

CO-AGONISTS OF THE GLP-1 AND AMYLIN RECEPTORS

TECHNICAL FIELD

5 The invention relates to a compound comprising a GLP-1 receptor agonist and an amylin receptor agonist. The invention also relates to a pharmaceutical formulation, suitable for but not limited to oral administration, which comprises such a compound. The compound and pharmaceutical formulation comprising it may be used for the medical treatment of subjects with overweight or obesity, with or without associated comorbidities; diabetes with
10 or without associated comorbidities; cardiovascular diseases, non-alcoholic steatohepatitis (NASH) and cognitive impairment, such as that caused by Alzheimer's disease.

BACKGROUND

15 Overweight and obesity are the abnormal or excessive accumulation of body fat that present a risk to an individual's overall health. A body mass index (BMI) over 25 is considered overweight, and a BMI of over 30 is considered obese. Obesity is a leading risk factor in a large number of serious conditions, including type 2 diabetes and its associated co-morbidities, and cardiovascular diseases such as heart disease and stroke, which are
20 the leading causes of death worldwide. Obesity is now recognised by the World Health Organization (WHO) as an issue that has grown to epidemic proportion, even in children: in 2016, 1.9 billion adults worldwide were reportedly obese; in 2019, 38.3 million children under the age of 5 worldwide were reportedly obese. According to the WHO, 422 million people worldwide have diabetes and 1.6 million deaths are directly attributed to diabetes
25 each year. There is, therefore, a huge incentive for the individual, as well as society, to try to prevent and/or treat obesity.

 When diet and exercise alone do not suffice in reducing the body mass index (BMI) of an obese individual to an acceptable level, treatment with pharmaceutical drugs such as liraglutide, orlistat and naltrexone–bupropion have been shown to cause some
30 weight loss. Nonetheless, bariatric surgery has proven necessary in many cases. While bariatric surgery is currently the most effective treatment in terms of obtaining long-term weight loss, it is an invasive procedure associated with high risk to the patient and high cost. Therefore, an efficacious and minimally invasive treatment would be a significant improvement in the treatment of obesity.

Amylin is a 37-amino acid long polypeptide hormone that is produced in and co-secreted with insulin from the pancreatic beta (β)-cell. Endogenous amylin has a half-life of approximately 15-20 minutes. It produces its effects in several different organ systems, primarily acting via amylin receptors 1-3 (AMYR1-3). Amylin is an important regulator of energy metabolism in health and disease, inhibiting glucagon secretion, delaying gastric emptying, signalling satiety and suppressing appetite. Other amylin actions have also been reported, such as on the cardiovascular system and on bone.

Clinical studies have shown that amylin receptor agonists may be useful for the treatment of overweight, obesity, type 1 diabetes and/or type 2 diabetes. Currently, there is one product on the market (Symlin®) which contains an amylin receptor agonist (pramlintide acetate) as the active pharmaceutical ingredient. Symlin®, a liquid pharmaceutical formulation for subcutaneous administration, is approved for use in patients with type 1 or type 2 diabetes who use basal and mealtime insulin and have failed to achieve desired glycemic control, despite optimal insulin therapy. Pramlintide for use in overweight and obese patients was also investigated in the clinic. Pramlintide has a short biological half-life (less than 1 hour), necessitating administration thrice daily. Consequently, there is a large diurnal difference in the pramlintide plasma levels of patients treated with it.

GLP-1 is a 30 or 31-amino acid polypeptide hormone that is synthesised and secreted from enteroendocrine L-cells. GLP-1 is an incretin, decreasing blood sugar levels in a glucose-dependent manner by enhancing the secretion of insulin. Endogenous GLP-1 is rapidly degraded, primarily by dipeptidyl peptidase-4 (DPP-4), resulting in a half-life of approximately 2 minutes.

Several marketed products containing a GLP-1 receptor agonist as the active pharmaceutical ingredient are approved for use in individuals with type 2 diabetes. These include dulaglutide (Trulicity®), exenatide (Byetta®, Bydureon®), liraglutide (Victoza®), lixisenatide (Lyxumia®) and semaglutide (Ozempic®).

Semaglutide is the first GLP-1 receptor agonist to be approved in the form of a tablet (Rybelsus®). It is safe and effective as a monotherapy, and as add-on pharmacological therapy, for the treatment of type 2 diabetes mellitus.

Two marketed products containing a GLP-1 receptor agonist as the active pharmaceutical ingredient are approved for use in individuals who are overweight or obese and have at least one weight-related co-morbidity: liraglutide (Saxenda®) and semaglutide (Wegovy®). The maximum efficacy that can be achieved with a GLP-1 receptor agonist is limited by tolerability. At increasing doses, side-effects such as nausea and vomiting

become increasingly pronounced. Amylin receptor agonist therapy is limited by tolerability in much the same way as GLP-1 receptor agonist therapy (and by similar side-effects such as nausea and vomiting). There has been a similar desire to be able to prolong the action of amylin.

5 A fixed-dose combination of an amylin receptor agonist, cagrilintide, and a GLP-1 receptor agonist, semaglutide, is currently under investigation for the treatment of overweight and obesity (Lancet 2021; 397: 1736–48). The drug products being investigated are separate liquid pharmaceutical formulations for subcutaneous use. A clinical trial has demonstrated that a combination of semaglutide and cagrilintide induced a greater weight
10 loss in obese patients than the maximal approved dose of semaglutide monotherapy, without resulting in a significant worsening of the side-effect profile.

 Whilst current therapy options and investigatory drugs provide promise, individuals with overweight, obesity and/or associated comorbidities can, at best, hope to be treated with injectable pharmaceutical formulations or medications with modest efficacy. There still
15 remains a need in the art for a more efficacious medicament, which does not simultaneously result in a proportionally increased level of side-effects and which is suitable for oral administration.

SUMMARY OF THE INVENTION

20 Disclosed herein is a GLP-1 receptor-amylin receptor co-agonist comprising a polypeptide (R1) according to Formula I:

 Z1-Z2-Z3,

 comprising 1-3 lysine (Lys, K) residues and no disulfide bridge; wherein:

- 25 • Z1 is a GLP-1 receptor agonist peptide comprising a maximum of 9 amino acid modifications, relative to SEQ ID NO: 1 (human GLP-1(7-37), with the proviso that Z1 does not comprise an isoleucine (Ile, I) at position 22, relative to SEQ ID NO: 1 or SEQ ID NO: 255;
- Z2 is an optional peptide linker;
- 30 • Z3 is an amylin receptor agonist peptide comprising a C-terminal amide and a maximum of 4 amino acid modifications, relative to SEQ ID NO: 79, with the proviso that Z3 does not comprise a proline (Pro, P) at position 12 (relative to SEQ ID NO: 79 or SEQ ID NO: 256).

 Disclosed herein is a GLP-1 receptor-amylin receptor co-agonist comprising a
35 polypeptide (R1) according to Formula I:

Z1-Z2-Z3,

comprising 1-3 lysine (Lys, K) residues and no disulfide bridge;

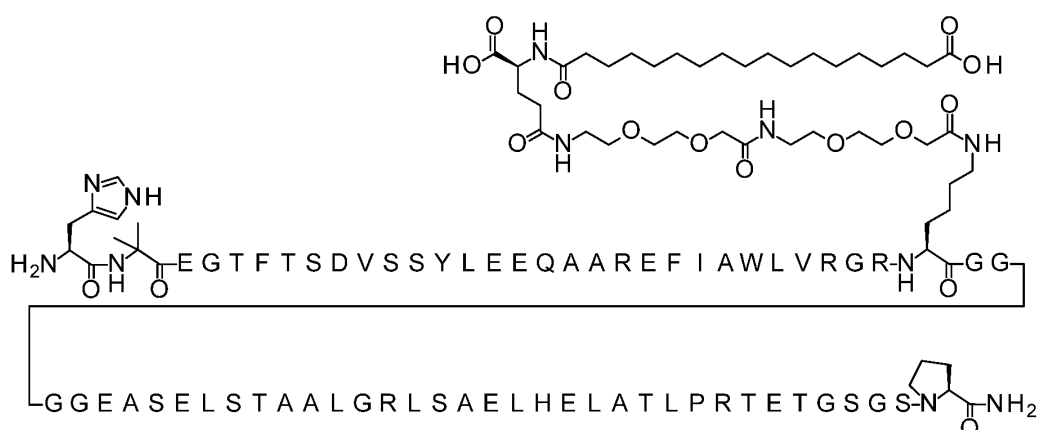
wherein:

- Z1 is a GLP-1 receptor agonist peptide comprising a maximum of 9 amino acid modifications, relative to SEQ ID NO: 1 (human GLP-1(7-37);
- Z2 is an optional peptide linker;
- Z3 is an amylin receptor agonist peptide comprising a C-terminal amide and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79.

Disclosed herein is said GLP-1 receptor-amylin receptor co-agonist, further comprising 1-3 protraction moieties attached via the 1-3 lysine residues.

One preferred compound is "compound 0111", that is:

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVGR-K([2-[2-[2-[[2-[2-[2-[[[4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide (see Fig 46):



Disclosed herein is a GLP-1 receptor-amylin receptor co-agonist for use as a medicament. Disclosed herein is a GLP-1 receptor-amylin receptor co-agonist for use in the treatment of subjects with; an initial body mass index (BMI) of 27 or more, such as 30 or more, optionally in the presence of at least one weight-related comorbidity; diabetes, optionally in the presence of at least one comorbidity, cardiovascular disease, non-steroidal steatohepatitis and/or cognitive impairment, such as that caused by Alzheimer's disease.

Disclosed herein is a pharmaceutical formulation comprising a GLP-1 receptor-amylin receptor co-agonist and pharmaceutically acceptable excipients.

SEQUENCE LISTING

SEQ ID NO: 1 represents the amino acid sequence of human GLP-1(7-37).

5 SEQ ID NOs: 2-78 represent the amino acid sequences of exemplified GLP-1
receptor agonist peptide backbones.

SEQ ID NOs: 79-88 represent the amino acid sequences of exemplified amylin
receptor agonist peptide backbones.

SEQ ID NOs: 89-116 represent the amino acid sequences of exemplified optional
peptide linkers.

10 SEQ ID NOs: 117-236 represent the amino acid sequences of exemplified GLP-1
receptor-amylin receptor co-agonist polypeptide backbones.

