEG polymer is PEG_{30}.

10. A pharmaceutical composition comprising the composition of any one of claims 6 to 9 and a pharmaceutical acceptable carrier.

11. A composition consisting of an oxyntomodulin, a polyethylene glycol polymer (PEG polymer) and 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS), wherein said PEG polymer is attached to lysine residue number thirteen (Lys_{30}) of said oxyntomodulin via Fmoc or FMS.

12. The composition of claim 11, wherein said oxyntomodulin consists of the amino acid sequence set forth in SEQ ID NO: 1.

13. The composition of any one of claims 11 to 12, wherein said PEG polymer is a PEG polymer with a sulfhydryl moiety.

14. The composition of any one of claims 11 to 13, wherein said PEG polymer is PEG_{30}.

15. A pharmaceutical composition comprising the composition of any one of claims 11 to 14 and a pharmaceutical acceptable carrier.

16. A method of improving the area under the curve (AUC) of oxyntomodulin, consisting of the step of conjugating a polyethylene glycol polymer (PEG polymer) to the Lysine residue on position number 12 or to the Lysine residue on position number 30 or to the amino terminus of said oxyntomodulin amino acid sequence via 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS).

17. The method of claim 16, wherein said oxyntomodulin consists of an amino acid sequence of SEQ ID NO: 1.

18. The method of any one of claims 16 to 17, wherein said PEG polymer is conjugated to the amino terminus or lysine residue of said oxyntomodulin via Fmoc or FMS.

19. The method of any one of claims 16 to 18, wherein said PEG polymer is a PEG polymer with a sulfhydryl moiety.

20. The method of any one of claims 16 to 19, wherein said PEG polymer is PEG_{30}.

21. A method of reducing the dosing frequency of oxyntomodulin, consisting of the step of conjugating a polyethylene glycol polymer (PEG polymer) to the Lysine residue on position number 12 or to the Lysine residue on position number 30 or to the amino terminus of said oxyntomodulin amino acid sequence via 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS).

22. The method of claim 21, wherein said oxyntomodulin consists of an amino acid sequence of SEQ ID NO: 1.

23. The method of any one of claims 21 to 22, wherein said PEG polymer is a PEG polymer with a sulfhydryl moiety.

24. The method of any one of claims 21 to 23, wherein said PEG polymer is PEG_{30}.

25. A method for extending the biological half-life of oxyntomodulin, consisting of the step of conjugating oxyntomodulin, a polyethylene glycol polymer (PEG polymer) and 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS), wherein said PEG polymer is conjugated to a Lysine residue on position number 12 or to a Lysine residue on position number 30 or to the amino terminus of said oxyntomodulin amino acid sequence via 9-fluorenylmethoxycarbonyl (Fmoc) or 2-sulfo-9-fluorenylmethoxycarbonyl (FMS).

26. The method of claim 25, wherein said oxyntomodulin consists of an amino acid sequence of SEQ ID NO: 1.

27. The method of any one of claims 25 to 26, wherein said PEG polymer is conjugated to the amino terminus or lysine residue of said oxyntomodulin via Fmoc or FMS.

28. The method of any one of claims 25 to 27, wherein said PEG polymer is a PEG polymer with a sulfhydryl moiety.

29. The method of any one of claims 25 to 28, wherein said PEG polymer is PEG_{30}.

30. The method of any one of claims 25 to 29, wherein said oxyntomodulin is released into a biological fluid by chemically hydrolyzing said FMS or said Fmoc linker from said composition.

31. The method of 30, wherein the released oxyntomodulin is intact and regains GLP-1 and glucagon receptor binding activity.

32. The method of any one of claims 30 to 31, wherein said biological fluid is blood, sera, or cerebrospinal fluid.

33. The method of any one of claims 30 to32, wherein said biological fluid is in a human subject.

34. The method of any one of claims 30 to 33, wherein chemically hydrolyzing said FMS or said Fmoc extends the circulating time of said OXM peptide in said biological fluid.

35. The method of claim 34, wherein extending the circulating time of said OXM allows said OXM to cross the blood brain barrier and target the CNS.

36. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for inducing glucose tolerance in a subject in need thereof.

37. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for inducing glycemic control in a subject in need thereof.

38. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for reducing food intake in a subject.

39. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for reducing body weight in a subject.

40. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for improving the cholesterol levels in a subject.

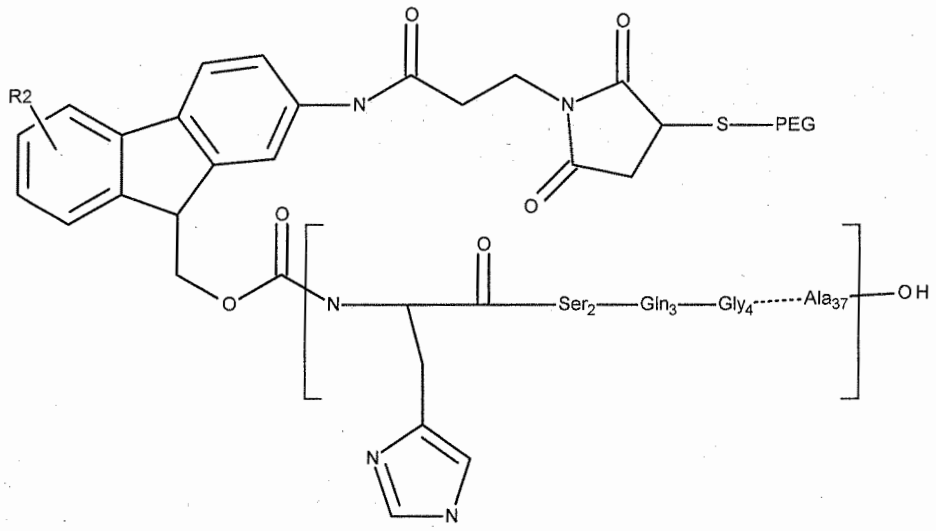
41. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for method of increasing insulin sensitivity in a subject.

42. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for reducing insulin resistance in a subject.

43. Use of the composition of any one of claims 1-5, 6-10, or 11-15 for method for increasing energy expenditure in a subject.

