

AMENDMENTS TO THE CLAIMS

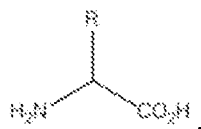
This claim listing will serve to replace all prior versions of the claims in the application.

What is claimed is:

1. (Currently Amended) A modified relaxin polypeptide comprising a non-naturally encoded amino acid, wherein:

(a) the modified relaxin polypeptide comprises the relaxin A chain polypeptide of SEQ ID NO: 4 and the relaxin B chain polypeptide of SEQ ID NO: 5 or SEQ ID NO: 6, substituted with a non-naturally encoded amino acid at ~~a position selected from the group consisting of: A chain residue 1, A chain residue 2, A chain residue 5, A chain residue 13, A chain residue 18, B chain residue 5, B chain residue 7, and B chain residue 25,~~ and having zero, one, or two additional amino acid changes in said relaxin A chain and having zero, one, or two additional amino acid changes in said relaxin B chain, wherein each of said amino acid changes is independently a substitution, insertion or deletion;

(b) said non-naturally encoded amino acid has the structure:



wherein the R group is any substituent other than the side chain found in alanine, arginine, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine or valine; and

(c) said non-naturally encoded amino acid is linked to a pharmacokinetic enhancer comprising a peptide component having between 4 and 8 2 and 30 amino acids and a half-life extending moiety comprising a fatty acid of Formula I:

-C_n-COOH (Formula I)

wherein n is between 12 and 16, wherein said peptide component comprises Glu^γ.

2. (Cancelled)
3. (Cancelled)
4. (Cancelled)

5. (Cancelled)
6. (Previously Presented) The modified relaxin polypeptide of claim 1, wherein said non-naturally encoded amino acid comprises a carbonyl group, an aminooxy group, a hydrazide group, a hydrazine group, a semicarbazide group, an azide group, or an alkyne group.
7. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said non-naturally encoded amino acid comprises a phenylalanine derivative, ~~a para-substituted phenylalanine, an ortho-substituted phenylalanine, a meta-substituted phenylalanine, or para-acetyl L-phenylalanine.~~
8. (Cancelled)
9. (Cancelled)
10. (Cancelled)
11. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said non-naturally encoded amino acid is directly linked to said peptide component.
12. (Previously Presented) The modified relaxin polypeptide of claim 1, wherein said non-naturally encoded amino acid is linked to said pharmacokinetic enhancer through an oxime linkage or triazole linkage.
13. (Cancelled)
14. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said relaxin A chain polypeptide comprises SEQ ID NO: 35, and said relaxin B chain polypeptide comprises SEQ ID NO: 5 or SEQ ID NO: 6, ~~and having zero, one, or two additional amino acid changes in said relaxin A chain and having zero, one, or two additional amino acid changes in said relaxin B chain, wherein each of said amino acid changes is independently a substitution, insertion or deletion.~~
15. (Cancelled)
16. (Cancelled)
17. (Cancelled)
18. (Cancelled)
19. (Cancelled)
20. (Cancelled)
21. (Cancelled)

22. (Cancelled)

23. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said peptide component comprises Glu, ~~Glu^γ~~, GGGGS-Glu^γ (SEQ ID NO: 139), DRDDRD (SEQ ID NO: 102), KKKKKK-Glu^γ (SEQ ID NO: 103), ~~RGGEKKKKEKEK-Glu^γ~~ (~~SEQ ID NO: 104~~), GGEEEE-Glu^γ (SEQ ID NO: 105), EEEGGG-Glu^γ (SEQ ID NO: 106), KKKGGG-Glu^γ (SEQ ID NO: 107), ~~GETGSSGEGT-Glu^γ~~ (~~SEQ ID NO: 108~~), GGGKKK-Glu^γ (SEQ ID NO: 109), ~~GSHHHHHHGS-Glu^γ~~ (~~SEQ ID NO: 110~~), ~~Sar-Sar-Sar-Sar-Ser-Sar-Sar-Sar-Sar-Glu^γ~~ (~~SEQ ID NO: 111~~), Sar-Sar-Sar-Sar-Ser-Glu^γ (SEQ ID NO: 112), Sar-Sar-Sar-Glu-Glu-Glu^γ (SEQ ID NO: 113), KKKSGGSGG-Glu^γ (~~SEQ ID NO: 118~~), KKSAGSAG-Glu^γ (~~SEQ ID NO: 116~~), KSGGSGG-Glu^γ (SEQ ID NO: 117), ~~KKSGGSGGEE-Glu^γ~~ (~~SEQ ID NO: 119~~), dKdKdKdKdKdK-Glu^γ (SEQ ID NO: 120), ~~EESGGSGG-Glu^γ~~ (~~SEQ ID NO: 121~~), ~~GSGSGSGS-Glu^γ~~ (~~SEQ ID NO: 123~~), EEEGGG-dGlu^γ (SEQ ID NO: 128), EGGGGSK-Glu^γ (SEQ ID NO: 130), EEEEE-Glu^γ (SEQ ID NO: 131), ~~EEEEPEEEEEPEEEEEPEEEEE-Glu^γ~~ (~~SEQ ID NO: 133~~), ~~EEEEPEEEEEPEEEEEPEEGGG~~ (~~SEQ ID NO: 135~~), ~~EEEEGEEEEGEEEEGEEEE-Glu^γ~~ (~~SEQ ID NO: 136~~), ~~KGGEKKKKEKEKEPKGGEKKKKEKEK-Glu^γ~~ (~~SEQ ID NO: 137~~), ~~EAQKAQAEAQKAQAEAQKAQA-Glu^γ~~ (~~SEQ ID NO: 138~~), KK-Glu^γ (SEQ ID NO: 140), ~~Glu^γ~~, KGPKGP-Glu^γ (SEQ ID NO: 146), SGGGS-Glu^γ (SEQ ID NO: 147), KGGGS-Glu^γ (SEQ ID NO: 148), KGGGSE-Glu^γ (SEQ ID NO: 149), GSPGSP-Glu^γ (SEQ ID NO: 150), GGGGP-Glu^γ (SEQ ID NO: 151), EGGS-Glu^γ (SEQ ID NO: 152), EGGGP-Glu^γ (SEQ ID NO: 153), KGPGSE-Glu^γ (SEQ ID NO: 154), Spermine-Glu^γ, or KKGGS-Glu^γ (SEQ ID NO: 156).

24. (Cancelled)

25. (Cancelled)

26. (Cancelled)

27. (Previously Presented) The modified relaxin polypeptide of claim 1, wherein the relaxin polypeptide comprises the relaxin A chain polypeptide of SEQ ID NO: 4 and the relaxin B chain polypeptide of SEQ ID NO: 5 or SEQ ID NO: 6, substituted with a non-naturally encoded amino acid at A chain residue 1, wherein said non-naturally encoded amino acid is linked to said pharmacokinetic enhancer and said

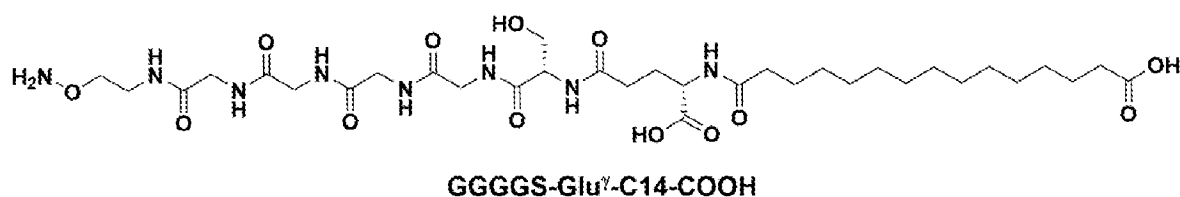
pharmacokinetic enhancer comprises the peptide component GGGGS-Glu^γ (SEQ ID NO: 139).

28. (Previously Presented) The modified relaxin polypeptide of claim 1, wherein said half-life extending moiety comprises a fatty acid or derivative thereof, wherein the fatty acid or derivative thereof is covalently linked to the peptide component.
29. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said half-life extending moiety comprises ~~a saturated fatty acid or derivative thereof, or comprises a fatty acid terminated with a carboxylic acid, or comprises -C14-COOH.~~
30. (Cancelled)
31. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said half-life extending moiety comprises a fatty acid of Formula I:

-C_n-COOH (Formula I)

wherein n is between 12 and 14~~10 and 18~~.