SEQ ID NO: 237 represents the amino acid sequence of human glucagon.

SEQ ID NO: 238 represents the amino acid sequence of a GLP-1 receptor agonist
(peptide Z1) according to Formula II.

15 SEQ ID NO: 239 represents the amino acid sequence of optional peptide linker Z2.

SEQ ID NO: 240 represents the amino acid sequence of an amylin receptor
agonist (peptide Z3) according to Formula III.

SEQ ID NOs: 241-245 represent the amino acid sequences of the polypeptide
backbones within exemplified comparator compounds.

20 SEQ ID NO: 246 represents the amino acid sequence of the polypeptide backbone
within semaglutide.

SEQ ID NO: 247 represents the amino acid sequence of the polypeptide backbone
within pramlintide.

25 SEQ ID NO: 248 represents the amino acid sequence of the polypeptide backbone
within cagrilintide.

SEQ ID NO: 249 represents the amino acid sequence of salmon calcitonin.

SEQ ID NO: 250 represents the amino acid sequence of the polypeptide backbone
within compound 1806.

30 SEQ ID NOs: 251-254 represent the amino acid sequences of the polypeptide
backbones within exemplified comparator compounds.

SEQ ID NO: 255 represents the amino acid sequence of a GLP-1 receptor agonist
(peptide Z1) according to Formula II.

SEQ ID NO: 256 represents the amino acid sequence of an amylin receptor
agonist (peptide Z3) according to Formula III.

35

DRAWINGS

Figs. 1-173 depict the manufactured compounds according to the invention that are further described in Example 1.

- Fig. 1 depicts compound 0007.
- 5 Fig. 2 depicts compound 0009.
- Fig. 3 depicts compound 0010.
- Fig. 4 depicts compound 0019.
- Fig. 5 depicts compound 0026.
- Fig. 6 depicts compound 0035.
- 10 Fig. 7 depicts compound 0039.
- Fig. 8 depicts compound 0040.
- Fig. 9 depicts compound 0042.
- Fig. 10 depicts compound 0044.
- Fig. 11 depicts compound 0045.
- 15 Fig. 12 depicts compound 0051.
- Fig. 13 depicts compound 0052.
- Fig. 14 depicts compound 0056.
- Fig. 15 depicts compound 0057.
- Fig. 16 depicts compound 0071.
- 20 Fig. 17 depicts compound 0072.
- Fig. 18 depicts compound 0073.
- Fig. 19 depicts compound 0074.
- Fig. 20 depicts compound 0075.
- Fig. 21 depicts compound 0076.
- 25 Fig. 22 depicts compound 0077.
- Fig. 23 depicts compound 0083.
- Fig. 24 depicts compound 0084.
- Fig. 25 depicts compound 0085.
- Fig. 26 depicts compound 0086.
- 30 Fig. 27 depicts compound 0087.
- Fig. 28 depicts compound 0089.
- Fig. 29 depicts compound 0090.
- Fig. 30 depicts compound 0092.
- Fig. 31 depicts compound 0093.
- 35 Fig. 32 depicts compound 0094.

Fig. 33 depicts compound 0095.
Fig. 34 depicts compound 0096.
Fig. 35 depicts compound 0097.
Fig. 36 depicts compound 0098.
5 Fig. 37 depicts compound 0099.
Fig. 38 depicts compound 0100.
Fig. 39 depicts compound 0101.
Fig. 40 depicts compound 0102.
Fig. 41 depicts compound 0103.
10 Fig. 42 depicts compound 0105.
Fig. 43 depicts compound 0106.
Fig. 44 depicts compound 0109.
Fig. 45 depicts compound 0110.
Fig. 46 depicts compound 0111.
15 Fig. 47 depicts compound 0114.
Fig. 48 depicts compound 0115.
Fig. 49 depicts compound 0116.
Fig. 50 depicts compound 0120.
Fig. 51 depicts compound 0124.
20 Fig. 52 depicts compound 0125.
Fig. 53 depicts compound 0127.
Fig. 54 depicts compound 0128.
Fig. 55 depicts compound 0129.
Fig. 56 depicts compound 0131.
25 Fig. 57 depicts compound 0132.
Fig. 58 depicts compound 0141.
Fig. 59 depicts compound 0142.
Fig. 60 depicts compound 0144.
Fig. 61 depicts compound 0145.
30 Fig. 62 depicts compound 0146.
Fig. 63 depicts compound 0147.
Fig. 64 depicts compound 0151.
Fig. 65 depicts compound 0156.

Fig. 66 depicts compound 0157.
Fig. 67 depicts compound 0159.
Fig. 68 depicts compound 0160.
Fig. 69 depicts compound 0179.
5 Fig. 70 depicts compound 0180.
Fig. 71 depicts compound 0191.
Fig. 72 depicts compound 0202.
Fig. 73 depicts compound 0231.
Fig. 74 depicts compound 0232.
10 Fig. 75 depicts compound 0233.
Fig. 76 depicts compound 0234.
Fig. 77 depicts compound 0235.
Fig. 78 depicts compound 0254.
Fig. 79 depicts compound 0255.
15 Fig. 80 depicts compound 0259.
Fig. 81 depicts compound 0260.
Fig. 82 depicts compound 0261.
Fig. 83 depicts compound 0263.
Fig. 84 depicts compound 0264.
20 Fig. 85 depicts compound 0265.
Fig. 86 depicts compound 0266.
Fig. 87 depicts compound 0267.
Fig. 88 depicts compound 0268.
Fig. 89 depicts compound 0269.
25 Fig. 90 depicts compound 0270.
Fig. 91 depicts compound 0271.
Fig. 92 depicts compound 0272.
Fig. 93 depicts compound 0273.
Fig. 94 depicts compound 0280.
30 Fig. 95 depicts compound 0281.
Fig. 96 depicts compound 0284.
Fig. 97 depicts compound 0285.
Fig. 98 depicts compound 0292.

Fig. 99 depicts compound 0294.
Fig. 100 depicts compound 0295.
Fig. 101 depicts compound 0296.
Fig. 102 depicts compound 0297.
5 Fig. 103 depicts compound 0299.
Fig. 104 depicts compound 0396.
Fig. 105 depicts compound 0397.
Fig. 106 depicts compound 0411.
Fig. 107 depicts compound 0414.
10 Fig. 108 depicts compound 0415.
Fig. 109 depicts compound 0416.
Fig. 110 depicts compound 0417.
Fig. 111 depicts compound 0431.
Fig. 112 depicts compound 0433.
15 Fig. 113 depicts compound 0434.
Fig. 114 depicts compound 0435.
Fig. 115 depicts compound 0436.
Fig. 116 depicts compound 0437.
Fig. 117 depicts compound 0438.
20 Fig. 118 depicts compound 0439.
Fig. 119 depicts compound 0440.
Fig. 120 depicts compound 0472.
Fig. 121 depicts compound 0473.
Fig. 122 depicts compound 0474.
25 Fig. 123 depicts compound 0475.
Fig. 124 depicts compound 0482.
Fig. 125 depicts compound 0483.
Fig. 126 depicts compound 0484.
Fig. 127 depicts compound 0502.
30 Fig. 128 depicts compound 0503.
Fig. 129 depicts compound 0504.
Fig. 130 depicts compound 0506.
Fig. 131 depicts compound 0509.

Fig. 132 depicts compound 0511.
Fig. 133 depicts compound 0512.
Fig. 134 depicts compound 0516.
Fig. 135 depicts compound 0518.
5 Fig. 136 depicts compound 0528.
Fig. 137 depicts compound 0529.
Fig. 138 depicts compound 0539.
Fig. 139 depicts compound 0552.
Fig. 140 depicts compound 0560.
10 Fig. 141 depicts compound 0561.
Fig. 142 depicts compound 0562.
Fig. 143 depicts compound 0564.
Fig. 144 depicts compound 0565.
Fig. 145 depicts compound 0575.
15 Fig. 146 depicts compound 0576.
Fig. 147 depicts compound 0577.
Fig. 148 depicts compound 0578.
Fig. 149 depicts compound 0580.
Fig. 150 depicts compound 0581.
20 Fig. 151 depicts compound 0629.
Fig. 152 depicts compound 0630.
Fig. 153 depicts compound 0631.
Fig. 154 depicts compound 0632.
Fig. 155 depicts compound 0633.
25 Fig. 156 depicts compound 0634.
Fig. 157 depicts compound 0635.
Fig. 158 depicts compound 0636.
Fig. 159 depicts compound 0637.
Fig. 160 depicts compound 0638.
30 Fig. 161 depicts compound 0639.
Fig. 162 depicts compound 0640.
Fig. 163 depicts compound 0648.
Fig. 164 depicts compound 0654.
Fig. 165 depicts compound 0655.
35 Fig. 166 depicts compound 0656.

Fig. 167 depicts compound 0657.

Fig. 168 depicts compound 0658.

Fig. 169 depicts compound 0659.

Fig. 170 depicts compound 0660.

5 Fig. 171 depicts compound 0661.

Fig. 172 depicts compound 0662.

Fig. 173 depicts compound 0663.

Figs. 174-182 depict the manufactured comparator compounds that are further described in Example 1.

10 Fig. 174 depicts comparator compound 0164.

Fig. 175 depicts comparator compound 0185.

Fig. 176 depicts comparator compound 0015.

Fig. 177 depicts comparator compound 0016.

Fig. 178 depicts comparator compound 0668.

15 Fig. 179 depicts comparator compound 0671.

Fig. 180 depicts comparator compound 0672.

Fig. 181 depicts comparator compound 0167.

Fig. 182 depicts comparator compound 0192.

Fig. 183 depicts amylin receptor agonist 1806.

20

DESCRIPTION

The current invention relates to a compound comprising an amylin receptor agonist and a GLP-1 receptor agonist. The compound disclosed herein is capable of activating or
25 “agonising” both the GLP-1 receptor and the amylin receptor system: it is a “GLP-1 receptor-amylin receptor co-agonist”. The compound may further comprise one, two or three protraction moieties.

The compound disclosed herein may be a potent GLP-1 receptor agonist.

The compound disclosed herein may be a potent amylin receptor agonist.