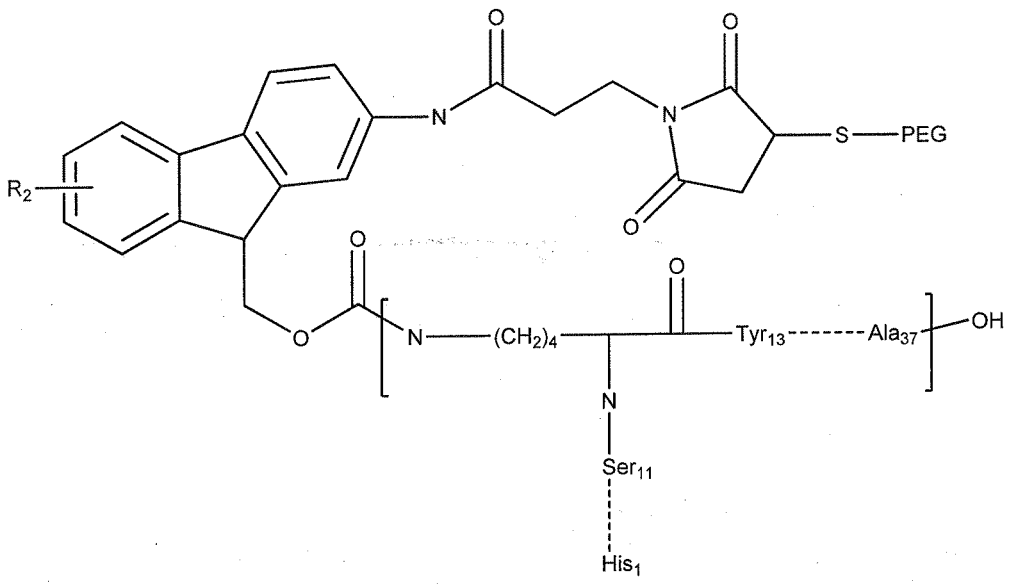
CLAIMS

1. A conjugate represented by the following structure:



oxyntomodulin

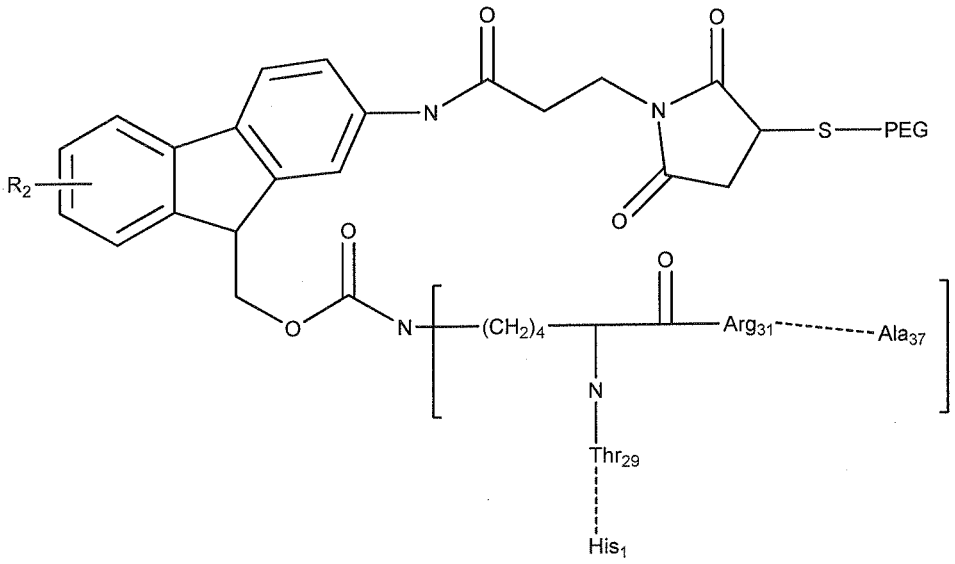
His[1] - Ser[2] - Gln[3] - Gly[4] - Thr[5]- Phe[6] - Thr[7] - Ser[8] - Asp[9] - Tyr[10] - Ser[11] - Lys[12] - Tyr[13] - Leu[14] - Asp[15] - Ser[16] - Arg[17] - Arg[18]- Ala[19] - Gln[20] - Asp[21] - Phe[22] - Val[23] - Gln[24] - Trp[25] - Leu[26] - Met[27] - Asn[28] - Thr[29] - Lys[30] - Arg[31]- Asn[32] - Arg[33] - Asn[34] - Asn[35] - Ile[36] - Ala[37] (SEQ ID NO:1)



oxyntomodulin

His [1] - Ser [2] - Gln [3] - Gly [4] - Thr [5]- Phe [6] - Thr [7] - Ser [8] - Asp [9] - Tyr [10] - Ser [11] - Lys [12] - Tyr [13] - Leu [14] - Asp [15] - Ser [16] - Arg [17] - Arg [18]- Ala [19] - Gln [20] - Asp [21] - Phe [22] - Val [23] - Gln [24] - Trp [25] - Leu [26] - Met [27] - Asn [28] - Thr [29] - Lys [30] - Arg [31]- Asn [32] - Arg [33] - Asn [34] - Asn [35] - Ile [36] - Ala [37] (SEQ ID NO: 1)

or



Oxyntomodulin

His [1] - Ser [2] - Gln [3] - Gly [4] - Thr [5]- Phe [6] - Thr [7] - Ser [8] - Asp [9] - Tyr [10] - Ser [11] - Lys [12] - Tyr [13] - Leu [14] - Asp [15]- Ser [16] - Arg [17] - Arg [18]- Ala [19] - Gln [20] - Asp [21] - Phe [22] - Val [23] - Gln [24] - Trp [25] - Leu [26] - Met [27] - Asn [28] - Thr [29]- Lys [30] - Arg [31]- Asn [32] - Arg [33] - Asn [34] - Asn [35] - Ile [36] - Ala [37] (SEQ ID NO: 1)

wherein R_2 is H or SO_3H.

