The present description refers to a number of documents, the contents of which are herein incorporated by reference in their entirety.

SUMMARY

The present disclosure relates to bioactive peptides conjugated to polyethylene glycol (PEG) and uses thereof, such as analogs of GHRH with one or more attached PEG moieties.

Disclosed is a growth hormone releasing hormone (GHRH) analog comprising a peptide having the sequence set forth in formula (I) (SEQ ID NO: 1):

X1-X2-Asp-Ala-Ile-Phe-Thr-X8-X9-Tyr-X11-X12-Val-Leu-X15-Gln-Leu-X18-Ala-X20-X21-X22-Leu-X24-X25-X26-X27-X28-X29-X30

(I)

wherein:

X1 is Tyr, His, N-methyl Tyr or Desamino Tyr,

X2 is Ala, D-Ala, Ser or alpha aminoisobutyric acid (Aib);

X8 is Asn, Asp, Ala, Gln, Ser or Aib;

X9 is Ser, Asp, Ala or Cys conjugated to a PEG polymer of less than or equal to about 5 kDa;

X11 is Arg or L-Homoarginine (Har);

X12 is Lys or L-Ornithine (Orn);

X15 is Gly or Ala;

X18 is Ser or Ala;

X20 is Arg or Har;

X21 is Lys or Orn;

X22 is Leu, Val, Ala or Cys conjugated to a PEG polymer of less than or equal to about 5 kDa;

X24 is Gln or Cys conjugated to a PEG polymer of less than or equal to about 5 kDa;

X25 is Asp or Ala;

X26 is Ile or Ala;

X27 is Met, Leu or Norleucine;

X28 is Ser, Ala or Cys conjugated to a PEG polymer of less than or equal to about 5 kDa;

X29 is Lys, Arg, Orn, Har, or Agmantine; and

X30 is an NH_2 group or Asn-NH_2;

wherein at least one of X9, X22, X24 and X28 is Cys conjugated to a PEG polymer of less than or equal to about 5 kDa;

or a pharmaceutically acceptable salt thereof.

In one aspect, there is provided a growth hormone releasing hormone (GHRH) analog comprising a peptide having the sequence set forth in formula (I) (SEQ ID NO: 1):

X1-X2-Asp-Ala-Ile-Phe-Thr-X8-X9-Tyr-X11-X12-Val-Leu-X15-Gln-Leu-X18-Ala-X20-X21-X22-Leu-X24-X25-X26-X27-X28-X29-X30

(I)

wherein:

X1 is Tyr, His, N-methyl Tyr or Desamino Tyr;

X2 is Ala, D-Ala, Ser or alpha aminoisobutyric acid (Aib);

X8 is Asn, Asp, Ala, Gln, Ser or Aib;

X9 is Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X11 is Arg or L-Homoarginine (Har);

X12 is Lys or Orn;

X15 is Gly or Ala;

X18 is Ser or Ala;

X20 is Arg or Har;

X21 is Lys or Orn;

X22 is Leu, Val, Ala or Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X24 is Gln or Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X25 is Asp or Ala;

X26 is Ile or Ala;

X27 is Met, Leu or Norleucine;

X28 is Ser, Ala or Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X29 is Lys, Arg, Orn, Har or Agmatine; and

X30 is an NH_2 group or Asn-NH_2;

or a pharmaceutically acceptable salt thereof.

In an embodiment, X9 is Cys conjugated to a PEG polymer of less than or equal to about 5 kDa. In another embodiment, X22 is Cys conjugated to a PEG polymer of less than or equal to about 5 kDa. In another embodiment, X24 is Cys conjugated to a PEG polymer of less than or equal to about 5 kDa. In yet another embodiment, X28 is Cys conjugated to a PEG polymer of less than or equal to about 5 kDa.

In an embodiment, X9 and X22 are each independently Cys conjugated to a PEG polymer of less than or equal to about 5 kDa. In another embodiment, X9 and X24 are each independently Cys conjugated to a PEG polymer of less than or equal to about 5 kDa. In yet another embodiment, X9 and X28 are each independently Cys conjugated to a PEG polymer

of less than or equal to about 5 kDa.

In some embodiments, X1 is Tyr. In some embodiments, X2 is Ala or D-Ala. In an embodiment, X2 is D-Ala. In some embodiments, X8 is Ala.In some embodiments, X9 is Ser. In some embodiments, X11 is Arg or Har; in an embodiment, X11 is Arg. In some embodiments, X12 is Lys or Orn; in an embodiment, X12 is Orn. In some embodiments, X15 is Ala. In some embodiments, X18 is Ser. In some embodiments, X20 is Arg or Har; in an embodiment, X20 is Arg. In some embodiments, X21 is Lys or Orn; in an embodiment, X21 is Orn. In some embodiments, X22 is Ala. In some embodiments, X24 is Gln. In some embodiments, X25 is Asp. In some embodiments, X26 is Ile. In some embodiments, X27 is Leu. In some embodiments, X28 is Ser. In some embodiments, X29 is Lys, Orn or Har; in an embodiment, X29 is Lys; in another embodiment, X29 is Har. In In some embodiments, X30 is Asn-NH_2.