30 The compound disclosed herein may provide a similar level of activation of both receptor systems; that is, it may be “balanced”. Relatively “balanced” receptor activation is advantageous because the relative ratio of the compound’s GLP-1 and amylin receptor agonist portions is locked to the molecule; it is not possible to titrate the two receptor agonists, relative to one another. Ultimately, where a molecule is “balanced”, it may be

dosed such that both hormone systems are activated without side-effects outweighing benefits.

A compound that is highly potent on one receptor and much less potent on the other would be “unbalanced”. For example, a compound that were highly potent on the GLP-1 receptor (for example, having an EC_{50} value of <50 pM) and considerably less potent on the amylin receptor system (for example, having an EC_{50} value of >200 pM) would be “unbalanced”; that is, it would be likely to behave as a GLP-1 receptor agonist only. Such a compound would be unable to achieve optimal efficacy from both hormone systems because side-effects arising from activation of the GLP-1 receptor would prevent administration of a dose sufficiently high to achieve activation of the amylin hormone system as well. The opposite situation might occur if the compound were highly potent on the amylin receptor (for example, having an EC_{50} value of <50 pM) and considerably less potent on the GLP-1 receptor (for example, having an EC_{50} value of >200 pM).

Furthermore, the compounds disclosed herein have a long half-life compared to their native ligands. The compound disclosed herein may possess a long biological half-life, relative to dosing interval, thus reducing the variability in steady state exposure.

The compound disclosed herein may be orally bioavailable, hence suitable for oral administration of subjects in need thereof.

Both the polypeptide backbone and the protraction moiety have been engineered and refined in order to achieve a compound having all of the above properties.

Receptor agonist

A “receptor agonist” or “agonist” is a ligand, such as a compound, that binds to and activates a biological receptor to produce a biological response. A full agonist may be defined as being one that elicits a response of the same magnitude as the natural ligand (see e.g. “Principles of Biochemistry”, AL Lehninger, DL Nelson, MM Cox, Second Edition, Worth Publishers, 1993, page 763). Receptors can be activated by either endogenous agonists, such as endogenous hormones, or exogenous agonists, such as pharmaceutical drugs.

Co-agonist

In the context of the current invention, a “co-agonist” is a compound comprising two different ligands, each of which binds to a given biological receptor to produce a biological response that is characteristic of the natural ligand. The co-agonists disclosed

herein are herein referred to as “GLP-1-amylin co-agonists” or “GLP-1 receptor-amylin receptor co-agonists”.

GLP-1 receptor-amylin receptor co-agonist

5 The compound disclosed herein is a “GLP-1 receptor-amylin receptor co-agonist” or “GLP-1-amylin receptor co-agonist”. The GLP-1 receptor-amylin receptor co-agonist comprises a GLP-1 receptor agonist, an optional peptide linker and an amylin receptor agonist. The GLP-1 receptor agonist component binds to and activates the GLP-1 receptor and the amylin receptor agonist component binds to and activates at least the human
10 amylin 3 receptor (AMYR3).

 The molecular format may be a single chain polypeptide backbone comprising one, two or three lysine (Lys, K) residues. The molecular format may be a single chain polypeptide backbone comprising one, two or three cysteine (Cys, C) residues. The molecular format may be a single chain polypeptide backbone comprising one, two or three
15 lysine (Lys, K) and/or cysteine (Cys, C) residues. The GLP-1 receptor-amylin receptor co-agonist may further comprise 1-3 protraction moieties. A protraction moiety may be covalently bound to said lysine (Lys, K) or cysteine (Cys, C) residue(s).

 The amide moiety at the C-terminus of the amylin receptor agonist must be free in order for the amylin receptor agonist to retain maximal bioactivity. Hence, the C-terminal
20 residue of the GLP-1 receptor agonist is covalently linked to the N-terminal residue of either the optional peptide linker or the amylin receptor agonist and the N-terminal residue of the amylin receptor agonist is covalently linked to the C-terminal of either the optional peptide linker or the GLP-1 receptor agonist. When present, the peptide linker comprises 1-30 naturally occurring amino acids.

25 The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may comprise 1, 2 or 3 lysine residues. The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may comprise 1 or 2 lysine residues. The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may comprise 1 lysine residue.

 The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may
30 comprise 1, 2 or 3 cysteine residues. The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may comprise 1 or 2 cysteine residues. The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may comprise 1 cysteine residue.

 The polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist may
35 herein be referred to as “R1” and described by Formula I:

Z1-Z2-Z3,

wherein Z1 is a GLP-1 receptor agonist peptide, Z2 is an optional peptide linker and Z3 is an amylin receptor agonist peptide.

5 Z1 may comprise a maximum of 9 amino acid modifications, relative to wild type GLP-1 (7-37) (SEQ ID NO: 1). The C-terminus of Z1 is attached either to Z2, when Z2 is present, or to Z3, when Z2 is absent.

Z2 is an optional peptide linker. When present, its N-terminus is attached to the C-terminus of Z1 and its C-terminus is attached to the N-terminus of Z3.

10 Z3 may comprise a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79. The C-terminus of Z3 is modified with an amide group. The N-terminus of Z3 is attached to the C-terminus of Z2, when Z2 is present, or to the C-terminus of Z1, when Z2 is absent.

Z1-Z2-Z3 comprises one, two or three lysine and/or cysteine residues. Each lysine and/or cysteine residue may be covalently bound to a protraction moiety, which may be referred to herein as "R2-R3", wherein "R2" is an optional linker and "R3" is a protractor.

The GLP-1 receptor-amylin receptor co-agonist may exhibit a variety of properties rendering it useful as a medicament, as described herein.

The GLP-1 receptor-amylin receptor co-agonist may be potent on the GLP-1 receptor and on the amylin receptor. The *in vitro* potency of the GLP-1 receptor-amylin receptor co-agonist on the GLP-1 receptor and on the amylin-3 receptor may be measured as described in Assays 1 and 2, respectively. The potency of the compound may be described by means of its EC₅₀ values. EC₅₀ represents the concentration of compound upon which 50% of its maximal effect is observed. The lower the EC₅₀ value, the more potent the compound.

25 When tested as described in Assay 1, the GLP-1 receptor-amylin receptor co-agonist disclosed herein may have an EC₅₀ value of less than 300 pM, such as less than 200 pM, such as less than 150 pM, preferably less than 100 pM, such as less than 75 pM, even more preferably less than 50 pM, such as less than 40 pM, such as less than 30 pM, such as less than 20 pM, such as less than 10 pM.

30 When tested as described in Assay 2, the GLP-1 receptor-amylin receptor co-agonist disclosed herein may have an EC₅₀ value of less than 300 pM, such as less than 200 pM, such as less than 150 pM, preferably less than 100 pM, such as less than 75 pM, preferably less than 50 pM, such as less than 40 pM, such as less than 30 pM, such as less than 20 pM, such as less than 10 pM.

The *in vivo* pharmacology, including half-life, of the GLP-1 receptor-amylin receptor co-agonist described herein may be assessed as described in Examples 4 and 5.

5 The half-life of the GLP-1 receptor-amylin receptor co-agonist in animal subjects may be as long as 125 hours, or longer. The half-life of the GLP-1 receptor-amylin receptor co-agonist in animal subjects may be at least 4 hours. The half-life of the GLP-1 receptor-amylin receptor co-agonist may be more than 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 105, 110, 115, 120 hours. The half-life of the GLP-1 receptor-amylin receptor co-agonist may be 15-60 hours, such as 20-55 hours, such as 25-50 hours.

10 The GLP-1 receptor-amylin receptor co-agonist may be orally bioavailable; that is, present in the bloodstream following per oral administration.

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may reduce food intake in a subject. Administration of the GLP-1 receptor-amylin receptor co-agonist disclosed herein may result in an acute reduction in the intake of food. The *in vivo* effect of
15 the GLP-1 receptor-amylin receptor co-agonist on food intake in rats may be assessed as described in Example 5. Administration of the GLP-1 receptor-amylin receptor co-agonist disclosed herein may result in a food intake in rats, relative to vehicle, which is 0-90%, such as 0-80%, such as 0-70%, such as 0-60%, preferably 0-50%, even more preferably 0-40%, within 0-24 hours after a single subcutaneous injection of 10 nmol/kg of said co-agonist,
20 wherein a food intake of 0% relative to vehicle means that the rat does not eat. Administration of the GLP-1 receptor-amylin receptor co-agonist disclosed herein may result in a food intake in rats, relative to vehicle, which is 0-90%, such as 0-80%, such as 0-70%, such as 0-60%, preferably 0-50%, even more preferably 0-40%, within 24-48 hours after a single subcutaneous injection of 10 nmol/kg of said co-agonist, wherein a food
25 intake of 0% relative to vehicle means that the rat does not eat.

GLP-1

The term "GLP-1" or "native GLP-1" herein refers to human Glucagon-Like Peptide-1 (GLP-1(7-37)), shown in the sequence listing as SEQ ID NO: 1. In the sequence
30 listing, the amino acid residues are consecutively numbered 1-31. Therefore, the first amino acid in wild type human GLP-1(7-37) – that is, the N-terminal histidine – is number "1" in SEQ ID NO: 1.

GLP-1 receptor agonist

The compounds disclosed herein comprise a GLP-1 receptor agonist. A "GLP-1 receptor agonist" may be defined as a ligand which is capable of binding to the GLP-1 receptor and producing a biological response similar to that of the natural ligand, glucagon-like peptide 1 (GLP-1). A "full" GLP-1 receptor agonist may be defined as a GLP-1 receptor agonist which is capable of eliciting a biological response of the same magnitude as GLP-1.

Semaglutide, disclosed in WO2006/097537, Example 4, is an example of an exogenous GLP-1 receptor agonist.

The GLP-1 receptor agonist must have a free N-terminus. Therefore, the compounds disclosed herein comprise a GLP-1 receptor agonist which is attached to either the optional peptide or the amylin receptor agonist at its C-terminus.

The GLP-1 receptor agonist comprises polypeptide "Z1".