2. The conjugate according to claim 1, wherein the PEG is a PEG 30.

3. The conjugate according to claim 1, wherein said R_2 is at position 2 of the fluorene ring.

4. A pharmaceutical composition comprising said conjugate of claim 1 or 2, and a pharmaceutically acceptable carrier.

5. A method for extending the biological half-life of oxyntomodulin, said method comprising administering a pharmaceutical composition of claim 4.

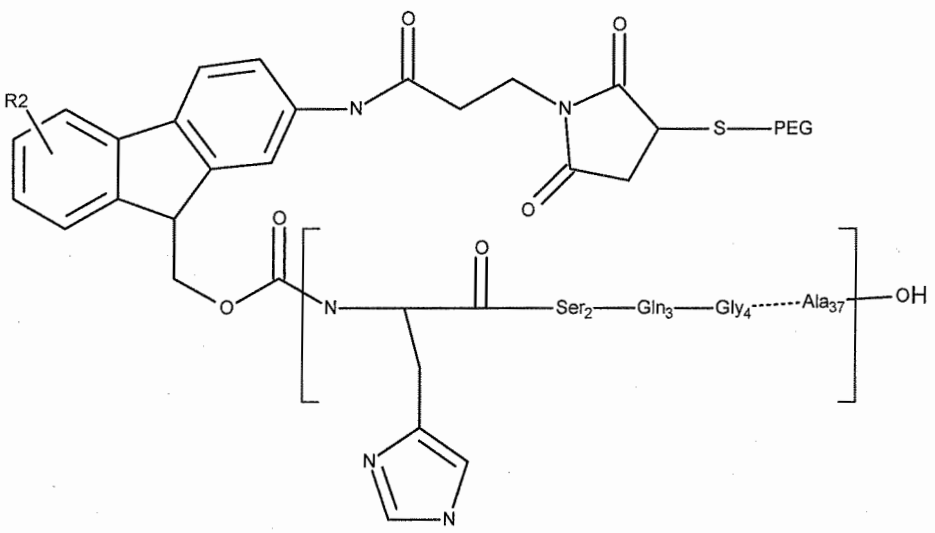
6. Use of a conjugate according to any one of claim 1 or claim 2, or use of pharmaceutical composition according to claim 4 in the preparation of a medicament for extending the biological half-life of oxyntomodulin.

7. A method of reducing the dosing frequency of oxyntomodulin, said method comprising administering a pharmaceutical composition of claim 4.

8. Use of a conjugate according to any one of claim 1 or claim 2, or use of pharmaceutical composition according to claim 4 in the preparation of a medicament for reducing the dosing frequency of oxyntomodulin.

9. Use of a conjugate according to any one of claim 1 or claim 2, or use of a composition according to claim 4, in the manufacture of a medicament for treating obesity.

10. A process for preparing a conjugate of claim 1, represented by the following structure:

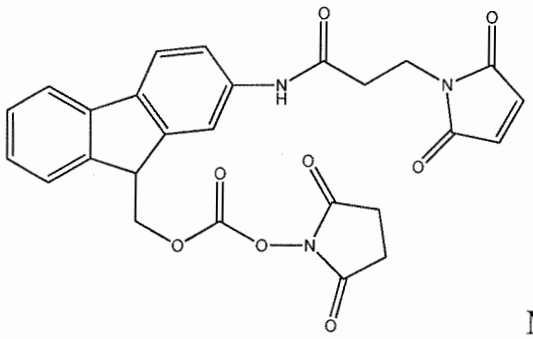


oxyntomodulin

His [1] - Ser [2] - Gln [3] - Gly [4] - Thr [5]- Phe [6] - Thr [7] - Ser [8] - Asp [9] - Tyr [10] - Ser [11] - Lys [12] - Tyr [13] - Leu [14] - Asp [15] - Ser [16] - Arg [17] - Arg [18]- Ala [19] - Gln [20] - Asp [21] - Phe [22] - Val [23] - Gln [24] - Trp [25] - Leu [26] - Met [27] - Asn [28] - Thr [29] - Lys [30] - Arg [31]- Asn [32] - Arg [33] - Asn [34] - Asn [35] - Ile [36] - Ala [37] (SEQ ID NO: 1)