In some embodiments of formula (I), the PEG polymer of less than or equal to about 5 kDa described herein is a PEG polymer of about 2 kDa.

In some embodiments, the GHRH analog provided herein comprises a peptide which is:

YaDAIFTAC(Peg-5K)YROVLAQLSAROALQDILSZ (SEQ ID NO: 2);

YaDAIFTASYROVLAQLSAROC(Peg-5K)LQDILSZ (SEQ ID NO: 3);

YaDAIFTASYROVLAQLSAROALC(Peg-5K)DILSZ (SEQ ID NO: 4);

YaDAIFTASYROVLAQLSAROALQDILC(Peg-5K)Z (SEQ ID NO: 5);

YaDAIFTAC(Peg-5K)YROVLAQLSAROC(Peg-5K)LQDILSZ (SEQ ID NO: 6);

YaDAIFTAC(Peg-5K)YROVLAQLSAROALC(Peg-5K)DILSZ (SEQ ID NO: 7); or

YaDAIFTAC(Peg-5K)YROVLAQLSAROALQDILC(Peg-5K)Z (SEQ ID NO: 8);

wherein: a is D-Ala; O is L-Ornithine (Orn); C(Peg-5K) is Cys conjugated to a PEG polymer of less than or equal to about 5 kDa; and Z is Lys, Arg, Har or Agmantine; or a pharmaceutically acceptable salt thereof. In some embodiments, the GHRH analog

GHRH analog or salt thereof provided herein. In an embodiment, the pharmaceutical composition further comprises one or more pharmaceutically acceptable carriers, excipients, and/or diluents.

Disclosed is a method for inducing growth hormone secretion in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein.

Disclosed is a method for increasing the level of growth hormone in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein.

Disclosed is a use of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein, for inducing growth hormone secretion in a subject.

Disclosed is a use of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein, for increasing the level of growth hormone in a subject.

Disclosed is a use of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein, for the preparation of a medicament for inducing growth hormone secretion or increasing the level of growth hormone in a subject.

In another aspect, there is provided use of the GHRH analog or salt thereof as described herein or of the pharmaceutical composition as described herein for the preparation of a medicament for inducing growth hormone secretion in a subject.

Disclosed is a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein, for use in inducing growth hormone secretion or increasing the level of growth hormone in a subject.

Disclosed is a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein, for the preparation of a medicament for inducing growth hormone secretion or increasing the level of growth hormone in a subject.

Disclosed is a method for treating, preventing, or diagnosing a GH-deficient condition in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein. Non-limiting examples of GH-deficient conditions that may be treated, prevented and/or diagnosed using methods provided herein include: HIV-associated lipohypertophy; HIV-associated dysmorphia/dysmetabolic syndrome (HADDS); growth hormone deficiency; Hypothalamic GH-RH deficiency; constitutional growth delay; pituitary dwarfism; growth retardation; wound or bone healing; osteoporosis; Turner Syndrome; Familial short stature; chronic renal insufficiency and severe growth retardation; intrauterine

growth retardation; GH deficiency following radiotherapy for pituitary or hypothalamic lesions; long-term treatment with glucocorticoids associated with subnormal rate of growth; and abdominal obesity associated with metabolic syndrome or HIV-associated lipohypertrophy.

In another aspect, there is provided use of the GHRH analog or salt thereof as described herein or the pharmaceutical composition as described herein in the manufacture of a medicament for treating, preventing, or diagnosing a GH-deficient condition in a subject in need thereof.

Disclosed is a method for diagnosing a growth hormone deficiency in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein, and measuring the growth hormone response.

In an embodiment, there is provided a method for treating HIV-related lipodystrophy in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein. In embodiments, the treating comprises treating visceral fat accumulation. In embodiments, the subject is receiving antiviral therapy.

In an embodiment, there is provided a method for treating pituitary dwarfism or growth retardation in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein.

In an embodiment, there is provided a method for the treatment of wound or bone healing in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein.

In an embodiment, there is provided a method for the treatment of osteoporosis in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein.

In an embodiment, there is provided a method for improving protein anabolism in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein.

In an embodiment, there is provided a method for inducing a lipolytic effect in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described herein, or a pharmaceutical composition described herein. In embodiments, the subject is inflicted with clinical obesity.