The compound disclosed herein may comprise a GLP-1 receptor agonist which is a polypeptide variant of GLP-1(7-37) (SEQ ID NO: 1). The GLP-1 receptor agonist may be a derivative of a polypeptide variant of GLP-1(7-37).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 9 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 9 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 9 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 8 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 8 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 8 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 7 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 7 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 7 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 6 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 6 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 6 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 5 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 5 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 5 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 4 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 4 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 4 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 3 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 3 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 3 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 2 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 2 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 2 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 9 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 9 amino acid modifications relative to wild type human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 9 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 8 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 8 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 8 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 7 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 7 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 7 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 6 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 6 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 6 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 5 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 5 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 5 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 4 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 4 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 4 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 3 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 3 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 3 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

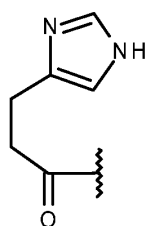
The GLP-1 receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 2 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 2 amino acid modifications relative to human GLP-1 (SEQ ID NO: 1). The GLP-1 receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 2 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1).

The GLP-1 receptor agonist may comprise a phenylalanine residue (Phe, F), tryptophan (Trp, W) or tyrosine (Tyr, Y) residue at position 28 relative to the amino acid sequence and numbering of human GLP-1(7-37). Relative to SEQ ID NO: 1, the numbering is shifted by -6, as shown in Formula II, SEQ ID NO: 238 and SEQ ID NO: 255. Hence, the GLP-1 receptor agonist may comprise a phenylalanine residue (Phe, F), tryptophan (Trp, W) or tyrosine (Tyr, Y) residue at position 22, relative to SEQ ID NO: 1, SEQ ID NO: 238 or SEQ ID NO: 255. The GLP-1 receptor agonist may not comprise an isoleucine (Ile, I) at position 22, relative to SEQ ID NO: 1, SEQ ID NO: 238 or SEQ ID NO: 255.

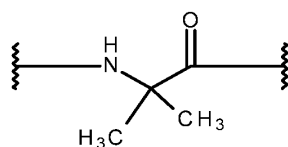
The GLP-1 receptor agonist may comprise an isoleucine (Ile, I) leucine (Leu, L) or valine (Val, V) residue at position 29, relative to the amino acid sequence and numbering of human GLP-1(7-37). Relative to SEQ ID NO: 1, the numbering is shifted by -6, as shown in Formula II and SEQ ID NO: 238. Hence, the GLP-1 receptor agonist may comprise an isoleucine (Ile, I) leucine (Leu, L) or valine (Val, V) residue at position 23, relative to SEQ ID NO: 1, SEQ ID NO: 238 or SEQ ID NO: 255.

The GLP-1 receptor agonist may comprise an Imp (imidazopropionyl or deamino histidine) of formula Chem. 1 at position 7, relative to the amino acid sequence and numbering of human GLP-1(7-37). Relative to SEQ ID NO: 1, the numbering is shifted by -6, as shown in Formula II and SEQ ID NO: 238.

5 Chem. 1:



The GLP-1 receptor agonist may comprise an Aib (alpha (α)-aminoisobutyryl, α-aminoisobutyric acid or α-methylalanine) of formula Chem. 2 at position 8, relative to the relative to the amino acid sequence and numbering of human GLP-1(7-37). Relative to
 10 SEQ ID NO: 1, the numbering is shifted by -6, as shown in Formula II and SEQ ID NO: 238.
 Chem 2:



The GLP-1 receptor agonist may comprise a lysine (Lys, K) residue at any one of
 15 positions 9, 10, 12, 16, 17, 20, 21, 24, 25, 28, 29, 30 or 31, relative to SEQ ID NO: 238 or
 SEQ ID NO: 255.

The GLP-1 receptor agonist disclosed herein may have Formula II, depicted in the sequence listing (SEQ ID NO: 238) as follows:

20 Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-Ser-Xaa12-Tyr-Leu-Glu-Xaa16-
 Xaa17-Ala-Xaa19-Xaa20-Xaa21-Xaa22-Xaa23-Xaa24-Xaa25-Leu-Val-Xaa28-
 Xaa29-Xaa30-Xaa31,

wherein

25 Xaa1 is His (H) or Imp,
 Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),
 Xaa9 is Cys (C), Asp (D) or Lys (K),
 Xaa10 is Cys (C), Lys (K) or Val (V),
 Xaa12 is Cys (C), Lys (K), Arg (R) or Ser (S),

Xaa16 is Cys (C), Glu (E), Gly (G) or Lys (K),
Xaa17 is Cys (C), Gln (Q) or Lys (K),
Xaa19 is Ala (A) or Val (V),
Xaa20 is Cys (C), Lys (K) or Arg (R),
5 Xaa21 is Cys (C), Glu (E) or Lys (K),
Xaa22 is Phe (F), Trp (W) or Tyr (Y),
Xaa23 is Ile (I), Leu (L) or Val (V),
Xaa24 is Ala (A), Cys (C), Glu (E) or Lys (K),
Xaa25 is Cys (C), Lys (K) or Trp (W),
10 Xaa28 is Cys (C), Lys (K) or Arg (R),
Xaa29 is Cys (C), Lys (K) or Gly (G),
Xaa30 is Cys (C), Ala (A), Gly (G), Lys (K), Arg (R) or absent,
Xaa31 is Ala (A), Cys (C), Lys (K), Gly (G), Gln (Q) or absent.

15 The skilled person often refers to the amino acid modifications of a GLP-1 receptor agonist by reference to the standard human, wild-type GLP-1(7-37) nomenclature. In the literature, the first amino acid in GLP-1(7-37) is referred to as being number 7, and subsequent amino acid residues are numbered accordingly, ending with a glycine at number 37.

20 Therefore, the skilled person might number the residues in Formula II as follows, Xaa1 of Formula II corresponding to Xaa7 in GLP-1(7-37), and so on:

Xaa7-Xaa8-Glu-Gly-Thr-Phe-Thr-Ser-Xaa15-Xaa16-Ser-Xaa18-Tyr-Leu-Glu-
Xaa22-Xaa23-Ala-Xaa25-Xaa26-Xaa27-Xaa28-Xaa29-Xaa30-Xaa31-Leu-Val-
25 Xaa34-Xaa35-Xaa36-Xaa37,

wherein:

Xaa7 is His (H) or Imp,
Xaa8 is Aib, Ala (A), Gly (G), Trp (W),
Xaa15 is Cys (C), Asp (D) or Lys (K),
30 Xaa16 is Cys (C), Lys (K) or Val (V),
Xaa18 is Cys (C), Lys (K), Arg (R) or Ser (S),
Xaa22 is Cys (C), Glu (E), Gly (G) or Lys (K),
Xaa23 is Cys (C), Gln (Q) or Lys (K),
Xaa25 is Ala (A) or Val (V),
35 Xaa26 is Cys (C), Lys (K) or Arg (R),

Xaa27 is Cys (C), Glu (E) or Lys (K),
Xaa28 is Phe (F), Trp (W) or Tyr (Y),
Xaa29 is Ile (I), Leu (L) or Val (V),
Xaa30 is Ala (A), Cys (C), Glu (E) or Leu (L),
5 Xaa31 is Cys (C), Lys (K) or Trp (W),
Xaa34 is Cys (C), Lys (K) or Arg (R)
Xaa35 is Cys (C), Lys (K) or Gly (G),
Xaa36 is Cys (C), Ala (A), Gly (G), Lys (K), Arg (R) or absent,
Xaa37 is Ala (A), Cys (C), Lys (K), Gly (G), Gln (Q) or absent.

10

The GLP-1 receptor agonist disclosed herein may have Formula II, depicted in the sequence listing (SEQ ID NO: 255) as follows:

Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-Ser-Xaa12-Tyr-Leu-Glu-Xaa16-
15 Xaa17-Ala-Xaa19-Xaa20-Xaa21-Xaa22-Xaa23-Xaa24-Xaa25-Leu-Val-Xaa28-
Xaa29-Xaa30-Xaa31,

wherein

Xaa1 is His (H) or Imp,
Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),
20 Xaa9 is Asp (D) or Lys (K),
Xaa10 is Lys (K) or Val (V),
Xaa12 is Lys (K), Arg (R) or Ser (S),
Xaa16 is Glu (E), Gly (G) or Lys (K),
Xaa17 is Gln (Q) or Lys (K),
25 Xaa19 is Ala (A) or Val (V),
Xaa20 is Lys (K) or Arg (R),
Xaa21 is Glu (E) or Lys (K),
Xaa22 is Phe (F),
Xaa23 is Ile (I),
30 Xaa24 is Ala (A), Glu (E) or Lys (K),
Xaa25 is Lys (K) or Trp (W),
Xaa28 is Lys (K) or Arg (R),
Xaa29 is Lys (K) or Gly (G),
Xaa30 is Ala (A), Gly (G), Lys (K), Arg (R) or absent,
35 Xaa31 is Ala (A), Lys (K), Gly (G), Gln (Q) or absent.

The GLP-1 receptor agonist peptide (Z1) may be described by reference to sequences in the sequence listing. The GLP-1 receptor co-agonist disclosed herein may comprise a GLP-1 receptor agonist peptide (Z1) which is selected from any one of those depicted in SEQ ID NOs 2-78.

The GLP-1 receptor agonists disclosed herein agonise, or activate, the GLP-1 receptor. This term refers to the ability to bind to the GLP-1 receptor and initiate a signal transduction pathway resulting in insulinotropic action or other physiological effects as is known in the art. The GLP-receptor agonists disclosed herein may be tested for GLP-1 receptor activation as described in Examples 2 (*in vitro*), 4 and 5 (*in vivo*).

The more potent the compound, the lower its EC_{50} value. A compound is considered a highly potent GLP-1 receptor agonist when its EC_{50} value is below approximately 50 pM. A compound is considered to have medium potency when its EC_{50} value is 50-250 pM. A compound is considered to have a poorer potency when its EC_{50} value value is 250-1000 pM. A compound is considered inactive when its EC_{50} value is above 1000 pM.

The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay (see Assay 1) of about 300 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 200 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 150 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 100 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 90 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 80 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 70 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 60 pM or less. Preferably, the GLP-1 receptor agonist has an EC_{50} in a human GLP-1 receptor functional assay of about 50 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 40 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 30 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 25 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 20 pM or less. The GLP-1 receptor agonist may have an EC_{50} in a human GLP-1 receptor functional assay of about 19 pM or less. The GLP-1 receptor agonist may

have an EC₅₀ in a human GLP-1 receptor functional assay of about 18 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 17 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 16 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 15 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 14 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 13 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 12 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 11 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 10 pM or less. The GLP-1 receptor agonist may have an EC₅₀ in a human GLP-1 receptor functional assay of about 5 pM or less.