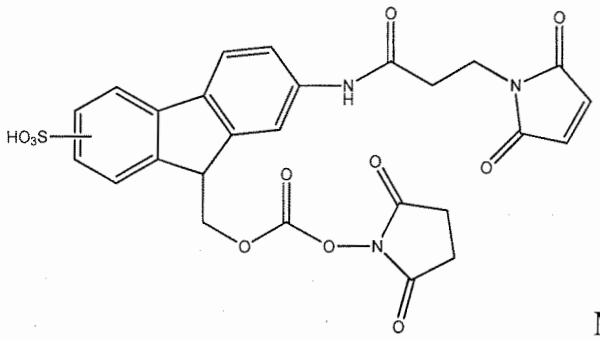
wherein said oxyntomodulin (OXM; of SEQ ID NO: 1) is linked *via* the terminal amino group of said oxyntomodulin and R_2 is hydrogen or SO_3H, representing PEG-Fmoc-OXM (N-terminus) and PEG-FMS-OXM (N-terminus), respectively; and

said process comprising reacting MAL-FMS-NHS or MAL-Fmoc-NHS:



MAL-Fmoc-NHS

or



MAL-FMS-NHS

with

a resin-bound oxyntomodulin of SEQ ID NO: 1, wherein the amino side chains of Lys^{12} and Lys^{30} of said resin-bound oxyntomodulin are each protected by a protecting group,

to obtain a resin-bound MAL-Fmoc-protected OXM (N-terminus) or a resin-bound MAL-FMS-protected OXM (N-terminus), respectively, wherein the amino side chains of said Lys^{12} and Lys^{30} are each protected by a protecting group,

followed by:

(a) reaction of the resin-bound MAL-Fmoc-protected OXM (N-terminus) or the resin-bound MAL-FMS-protected OXM (N-terminus) with sulfhydryl PEG polymer (PEG-SH) and subsequent removal of said protecting groups and said resin; or

(b) removal of the resin and protecting groups to provide a MAL-Fmoc-OXM (N-terminus) and a MAL-FMS-OXM (N-terminus), respectively, and subsequent reaction of the MAL-Fmoc-OXM or the MAL-FMS-OXM with sulfhydryl PEG polymer (PEG-SH),

to obtain PEG-Fmoc-OXM (N-terminus) or PEG-FMS-OXM (N-terminus), respectively.

11. The process of claim 10, wherein said PEG is represented PEG 30.

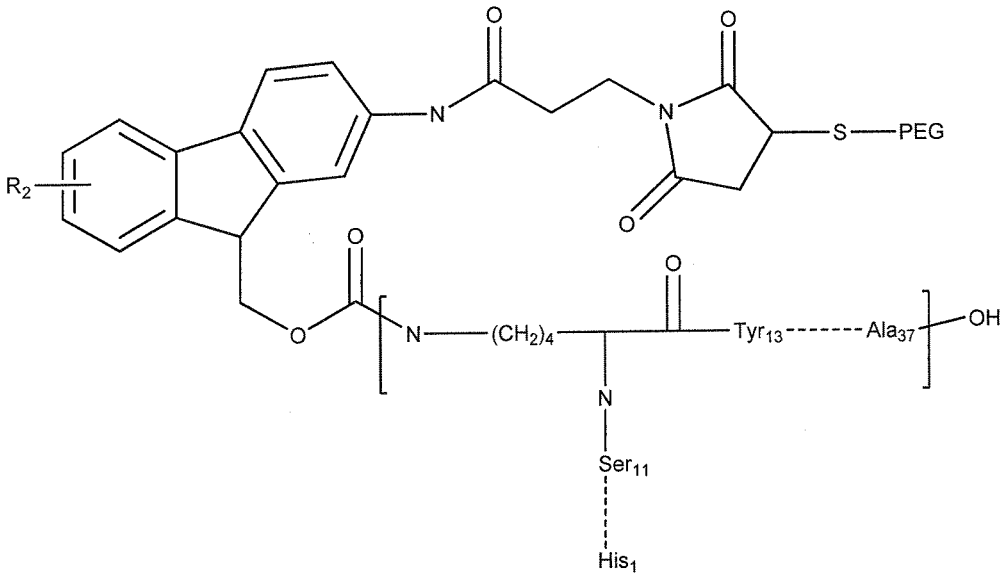
12. The process of claim 10, wherein said R_2, or SO_3H is at position 2 of the fluorene ring.