In an embodiment, there is provided a method for the treatment of HIV-associated dysmorphia/dysmetabolic syndrome (HADDS) in a subject in need thereof, comprising administering to the subject an effective amount of a GHRH analog or salt thereof described

CLAIMS

WHAT IS CLAIMED IS:

1. A growth hormone releasing hormone (GHRH) analog comprising a peptide having the sequence set forth in formula (I) (SEQ ID NO: 1):

X1-X2-Asp-Ala-Ile-Phe-Thr-X8-X9-Tyr-X11-X12-Val-Leu-X15-Gln-Leu-X18-Ala-X20-X21-X22-Leu-X24-X25-X26-X27-X28-X29-X30

(I)

wherein:

X1 is Tyr, His, N-methyl Tyr or Desamino Tyr;

X2 is Ala, D-Ala, Ser or alpha aminoisobutyric acid (Aib);

X8 is Asn, Asp, Ala, Gln, Ser or Aib;

X9 is Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X11 is Arg or L-Homoarginine (Har);

X12 is Lys or Orn;

X15 is Gly or Ala;

X18 is Ser or Ala;

X20 is Arg or Har;

X21 is Lys or Orn;

X22 is Leu, Val, Ala or Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X24 is Gln or Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X25 is Asp or Ala;

X26 is Ile or Ala;

X27 is Met, Leu or Norleucine;

X28 is Ser, Ala or Cys conjugated to a PEG polymer of less than or equal to 5 kDa;

X29 is Lys, Arg, Orn, Har or Agmatine; and

X30 is an NH_2 group or Asn-NH_2;

or a pharmaceutically acceptable salt thereof.

2. The GHRH analog or salt thereof of claim 1, wherein at least one of X22, X24 and X28 is Cys conjugated to a PEG polymer of less than or equal to 5 kDa.

3. The GHRH analog or salt thereof of claim 1, wherein X22 is Cys conjugated to a PEG polymer of less than or equal to 5 kDa.

4. The GHRH analog or salt thereof of any one of claims 1 and 3, wherein X24 is Cys

conjugated to a PEG polymer of less than or equal to 5 kDa.

5. The GHRH analog or salt thereof of any one of claims 1, 3, and 4, wherein X28 is Cys conjugated to a PEG polymer of less than or equal to 5 kDa.

6. The GHRH analog or salt thereof of any one of claims 1-5, wherein the PEG polymer of less than or equal to 5 kDa is a PEG polymer of about 2 kDa.

7. The GHRH analog or salt thereof of claim 1 or 2, wherein said GHRH analog or salt thereof is:

YaDAIFTAC(Peg-2K)YROVLAQLSAROALQDILSKN-NH_2 (SEQ ID NO: 14);
YaDAIFTAC(Peg-2K)YROVLAQLSAROALQDILS-(Har)-NH_2 (SEQ ID NO:18);
YaDAIFTAC(Peg-2K)YROVLAQLSAROALQDILC(Peg-2K)RN-NH_2 (SEQ ID NO: 20);
YaDAIFTAC(Peg-2K)YROVLAQLSAROALC(Peg-2K)DILSRN-NH_2 (SEQ ID NO: 21);
or
YaDAIFTAC(Peg-2K)YROVLAQLSAROC(Peg-2K)LQDILSRN-NH2 (SEQ ID NO : 22);
wherein a is D-Ala, O is L-Ornithine (Orn), Har is L-Homoarginine and C(Peg-2K) is Cys conjugated to a PEG polymer of about 2 kDa;
or a pharmaceutically acceptable salt thereof.

8. The GHRH analog or salt thereof of claim 7, wherein said GHRH analog or salt thereof is:

YaDAIFTAC(Peg-2K)YROVLAQLSAROALQDILSKN-NH_2 (SEQ ID NO: 14).

9. The GHRH analog or salt thereof of claim 7, wherein said GHRH analog or salt thereof is:

YaDAIFTAC(Peg-2K)YROVLAQLSAROALQDILS-(Har)-NH_2 (SEQ ID NO:18).

10. The GHRH analog or salt thereof of claim 7, wherein said GHRH analog or salt thereof is:

YaDAIFTAC(Peg-2K)YROVLAQLSAROALQDILC(Peg-2K)RN-NH2 (SEQ ID NO: 20).

11. The GHRH analog or salt thereof of claim 7, wherein said GHRH analog or salt thereof is:

YaDAIFTAC(Peg-2K)YROVLAQLSAROALC(Peg-2K)DILSRN-NH_2 (SEQ ID NO: 21).

12. The GHRH analog or salt thereof of claim 7, wherein said GHRH analog or salt thereof is:

YaDAIFTAC(Peg-2K)YROVLAQLSAROC(Peg-2K)LQDILSRN-NH_2 (SEQ ID NO: 22).

13. A pharmaceutical composition comprising the GHRH analog or salt thereof of any one of claims 1 to 12.

14. The pharmaceutical composition of claim 13, further comprising one or more pharmaceutically acceptable carriers, excipients, and/or diluents.

15. Use of the GHRH analog or salt thereof of any one of claims 1 to 12 or of the pharmaceutical composition of claim 13 or 14 for the preparation of a medicament for inducing growth hormone secretion in a subject.

16. Use of the GHRH analog or salt thereof of any one of claims 1 to 12 or the pharmaceutical composition of claim 13 or 14 in the manufacture of a medicament for treating, preventing, or diagnosing a GH-deficient condition in a subject in need thereof.

17. The use of claim 15 or 16, wherein said GHRH analog of salt thereof is to be administered at a daily dose of 0.1 mg to 10 mg.

18. The use of any one of claims 15, 16 and 17, wherein said GHRH analog or salt thereof is to be administered by an intravenous, oral, transdermal, subcutaneous, mucosal, intramuscular, intranasal, or pulmonary route.