The GLP-1 receptor agonist may have a similar potency as semaglutide.

Amylin

The term “amylin” herein refers to a polypeptide having the same amino acid sequence as an endogenous amylin, such as human amylin.

Amylin receptor

An amylin receptor agonist may activate or agonise the calcitonin receptor (CTR) and/or the amylin receptors (AMYRs). Amylin receptors consist of heterodimers of two components: the calcitonin receptor (CTR) and one of three receptor activity-modifying proteins (RAMP1–3), resulting in three possible complexes, AMYR1–3. Unless otherwise specified herein, “amylin receptor” at least refers to amylin receptor 3 (AMYR3). Nonetheless, some cross-reactivity can be expected.

Amylin receptor agonists

The compounds disclosed herein comprise an amylin receptor agonist. An “amylin receptor agonist” may be defined as a chemical entity which is capable of binding to the amylin receptor and is capable of activating it. In the context of the current invention, an “amylin receptor agonist” is capable of binding to and activating at least the AMYR3 complex. The amylin receptor agonist may also be capable of agonising the calcitonin receptor and AMYR1-2.

Examples of endogenous amylin receptor agonists are human amylin and human calcitonin. Examples of exogenous amylin receptor agonists are pramlintide and cagrilintide (disclosed in WO2012/168432).

5 The amylin receptor agonist disclosed herein comprises peptide "Z3". The amylin receptor agonist disclosed herein comprises a C-terminal amide, which is essential for bioactivity.

The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues
10 and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79.

The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 6 amino acid modifications, relative to SEQ ID NO: 79.
15 The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 6 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 6 amino acid modifications, relative to SEQ ID NO: 79.

The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 5 amino acid modifications, relative to SEQ ID NO: 79.
20 The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 5 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 5 amino acid modifications, relative to SEQ ID NO: 79.

25 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 4 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 4 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum
30 of 4 amino acid modifications, relative to SEQ ID NO: 79.

The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 3 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 3 amino acid modifications, relative to SEQ ID NO: 79. The amylin

receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 3 amino acid modifications, relative to SEQ ID NO: 79.

The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and a maximum of 2 amino acid modifications, relative to SEQ ID NO: 79.

- 5 The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and a maximum of 2 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 lysine residue and a maximum of 2 amino acid modifications, relative to SEQ ID NO: 79.

- 10 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 lysine residues and 1 amino acid modification, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 lysine residues and 1 amino acid modification, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 lysine residue and 1 modification, relative to SEQ ID NO: 79.

- 15 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79.

- 20 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 6 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 6 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 6 amino acid modifications, relative to SEQ ID NO: 79.

- 25 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 5 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 5 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 5 amino acid modifications, relative to SEQ ID NO: 79.

- 30 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 4 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 4 amino acid modifications, relative to SEQ ID NO: 79. The

amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 4 amino acid modifications, relative to SEQ ID NO: 79.

The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 3 amino acid modifications, relative to SEQ ID NO: 79.

5 The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 3 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 3 amino acid modifications, relative to SEQ ID NO: 79.

10 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and a maximum of 2 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and a maximum of 2 amino acid modifications, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and a maximum of 2 amino acid modifications, relative to SEQ ID NO: 79.

15 The amylin receptor agonist may comprise a polypeptide comprising 0, 1, 2 or 3 cysteine residues and 1 amino acid modification, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 or 2 cysteine residues and 1 amino acid modification, relative to SEQ ID NO: 79. The amylin receptor agonist may comprise a polypeptide comprising 1 cysteine residue and 1 modification, relative to SEQ
20 ID NO: 79.

The amylin receptor agonist may not comprise a proline at position 12, relative to SEQ ID NO: 240 or SEQ ID NO: 256.

The amylin receptor agonist may comprise a lysine (Lys, K) residue at any one of positions 1, 2, 3, 7, 14, 18, 20, 23 or 29 relative to SEQ ID NO: 240 or SEQ ID NO: 256.

25 The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise an amylin receptor agonist according to Formula III (SEQ ID NO: 240):

Xaa1-Xaa2-Xaa3-Leu-Ser-Thr-Xaa7-Ala-Leu-Gly-Arg-Leu-Ser-Xaa14-Glu-Leu-His-
Xaa18-Leu-Xaa20-Thr-Leu-Xaa23-Arg-Thr-Glu-Thr-Gly-Xaa29-Gly-Ser-Xaa32,

30 wherein

Xaa1 is Ala (A), Cys (C), Lys (K) or absent,

Xaa2 is Cys (C), Lys (K) or Ser (S),

Xaa3 is Cys (C), Glu (E), Lys (K) or Arg (R),

Xaa7 is Ala (A), Cys (C), Glu (E) or Lys (K),

35 Xaa14 is Ala (A), Cys (C) or Lys (K),

Xaa18 is Cys (C), Glu (E), Lys (K) or Gln (Q),
Xaa20 is Ala (A), Cys (C) or Lys (K),
Xaa23 is Cys (C), Lys (K) or Pro (P),
Xaa29 is Cys (C), Ser (S) or Lys (K) and
5 Xaa32 is Pro (P) or Tyr (Y).

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise an amylin receptor agonist according to Formula III:

10 Xaa1-Xaa2-Xaa3-Leu-Ser Thr-Xaa7-Ala-Leu-Gly-Arg-Leu-Ser-Xaa14-Glu-Leu-His-
Xaa18-Leu-Xaa20-Thr-Leu-Xaa23-Arg-Thr-Glu-Thr-Gly-Xaa29-Gly-Ser-Xaa32,
wherein
Xaa1 is Ala (A), Lys (K) or absent,
Xaa2 is Lys (K) or Ser (S),
15 Xaa3 is Glu (E), Lys (K) or Arg (R),
Xaa7 is Ala (A), Glu (E) or Lys (K),
Xaa14 is Ala (A) or Lys (K),
Xaa18 is Glu (E), Lys (K) or Gln (Q),
Xaa20 is Ala (A) or Lys (K),
20 Xaa23 is Lys (K) or Pro (P),
Xaa29 is Ser (S) or Lys (K) and
Xaa32 is Pro (P) or Tyr (Y).

The amylin receptor agonist may comprise a polypeptide (Z3) represented by any
25 one of SEQ ID NOs: 79-88. Hence, the polypeptide backbone of the amylin receptor
agonist described herein (Z3) may be described by reference to sequences in the sequence
listing.

The amylin receptor agonists disclosed herein agonise, or activate, the amylin
receptors. The amylin agonists disclosed herein may be tested for amylin activity as
30 described in Examples 2 (*in vitro*), 4 and 5 (*in vivo*).

The more potent the compound, the lower its EC₅₀ value. A compound is
considered a highly potent amylin-receptor agonist when its EC₅₀ value is below 50 pM. A
compound is considered to have medium potency when its EC₅₀ value is 50-250 pM. A
compound is considered to have a poorer potency when its EC₅₀ value is 250-1000 pM. A
35 compound is considered impotent when its EC₅₀ value is above 1000 pM.

The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay (see Assay 2) of about 300 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 250 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 200 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 150 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 100 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 90 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 80 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 70 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 60 pM or less. Preferably, the amylin receptor agonist has an EC_{50} in a human amylin receptor functional assay of about 50 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 40 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 30 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 25 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 20 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 19 pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 18pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 17pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 16pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 15pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 14pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 13pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 12pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 11pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 10pM or less. The amylin receptor agonist may have an EC_{50} in a human amylin receptor functional assay of about 5pM or less.

The amylin receptor agonist disclosed herein may have a similar potency as that of cagrilintide, pramlintide or amylin receptor agonist 1806.

Peptide linkers

The GLP-1 receptor-amylin receptor co-agonist polypeptide backbone disclosed herein, R1, may comprise an optional peptide linker, Z2, which may be represented by Formula IV (SEQ ID NO: 239):

5

Xaa1-Xaa2-Xaa3-Xaa4-Xaa5-Xaa6-Xaa7-Xaa8-Xaa9-Xaa10-Xaa11-Xaa12-
Xaa13-Xaa14-Xaa15-Xaa16-Xaa17-Xaa18-Xaa19-Xaa20-Xaa21-Xaa22-Xaa23-
Xaa24-Xaa25-Xaa26-Xaa27-Xaa28-Xaa29-Xaa30

10 wherein Xaa1-30 is absent or is independently selected from any 7 naturally occurring, or canonical, amino acid residue(s).

Hence, the optional peptide linker may comprise 1-30 canonical amino acid residues. The optional peptide linker may comprise 1-25 canonical amino acid residues. The optional peptide linker may comprise 1-20 canonical amino acid residues, such as 1-
15 15, 1-10 or 1-5 canonical amino acid residues.

In the context of the optional peptide linker, Xaa may be selected from any non-aromatic amino acid residue. Xaa may be a charged amino acid. Xaa may be a polar amino acid. Xaa may be a hydrophobic amino acid.

20 Xaa may be selected from the group consisting of alanine (Ala, A), cysteine (Cys, C), glutamic acid (E), glycine (G), isoleucine (Ile, I), lysine (Lys, K), glutamine (Q), serine (S) and/or proline (Pro, P).

Xaa may be selected from the group consisting of alanine (A), glutamic acid (E), glycine (G), isoleucine (I), lysine (K), glutamine (Q), serine (S) and/or proline (P).

25 The optional peptide linker (Z2) may be any one of the peptide linkers represented by SEQ ID NOs 89-116. The optional peptide linker (Z2) may be any one of the peptide linkers listed in Table 1. The optional peptide linker may be GGGGE.

Table 1: Optional peptide linkers (Z2)

SEQ ID	Peptide linker	SEQ ID	Peptide linker
-	E	100	EGQEPPG
-	GE	101	QEPGQAPE
-	GG	102	QAPGQEPE
-	EA	103	QEPKGQAP

-	EGE	104	GGGGGGGK
-	GGE	105	GGGSGGGE
-	GKE	106	GQEPGQEPE
89	EGGE	107	KQEPGQEPE
90	GGGE	108	GQEKGQEPE
91	GGGG	109	QEPGQAPKE
92	KGGG	110	QEPKGQAPE
93	EAEAE	111	GGGGGGGGE
94	GGGGE	112	GQEPGQEPKE
95	EGEGEE	113	KGQEPGQAPE
96	EGGGGG	114	EGGGGGGGGGE
97	KGGGGE	115	GGGGGGGGGGGGE
98	EGEGEKE	116	GGGGGGGGGGGGGGGGE
99	EGGGGGE		

Polypeptide

The term "polypeptide" or "peptide", as used herein, refers to a compound consisting of a series of amino acid residues that are interconnected by amide (or peptide) bonds.