32. (Cancelled)
33. (Cancelled)
34. (Cancelled)
35. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said pharmacokinetic enhancer comprises:



(Formula II)

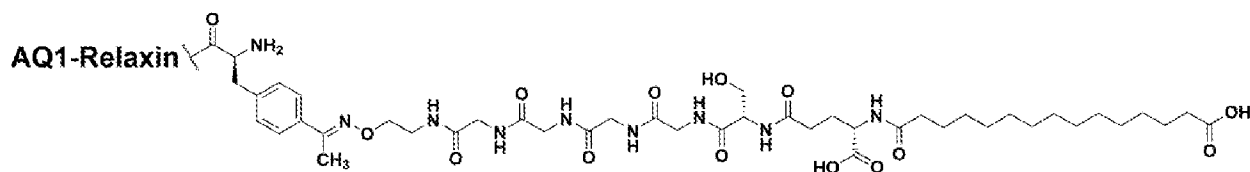
wherein the aminooxy group is linked to the non-natural amino acid in said modified relaxin polypeptide.

36. (Previously Presented) The modified relaxin polypeptide of claim 1, wherein said half-life extending moiety is conjugated to said peptide component through an amide bond.
37. (Original) The modified relaxin polypeptide of claim 1, wherein the relaxin polypeptide comprises the relaxin A chain polypeptide of SEQ ID NO: 4 and the

relaxin B chain polypeptide of SEQ ID NO: 5 or SEQ ID NO: 6, substituted with a non-naturally encoded amino acid at A chain residue 1;

wherein said non-naturally encoded amino acid comprises para-acetyl-L-phenylalanine linked to said pharmacokinetic enhancer which comprises
a peptide component comprising GGGGS-Glu^γ (SEQ ID NO: 139); and
a half-life extending moiety comprising -C14-COOH.

38. (Original) A modified relaxin polypeptide comprising:



(Formula III),

wherein said AQ1-Relaxin comprises a relaxin A chain polypeptide having ~~at least 90% the~~ amino acid sequence ~~of identity to~~ SEQ ID NO: 35 and a relaxin B chain polypeptide having ~~at least 90% the~~ amino sequence ~~identity to of~~ SEQ ID NO: 5 or SEQ ID NO: 6, and having zero, one, or two amino acid changes in said relaxin A chain and having zero, one, or two amino acid changes in said relaxin B chain, wherein the modified para-acetyl-phenylalanine depicted in Formula III is located at the N-terminus of said relaxin A chain polypeptide.

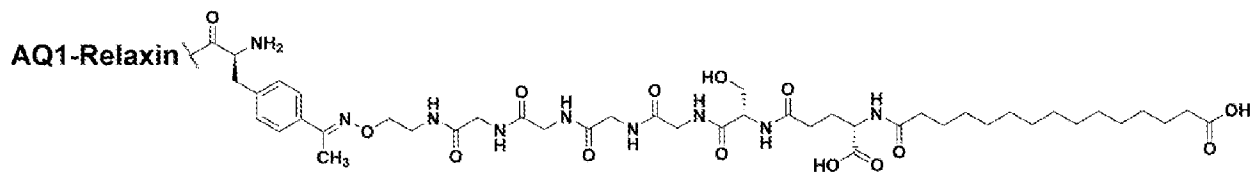
39. (Cancelled)

40. (Cancelled)

41. (Original) The modified relaxin polypeptide of claim 38, wherein AQ1-Relaxin comprises a relaxin A chain polypeptide of SEQ ID NO: 35 and a relaxin B chain polypeptide of SEQ ID NO: 5 or SEQ ID NO: 6.