13. The process of claim 10, wherein said protecting groups of said amino groups of said lysine^{12} and lysine^{30} of MAL-Fmoc-protected OXM (N-terminus) and said MAL-FMS-protected OXM (N-terminus) are removed to obtain MAL-Fmoc-OXM (N-terminus) and MAL-FMS-OXM (N-terminus), respectively, and said MAL-Fmoc-OXM (N-terminus) and MAL-FMS-OXM (N-terminus) are further purified.

14. The process of claim 10, wherein said protecting group of said side chain of said Lys^{12} and Lys^{30} is ivDde (1-[(4,4-dimethyl-2,6-dioxocyclohex-1-ylidine)ethyl]).

15. The process of claim 10, wherein said protecting group is removed under basic conditions.

16. A process for preparing a conjugate of claim 1 represented by the following structure:

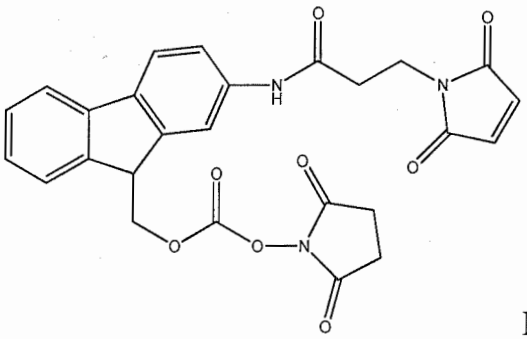


oxyntomodulin

His [1] - Ser [2] - Gln [3] - Gly [4] - Thr [5]- Phe [6] - Thr [7] - Ser [8] - Asp [9] - Tyr [10] - Ser [11] - Lys [12] - Tyr [13] - Leu [14] - Asp [15] - Ser [16] - Arg [17] - Arg [18]- Ala [19] - Gln [20] - Asp [21] - Phe [22] - Val [23] - Gln [24] - Trp [25] - Leu [26] - Met [27] - Asn [28] - Thr [29] - Lys [30] - Arg [31]- Asn [32] - Arg [33] - Asn [34] - Asn [35] - Ile [36] - Ala [37] (SEQ ID NO: 1)

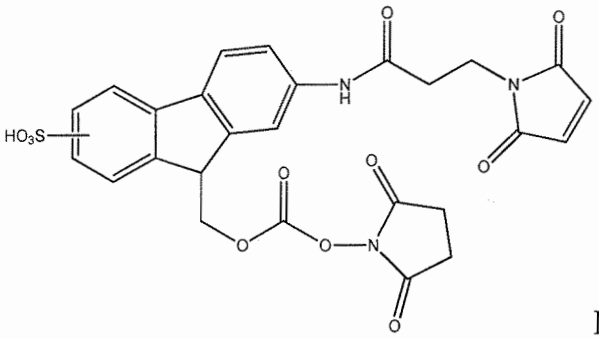
wherein said oxyntomodulin (OXM; of SEQ ID NO: 1) is linked *via* the amino side chain of said Lys^{12} of said oxyntomodulin and R_2 is hydrogen or SO_3H, representing PEG-Fmoc-OXM (Lys^{12}) and PEG-FMS-OXM (Lys^{12}), respectively; and

said process comprising reacting MAL-FMS-NHS or MAL-Fmoc-NHS:



MAL-Fmoc-NHS

or



MAL-FMS-NHS

with

a resin-bound oxyntomodulin of SEQ ID NO: 1, wherein the amino side chain of Lys^{30} and the amino terminus of His^1 of said oxyntomodulin are each protected by a protecting group;

to obtain a resin-bound MAL-Fmoc-protected OXM (Lys^{12}) or a resin-bound MAL-FMS-protected OXM (Lys^{12}), respectively, wherein the amino side chain of Lys^{30} and the amino terminus of His^1 of said oxyntomodulin are each protected by said protecting group;

followed by:

(a) reaction of the resin-bound MAL-Fmoc-protected OXM (Lys^{12}) or the resin-bound MAL-FMS-protected OXM (Lys^{12}) with sulfhydryl PEG polymer (PEG-SH) and subsequent removal of said protecting groups and said resin; or

(b) removal of the resin and protecting groups to provide a MAL-Fmoc-OXM (Lys^{12}) and a MAL-FMS-OXM (Lys^{12}), respectively, and subsequent reaction of the MAL-Fmoc-OXM (Lys^{12}) or the MAL-FMS-OXM (Lys^{12}) with sulfhydryl PEG polymer (PEG-SH),

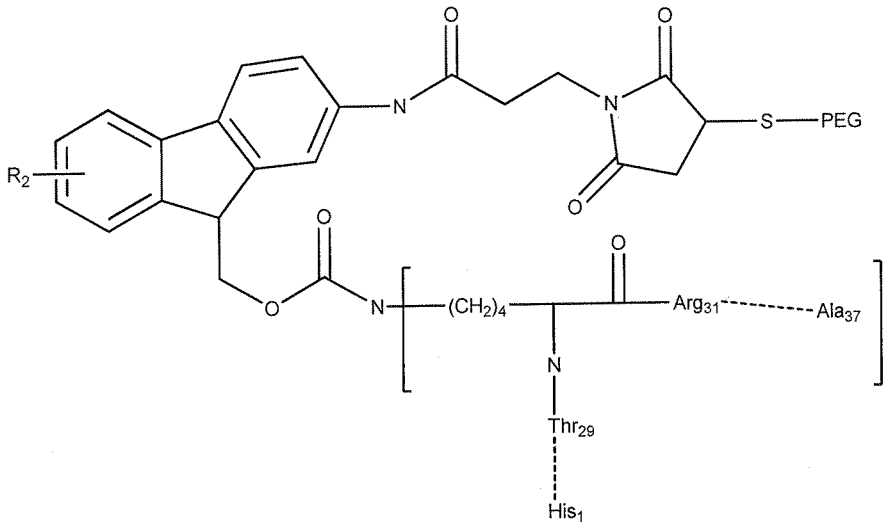
to obtain PEG-Fmoc-OXM (Lys^{12}) or PEG-FMS-OXM (Lys^{12}), respectively.

17. The process of claim 16, wherein said PEG is PEG 30.

18. The process of claim 16, wherein said R_2, or SO_3H is at position 2 of the fluorene ring.

19. The process of claim 16, wherein said protecting groups of said amino groups of said $lysine^{30}$ and $histidine^{1}$ of MAL-Fmoc-protected OXM (Lys^{12}) and said MAL-FMS-protected OXM (Lys^{12}) are removed to obtain MAL-Fmoc-OXM (Lys^{12}) and MAL-FMS-OXM (Lys^{12}), respectively, and said MAL-Fmoc-OXM (Lys^{12}) and MAL-FMS-OXM (Lys^{12}) are further purified.