The polypeptide backbone, of the GLP-1 receptor-amylin receptor co-agonist (R1) disclosed herein, typically comprises 60-85 amino acid residues linked together by peptide bonds. R1 comprises a peptide, Z1-Z2-Z3, which is a GLP-1 receptor agonist (Z1), an optional peptide linker (Z2) and a peptide which is an amylin receptor agonist (Z3).

Amino acids

Amino acids are molecules containing an amine group and a carboxylic acid group, and, optionally, one or more additional groups, often referred to as a side chain.

The term "amino acid" includes canonical amino acids (which are genetically encoded), and unnatural amino acids.

Non-limiting examples of unnatural amino acids are Aib (α -aminoisobutyric acid), deamino histidine (alternative name 3-(imidazol-4-yl)propanoic acid, abbreviated Imp (imidazopropionyl) and the D-isomers of the canonical amino acids.

All amino acid residues within the polypeptide for which the optical isomer is not stated is herein to be understood to mean the L-isomer, unless otherwise specified.

The GLP-1 receptor agonist peptide (Z1) disclosed herein may have a maximum of 9 amino acid modifications, relative to human GLP-1 (SEQ ID NO: 1). The amylin receptor agonist peptide (Z3) disclosed herein may have a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79. Herein, "amino acid modification" refers to the substitution, addition or deletion of an amino acid at a given position in the reference sequence.

Protraction moiety

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may further comprise one, two or three "protraction moieties". A protraction moiety may be represented by the general formula "R2-R3", in which R2 is an optional linker and R3 is a protractor. Each protraction moiety attaches to a lysine or a cysteine residue in the polypeptide backbone (R1) of the compound.

A protraction moiety may consist of one protractor.

A protraction moiety may comprise one linker and one protractor.

A protraction moiety may comprise one linker and two protractors.

When the linker (R2) is present, the protraction moiety attaches to the polypeptide backbone (R1) via R2. When the linker (R2) is absent, R3 attaches to the polypeptide backbone.

The protraction moiety (R2-R3) may attach to a lysine residue in the GLP-1 receptor agonist portion of the polypeptide backbone (the "Z1" in Z1-Z2-Z3). The protraction moiety may attach to a lysine residue in the optional peptide linker portion of the polypeptide backbone (the "Z2" in Z1-Z2-Z3). The protraction moiety may attach to a lysine residue in the amylin receptor agonist portion of the polypeptide backbone (the "Z3" in Z1-Z2-Z3). Where the protraction moiety attaches to a lysine residue via an amide linkage, the GLP-1 receptor-amylin receptor co-agonist is considered to be "acylated".

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise a single lysine residue, to which a single protraction moiety is attached.

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise two lysine residues and two protraction moieties. The compound disclosed herein may comprise two lysine residues and two identical protraction moieties.

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise three lysine residues and three protraction moieties. The compound disclosed herein may comprise three lysine residues and three identical protraction moieties.

The protraction moiety may attach to a cysteine residue in the GLP-1 receptor agonist portion of the polypeptide backbone (the "Z1" in Z1-Z2-Z3). The protraction moiety

may attach to a cysteine residue in the optional peptide linker portion of the polypeptide backbone (the "Z2" in Z1-Z2-Z3). The protraction moiety may attach to a cysteine residue in the amylin receptor agonist portion of the polypeptide backbone (the "Z3" in Z1-Z2-Z3).

Where the protraction moiety attaches to a cysteine residue via a thioether linkage, the
5 GLP-1 receptor-amylin receptor co-agonist is considered to be "alkylated".

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise a single cysteine residue, to which a single protraction moiety is attached.

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise two cysteine residues and two protraction moieties. The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise two cysteine residues and two identical protraction
10 moieties.

The compound disclosed herein may comprise three cysteine residues and three protraction moieties. The compound disclosed herein may comprise three cysteine residues and three identical protraction moieties.

Where the GLP-1 receptor-amylin receptor co-agonist comprises two or three
15 protraction moieties, the protracting moieties are similar, preferably substantially identical, or, most preferably, identical.

In the context of chemical moieties such as the protraction moieties disclosed herein, similarity and/or identity may be determined using any suitable computer program
20 and/or algorithm known in the art.

The skilled person may refer to compounds comprising a protraction moiety as being "derivatives". For example, an "amylin derivative" is understood to be an amylin receptor agonist comprising a protraction moiety.

Herein, the term "protraction moiety" refers to a half-life extending, synthetic moiety
25 comprising a "protractor" (R3) and an optional "side-chain linker" or "linker" (R2). R2 may join R3 to the side chain of a lysine or cysteine residue in R1, the polypeptide backbone of the GLP-1 receptor-amylin receptor co-agonist.

The protraction moiety may be capable of non-covalently binding to albumin, thereby promoting the circulation of the GLP-1 receptor-amylin receptor co-agonist in the
30 blood stream and prolonging its half-life. Thus, the skilled person may also refer to the protraction moiety as being an "albumin binding moiety".

The protractor (R3) may comprise an acyl group. The acyl group may be branched or unbranched. The acyl group may be saturated or unsaturated. The protractor (R3) may comprise a fatty acyl group. The acyl group may be branched or unbranched. The acyl
35 group may be saturated or unsaturated.

The protractor (R3) may comprise a distal carboxylic acid group.

The protractor (R3) may comprise a fatty acid group.

The protractor ("R3") may comprise a fatty acid group and an amide group.

5 The protractor (R3) may comprise a distal carboxylic acid group and an amide group.

The protractor (R3) may comprise an alkyl group.

The protractor (R3) may comprise an aryl group.

The protractor (R3) may comprise a tetrazole group.

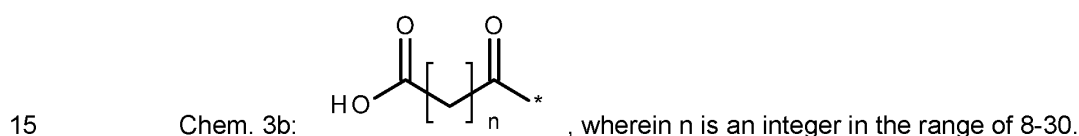
The protractor (R3) may comprise a sulfonic acid group.

10 The protractor (R3) may comprise a phenoxy group.

The protractor (R3) may comprise a benzoic acid group.

The protractor (R3) may comprise a group defined by:

Chem. 3: $\text{HOOC}-(\text{CH}_2)_n-\text{CO}-^*$ wherein n is an integer in the range of 8-30, which may also be referred to as a C(n+2) diacid or as



The protractor (R3) may comprise 8-30 carbon atoms. The protractor may comprise 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29 or 30 carbon atoms.

20 The protractor (R3) may comprise 6-30 consecutive $-\text{CH}_2-$ groups. The protractor (R3) may comprise a carbon chain comprising at least 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29 or 30 consecutive $-\text{CH}_2-$ groups.

The protractor (R3) may comprise 12-26 carbon atoms. The "protractor", or "side chain", may comprise 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 or 26 carbon atoms.

25 The protractor (R3) may comprise 10-26 consecutive $-\text{CH}_2-$ groups. The protractor (R3) may comprise a carbon chain comprising 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 or 26 consecutive $-\text{CH}_2-$ groups.

30 The protractor (R3) may comprise 16-22 carbon atoms. The compound may comprise a single protraction moiety which comprises a side chain comprising 16, 17, 18, 19, 20, 21 or 22 carbon atoms.

The protractor (R3) may comprise 14-20 consecutive $-\text{CH}_2-$ groups. The protractor (R3) may comprise a carbon chain comprising 14, 15, 16, 17, 18, 19 or 20 consecutive $-\text{CH}_2-$ groups.

CH₂- groups.

The protractor (R3) may comprise 16-22 consecutive carbon atoms and 14-20 consecutive –CH₂- groups.

5 The protractor (R3) may comprise 16 consecutive carbon atoms and 14 consecutive –CH₂- groups.

The protractor (R3) may comprise 18 consecutive carbon atoms and 16 consecutive –CH₂- groups.

The protractor (R3) may comprise 20 consecutive carbon atoms and 18 consecutive –CH₂- groups.

10 The protractor (R3) may comprise 22 consecutive carbon atoms and 20 consecutive –CH₂- groups.

The GLP-1 receptor-amylin receptor co-agonist may comprise two protraction moieties, each of which comprises 14, 15, 16, 17, 18, 19 or 20 carbon atoms. The GLP-1 receptor-amylin receptor co-agonist may comprise two protraction moieties, wherein each
15 protractor (R3) comprises 12, 13, 14, 15, 16, 17 or 18 consecutive –CH₂- groups.

The GLP-1 receptor-amylin receptor co-agonist may comprise two C14 diacids, two C16 diacids or two C18 diacids.

The GLP-1 receptor-amylin receptor co-agonist may comprise three protraction moieties, each of which comprises a protractor comprising 12, 13, 14, 15, 16, 17, 18, 19 or
20 20 carbon atoms. The GLP-1 receptor-amylin receptor co-agonist may comprise three protraction moieties, wherein the protractor (R3) comprises 10, 11, 12, 13, 14, 15, 16, 17 or 18 consecutive –CH₂- groups.

The protraction moiety (R2-R3) may be covalently attached to a lysine residue in the polypeptide backbone (R1). The protraction moiety may be attached via an amide bond
25 formed between a carboxylic acid group in the protraction moiety and the epsilon amino group of the lysine residue.

The protraction moiety (R2-R3) may be covalently attached to a cysteine residue in the polypeptide backbone (R1). The protraction moiety may be attached via a thioether bond formed between the protraction moiety and the sulphur atom of the cysteine residue in
30 the polypeptide.

As mentioned above, the compound disclosed herein may comprise one, two or three lysine or cysteine residues and thence one, two or three protraction moieties (R2-R3), wherein each protraction moiety is attached to a side chain of a single lysine or cysteine residue.