42. (Currently Amended) A modified relaxin polypeptide comprising AQ1-GGGGS-Glu^γ (SEQ ID NO: 139)-C14-COOH ~~which comprises the structure shown in FIG. 8,~~ which comprises the structure shown in FIG. 9, or which comprises a relaxin A chain polypeptide of SEQ ID NO: 35 and a relaxin B chain polypeptide of SEQ ID NO: 6, wherein the modified para-acetyl-phenylalanine located at the N-terminus of said

relaxin A chain polypeptide is linked to ~~PK~~ the pharmacokinetic enhancer GGGGS-
Glu⁺ (SEQ ID NO: 139) C14 COOH, as depicted in Formula III:



(Formula III).

43. (Cancelled)

44. (Cancelled)

45. (Cancelled)

46. (Cancelled)

47. (Cancelled)

48. (Cancelled)

49. (Cancelled)

50. (Cancelled)

51. (Currently Amended) The modified relaxin polypeptide of claim 1, wherein said modified relaxin polypeptide exhibits an increased in vivo half-life by at least 2-fold compared to the wild-type relaxin polypeptide consisting of the relaxin A chain of SEQ ID NO: 4 linked to the relaxin B chain of SEQ ID NO: 5.

52. (Cancelled)

53. (Previously Presented) A pharmaceutical composition comprising the modified relaxin polypeptide of claim 1 and a pharmaceutically acceptable carrier.

54. (Cancelled)

55. (Cancelled)

56. (Cancelled)

57. (Cancelled)

58. (Cancelled)

59. (Cancelled)

60. (Cancelled)

61. (Cancelled)

62. (Cancelled)
63. (Cancelled)
64. (Cancelled)
65. (Cancelled)
66. (Cancelled)
67. (Cancelled)
68. (Cancelled)
69. (Cancelled)
70. (Previously Presented) A method of manufacturing a modified relaxin polypeptide according to claim 1, comprising:
- (a) providing a polypeptide comprising said relaxin A chain and said relaxin B chain, wherein said polypeptide comprises said non-naturally encoded amino acid; and
 - (b) linking said non-naturally encoded amino acid to said pharmacokinetic enhancer.
71. (Cancelled)
72. (Cancelled)
73. (Cancelled)
74. (Cancelled)
75. (Cancelled)
76. (Previously Presented) The modified relaxin polypeptide of claim 42, which comprises the structure shown in FIG. 9.
77. (Previously Presented) The modified relaxin polypeptide of claim 76, which consists of the structure shown in FIG. 9.
78. (Currently Amended) The modified relaxin polypeptide of claim 76, wherein said modified relaxin polypeptide exhibits an increased in vivo half-life by at least 2-fold compared to the wild-type relaxin polypeptide consisting of the relaxin A chain of SEQ ID NO: 4 linked to the relaxin B chain of SEQ ID NO: 5.

79. (Currently Amended) The modified relaxin polypeptide of claim 77, wherein said modified relaxin polypeptide exhibits an increased in vivo half-life by at least 2-fold compared to the wild-type relaxin polypeptide consisting of the relaxin A chain of SEQ ID NO: 4 linked to the relaxin B chain of SEQ ID NO: 5.
80. (Previously Presented) A pharmaceutical composition comprising the modified relaxin polypeptide of claim 38 and a pharmaceutically acceptable carrier.
81. (Previously Presented) A pharmaceutical composition comprising the modified relaxin polypeptide of claim 42 and a pharmaceutically acceptable carrier.
82. (Previously Presented) A pharmaceutical composition comprising the modified relaxin polypeptide of claim 76 and a pharmaceutically acceptable carrier.
83. (Previously Presented) A pharmaceutical composition comprising the modified relaxin polypeptide of claim 77 and a pharmaceutically acceptable carrier.
84. (New) The modified relaxin polypeptide of claim 1, wherein said modified relaxin polypeptide exhibits an increased in vivo half-life by at least 5-fold compared to the wild-type relaxin polypeptide consisting of the relaxin A chain of SEQ ID NO: 4 linked to the relaxin B chain of SEQ ID NO: 5.
85. (New) The modified relaxin polypeptide of claim 1, wherein said modified relaxin polypeptide exhibits an increased in vivo half-life by at least 10-fold compared to the wild-type relaxin polypeptide consisting of the relaxin A chain of SEQ ID NO: 4 linked to the relaxin B chain of SEQ ID NO: 5.