20. A process for preparing a conjugate of claim 1 represented by the following structure:

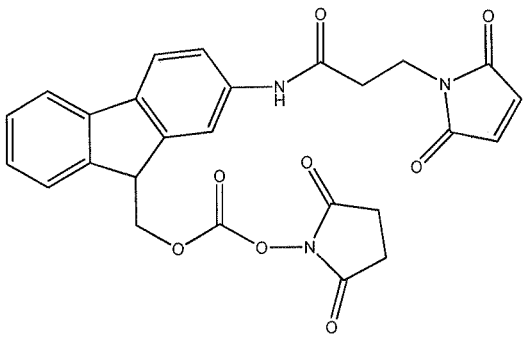


Oxyntomodulin

His [1] - Ser [2] - Gln [3] - Gly [4] - Thr [5]- Phe [6] - Thr [7] - Ser [8] - Asp [9] - Tyr [10] - Ser [11] - Lys [12] - Tyr [13] - Leu [14] - Asp [15]- Ser [16] - Arg [17] - Arg [18]- Ala [19] - Gln [20] - Asp [21] - Phe [22] - Val [23] - Gln [24] - Trp [25] - Leu [26] - Met [27] - Asn [28] - Thr [29]- Lys [30] - Arg [31]- Asn [32] - Arg [33] - Asn [34] - Asn [35] - Ile [36] - Ala [37] (SEQ ID NO: 1)

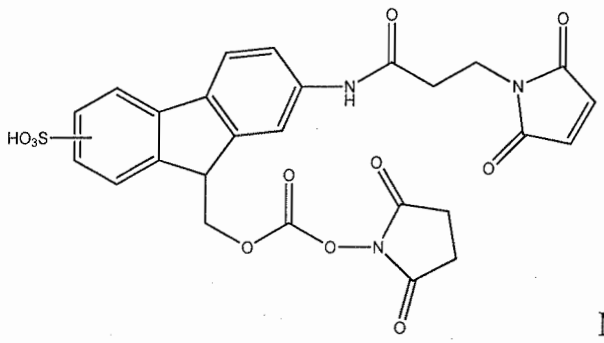
wherein said oxyntomodulin (OXM; of SEQ ID NO: 1) is linked *via* the amino side chain of said Lys^{30} of oxyntomodulin, R_2 is hydrogen or SO_3H, representing PEG-Fmoc-OXM (Lys^{30}) and PEG-FMS-OXM (Lys^{30}), respectively; and

said process comprising reacting MAL-FMS-NHS or MAL-Fmoc-NHS :



MAL-Fmoc-NHS

or



MAL-FMS-NHS

with

a resin-bound oxyntomodulin of SEQ ID NO: 1, wherein the amino side chain of Lys^{12} and the amino terminus of His^{1} of said oxyntomodulin are each protected by a protecting group;

to obtain a resin-bound MAL-Fmoc-protected OXM (Lys^{30}) or MAL-FMS-protected OXM (Lys^{30}), wherein the amino side chain of Lys^{12} and the amino terminus of His^{1} of said oxyntomodulin are each protected by a protecting group,

followed by:

(a) reaction of the resin-bound MAL-Fmoc-protected OXM (Lys^{30}) or the resin-bound MAL-FMS-protected OXM (Lys^{30}) with sulfhydryl PEG polymer (PEG-SH) and subsequent removal of said protecting groups and said resin; or

(b) removal of the resin and protecting groups to provide a MAL-Fmoc-OXM (Lys^{30}) and a MAL-FMS-OXM (Lys^{30}), respectively, and subsequent reaction of the MAL-Fmoc-OXM or the MAL-FMS-OXM with sulfhydryl PEG polymer (PEG-SH),

to yield PEG-Fmoc-OXM (Lys^{30}) or PEG-FMS-OXM (Lys^{30}).

21. The process of claim 20, wherein said PEG is PEG30.

22. The process of claim 20, wherein said R_2, or SO_3H is at position 2 of the fluorene ring.

23. The process of claim 20, wherein said protecting groups of said amino groups of said lysine^{12} and histidine^{1} of MAL-Fmoc-protected OXM (Lys^{30}) and said MAL-FMS-protected OXM (Lys^{30}) are removed to obtain MAL-Fmoc-OXM (Lys^{30}) and MAL-FMS-OXM (Lys^{30}), respectively, and said MAL-Fmoc-OXM (Lys^{30}) and MAL-FMS-OXM (Lys^{30}) are further purified.