The protraction moiety may be attached to the polypeptide backbone described herein (R1) via a lysine (K) residue at any one of positions 9, 10, 12, 16, 17, 20, 21, 24, 25, 28, 29, 30 or 31 of the GLP-1 receptor agonist portion of the polypeptide backbone (Z1), relative to SEQ ID NO: 238 or SEQ ID NO: 255.

- 5 The protraction moiety may be attached to the polypeptide backbone (R1) via a lysine (K) residue in the optional linker (Z2).

 The protraction moiety may be attached to the polypeptide backbone (R1) via a lysine (K) residue at any one of positions 1, 2, 3, 7, 10, 14, 18, 20, 23 or 29 of the amylin receptor agonist portion of the polypeptide backbone (Z3), relative to SEQ ID NO: 240 or
10 SEQ ID NO: 256.

 The protraction moiety may be attached to the polypeptide backbone (R1) via a cysteine (C) residue at any one of positions 9, 10, 12, 16, 17, 20, 21, 24, 25, 28, 29, 30 or 31 of the GLP-1 receptor agonist portion of the polypeptide backbone (Z1), relative to SEQ ID NO: 238.

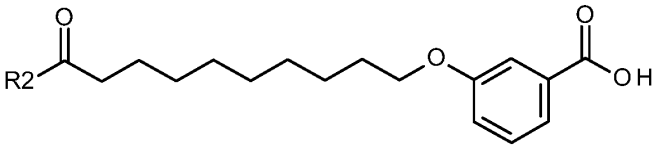
- 15 The protraction moiety may be attached to the polypeptide backbone (R1) via a cysteine (C) residue in the optional linker portion of the polypeptide backbone (Z2).

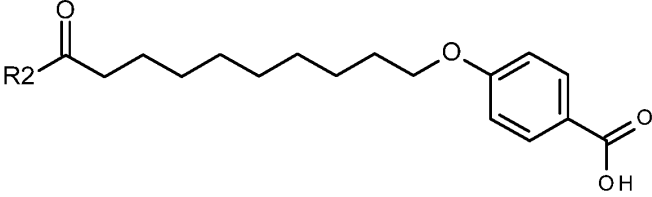
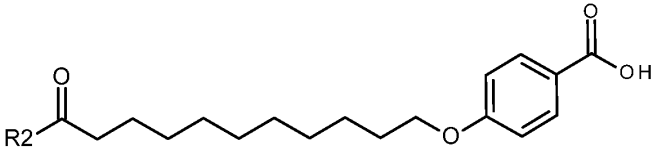
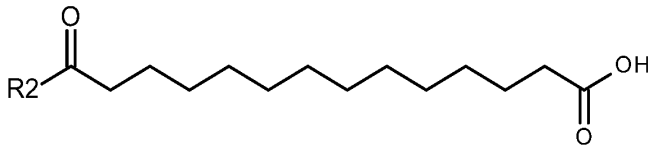
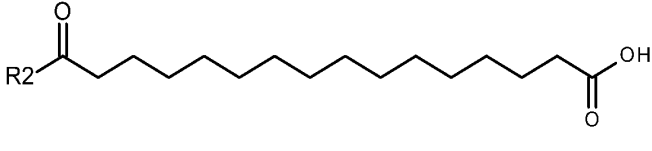
 The protraction moiety may be attached to the polypeptide described herein via a cysteine (C) residue in the amylin receptor agonist portion of the polypeptide backbone (Z3). The cysteine residue may be at any one of positions 1, 2, 3, 7, 10, 14, 18, 20, 23 or
20 29, relative to SEQ ID NO: 240.

 The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise a protractor (R3) which is selected from any one of those depicted in Table 2. In Table 2, R2 represents the optional linker which connects the depicted protractor (R3) to the polypeptide backbone (R1). R1 is not shown in the table.

25

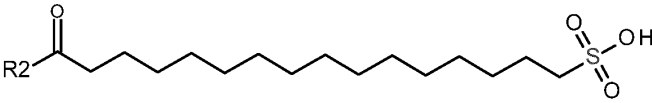
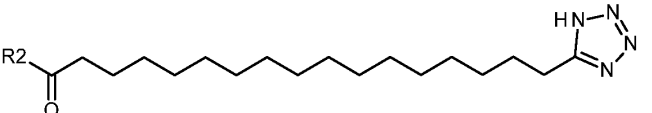
Table 2: Protractors (R3)

<i>Protractor number</i>	<i>Structure</i>	<i>In compounds</i>
1	 3-(9-carboxynonyloxy) benzoic acid	0235 (2x)

2	 4-(9-carboxynonyloxy) benzoic acid	0096 (2x), 0097 (2x), 0098 (2x), 0099 (2x), 0100 (2x), 0101 (2x), 0231 (2x), 0232 (2x), 0233 (2x), 0234 (2x),
3	 4-(10-carboxydecyloxy) benzoic acid	0255 (2x)
4	 C14 diacid	0039 (2x), 0484 (2x), 0635 (3x)
5	 C16 diacid	0560, 0564 (2x), 0630 (2x), 0631 (2x), 0634 (2x), 0072 (2x), 0073 (2x), 0074 (2x), 0075 (2x), 0076 (2x), 0077 (2x), 0086 (2x), 0087 (2x), 0105 (2x), 0259 (2x),

		0260 (2x), 0261 (2x), 0280 (2x), 0281 (2x), 0284 (2x), 0285 (2x), 0292 (2x), 0294 (2x), 0295 (2x), 0296 (2x), 0297 (2x), 0299 (2x), 0396 (2x), 0397 (2x), 0411 (2x), 0475 (2x), 0482 (2x), 0483 (2x), 0509 (2x), 0516 (2x), 0528 (2x), 0575 (2x), 0576 (2x), 0577 (2x), 0578 (2x), 0629 (2x)
6	C18 diacid	0658, 0656, 0007, 0009, 0010, 0019, 0026, 0040, 0042, 0051, 0052, 0083, 0084, 0085, 0092, 0093, 0111, 0114,

		0115, 0132, 0144, 0151, 0156, 0191, 0271, 0273, 0472, 0637, 0639, 0640, 0648, 0044, 0045, 0159, 0661, 0662, 0660, 0131 (2x), 0160 (2x), 0056 (2x), 0057 (2x), 0071 (2x), 0089 (2x), 0090 (2x), 0106 (2x), 0109 (2x), 0116 (2x), 0127 (2x), 0128 (2x), 0129 (2x), 0110 (2x)
7	C20 diacid	0636, 0632, 0633, 0141, 0142, 0659, 0657, 0094, 0157, 0202, 0473, 0474, 0502, 0529, 0562, 0565, 0580, 0581, 0638, 0552, 0663, 0035,

		0095, 0120, 0254, 0263, 0264, 0265, 0266, 0267, 0268, 0269, 0270, 0272, 0414, 0415, 0416, 0417, 0431, 0440, 0503, 0504, 0511, 0512, 0518, 0539, 0561, 0433, 0434, 0435, 0436, 0437, 0438, 0439, 0506, 0179, 0180
8	 C16-sulfonic acid	0654
9	 C17-tetrazole	0102, 0103, 0124, 0125, 0145, 0146, 0147, 0655

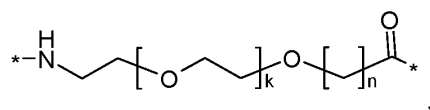
As mentioned above, the protraction moiety ("R2-R3") may comprise an optional side-chain linker, "R2".

The optional side-chain linker (R2) may comprise Ado, Aeep or Aeep, sulfonamide, Trx, ε-Lys, Ahx, Glu, γGlu, Gly, Ser, Ala, Thr and/or a bond.

The optional side-chain linker may comprise at least a moiety which may be represented by the following chemical formula:

Chem 4a: $^*\text{-NH-(CH}_2)_2\text{-(O-(CH}_2)_k\text{-O-(CH}_2)_n\text{-CO-}^*$.

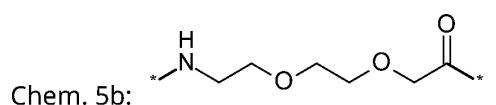
Chem. 4b:



wherein k is an integer in the range of 1-5, and n is an integer in the range of 1-5. When k=1 and n=1, the linker element may be designated Ado, or a 8-amino-3,6-dioxaoctanoyl, which may be represented by the following chemical formula:

Chem. 5a: $^*-\text{NH}-(\text{CH}_2)_2-\text{O}-(\text{CH}_2)_2-\text{O}-\text{CH}_2-\text{CO}-^*$.

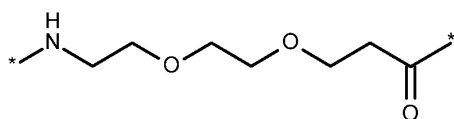
or



- 10 When k=1 and n=2, the linker element may be designated Aeep, which may be represented by the following chemical formula:

Chem. 6: $^*-\text{NH}-(\text{CH}_2)_2-\text{O}-(\text{CH}_2)_2-\text{O}-(\text{CH}_2)_2-\text{CO}-^*$,

or



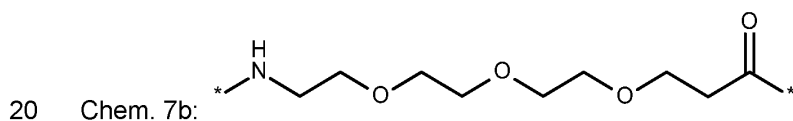
Chem. 6b:

15

When k=2 and n=2, the linker element may be designated Aeeep, which may be represented by the following chemical formula:

Chem. 7a: $^*-\text{NH}-(\text{CH}_2)_2-\text{O}-(\text{CH}_2)_2-\text{O}-(\text{CH}_2)_2-\text{O}-(\text{CH}_2)_2-\text{CO}-^*$,

or

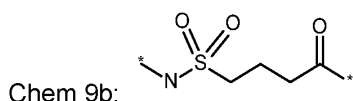


20

Chem. 7b:

The optional side-chain linker may comprise a sulfonamide-C4 moiety. A sulfonamide-C4 group is a sulfonamide group attached to a 4-butanoyl group and having the following chemical formula:

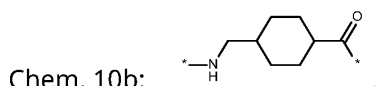
25 Chem 9a: $^*\text{NH}-\text{S}(\text{O})_2-\text{CH}_2-\text{CH}_2-\text{CH}_2-\text{CO}-^*$



Chem 9b:

The optional side-chain linker may comprise Trx. Trx is also referred to as Tranexamic acid, trans-4-(aminomethyl)cyclohexanecarboxylic acid and has the following chemical formula:

- 5 Chem. 10a: $^*\text{-NH-CH}_2\text{-(C}_6\text{H}_{10}\text{)-CO-}^*$ or



Chem. 10b:

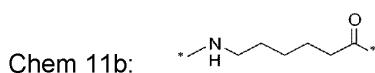
The optional side-chain linker may comprise epsilon-lysine (ϵ -Lys).

The optional side-chain linker may comprise lysine (Lys).

The optional side-chain linker may comprise Ahx. Ahx is also referred to as

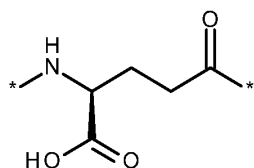
- 10 Aminocaproic acid, 6-aminohexanoic acid and is defined by

Chem 11a: $^*\text{-NH-(CH}_2\text{)}_5\text{-CO-}^*$ or



Chem 11b:

The optional side-chain linker may comprise a Glu di-radical, such as Chem. 12:



15

wherein the Glu di-radical may be included p times, where p is an integer in the range of 1-3.

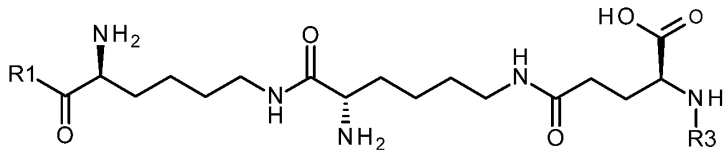
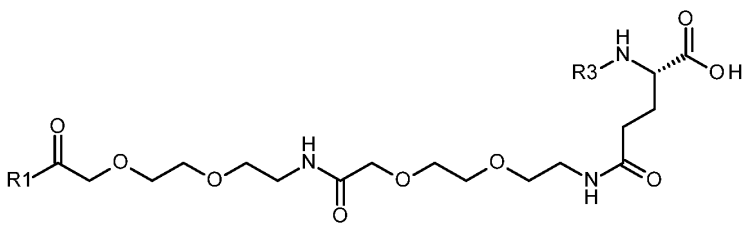
- 20 Chem. 12 may also be referred to as gamma-Glu, or briefly γ Glu, due to the fact that it is the gamma carboxy group of the amino acid glutamic acid which is here used for connection to another linker element, or to the epsilon-amino group of lysine. As explained above, the other linker element may, for example, be another Glu residue, or an Ado molecule. The amino group of Glu in turn forms an amide bond with the carboxy group of the protracting moiety, or with the carboxy group of, e.g., an Ado molecule, if present, or with the gamma-carboxy group of, e.g., another Glu, if present.

- 25 The GLP-1 receptor-amylin receptor co-agonist disclosed herein may comprise a side-chain linker (R2) which is selected from any one of those depicted in Table 3. In Table 3, "R1" represents the polypeptide backbone (Z1-Z2-Z3), R2 (depicted) represents the side-

chain linker and "R3" represents the protractor.)

Table 3: Optional linkers ("R2")

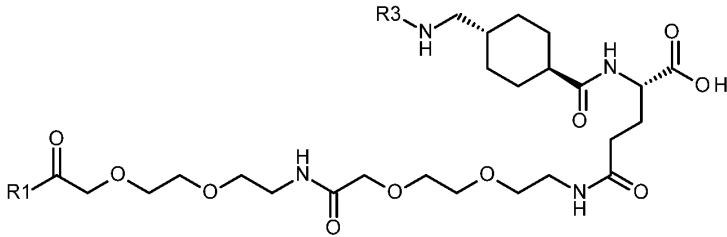
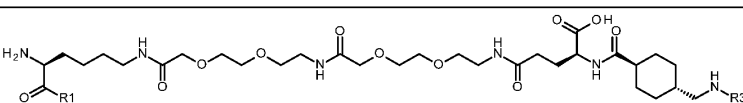
Linker number	Structure	In compounds
1		0564
2		0630 (2x)
3		0636
4		0621 (2x), 0632, 0633, 0634 (2x)
5		0141, 0142, 0131 (2x), 0160 (2x)
6		658, 659

7		656, 657
8		0560, 0007, 0009, 0010, 0019, 0026, 0040, 0042, 0051, 0052, 0083, 0084, 0085, 0092, 0093, 0111, 0114, 0115, 0132, 0144, 0151, 0156, 0191, 0271, 0273, 0472, 0637, 0639, 0640, 0648,

		0094, 0457, 0202, 0473, 0474, 0502, 0529, 0562, 0565, 0580, 0581, 0638, 0654, 0655, 0635 (3x), 0235 (2x), 0096 (2x), 0097 (2x), 0098 (2x), 0099 (2x), 0100 (2x), 0101 (2x), 0231 (2x), 0232 (2x), 0233 (2x), 0234 (2x), 0255 (2x), 0039 (2x), 0484 (2x), 0072 (2x), 0073 (2x), 0074 (2x), 0075 (2x), 0076 (2x), 0077 (2x),
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		0086 (2x), 0087 (2x), 0105 (2x), 0259 (2x), 0260 (2x), 0261 (2x), 0280 (2x), 0281 (2x), 0284 (2x), 0285 (2x), 0292 (2x), 0294 (2x), 0295 (2x), 0296 (2x), 0297 (2x), 0299 (2x), 0396 (2x), 0397 (2x), 0411 (2x), 0475 (2x), 0482 (2x), 0483 (2x), 0509 (2x), 0516 (2x), 0528 (2x), 0575 (2x), 0576 (2x), 0577 (2x), 0578 (2x), 0629 (2x), 0056 (2x), 0057 (2x), 0071 (2x), 0089 (2x), 0090 (2x),
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		0106 (2x), 0109 (2x), 0116 (2x), 0127 (2x), 0128 (2x), 0129 (2x)
9		0044, 0045, 0159, 0552, 0110 (2x)
10		0661
11		0662, 0663
12		0660
13		0102, 0103, 0124, 0125, 0145, 0146, 0147

14		0035, 0095, 0120, 0254, 0263, 0264, 0265, 0266, 0267, 0268, 0269, 0270, 0272, 0414, 0415, 0416, 0417, 0431, 0440, 0503, 0504, 0511, 0512, 0518, 0539, 0561, 0179
15		0433, 0434, 0435, 0436, 0437, 0438, 0439, 0506, 0180

The co-agonists may exist in different stereoisomeric forms having the same molecular formula and sequence of bonded atoms but differing only in the three-dimensional orientation of their atoms in space. The stereoisomerism of the exemplified co-agonists is indicated in the experimental section, in the names as well as the structures, using standard nomenclature. Unless otherwise stated the invention relates to all stereoisomeric forms of the embodied derivative.

Methods of production

The compounds disclosed herein may, for instance, be produced by classical peptide synthesis, e.g. solid phase peptide synthesis using t-Boc or Fmoc chemistry, or other well established techniques, see e.g. Greene and Wuts, "Protective Groups in Organic Synthesis", John Wiley & Sons, 1999; Florencio Zaragoza Dörwald, "Organic Synthesis on Solid Phase", Wiley-VCH Verlag GmbH, 2000; and "Fmoc Solid Phase Peptide Synthesis", Edited by W.C. Chan and P.D. White, Oxford University Press, 2000.

Alternatively, the compounds may be produced by recombinant methods, e.g. by culturing a host cell containing a DNA sequence encoding the peptide sequence and capable of expressing the peptide, in a suitable nutrient medium under conditions permitting the expression of the peptide. Non-limiting examples of host cells suitable for expression of these peptides are *Escherichia coli*, *Saccharomyces cerevisiae* and mammalian BHK or CHO cell lines.

The co-agonists that include non-natural amino acids and/or covalently attached substituents may be produced as described in the experimental part.

Specific examples of methods of preparing a number of the disclosed compounds are included in the examples.

A further aspect of the invention relates to a method for preparing the peptides described herein.

A further aspect of the invention relates to a method for preparing the receptor co-agonists described herein.

In one embodiment, the method for preparing a compound as described herein comprises a step of solid phase peptide synthesis. The substituent may be built sequentially as part of the solid phase peptide synthesis or produced separately and attached via the lysine residue after peptide synthesis.

In one embodiment, the compounds are produced by a two-step process whereby two peptide fragments are ligated after attachment of the substituent to one of the peptide fragments.

Pharmaceutical compositions

Also disclosed herein is a pharmaceutical composition comprising the GLP-1 receptor-amylin receptor co-agonist disclosed herein. Pharmaceutical compositions comprising the GLP-1 receptor-amylin receptor co-agonist, or a pharmaceutically acceptable salt thereof, and one or more pharmaceutically acceptable excipients, may be prepared using methods known to the person skilled in the art.

The term "pharmaceutically acceptable excipient" refers to any ingredient in the pharmaceutical composition which is not the active pharmaceutical ingredient. The excipient may be functional or inert and may serve one or more purposes. For example, the excipient may enhance absorption of the active substance. The excipient might be, amongst others, a buffer, an antimicrobial preservative, an isotonicity agent, a carrier, a vehicle, a filler, a binder, a lubricant, a glidant, a disintegrant, a flow control agent, a crystallization inhibitor, a solubilizer, a stabilizer, a colouring agent, a flavoring agent, a surfactant, an emulsifier. The amount of each excipient used may vary within ranges conventional in the art.

The pharmaceutical composition may be suitable for oral administration. Techniques and excipients which may be used to formulate orally administered pharmaceutical compositions are described in *Handbook of Pharmaceutical Excipients* (e.g. 8th edition, Sheskey et al., Eds., American Pharmaceuticals Association and Pharmaceutical Press, publications department of the Royal Pharmaceutical Society of Great Britain (2017) and later editions); and *Remington: The Science and Practice of Pharmacy* (e.g. 22nd edition, Remington and Allen, Eds., Pharmaceutical Press (2013), and later editions).

The pharmaceutical formulation may be a solid pharmaceutical formulation (e.g. a compressed tablet or capsule) containing the active pharmaceutical ingredient, for example as a freeze-dried or spray-dried composition, and may be used as is, dissolved prior to use, or combined with excipients in the formulation.

The pharmaceutical composition may be a solid formulation containing the compound disclosed herein, a salt of *N*-[8-(2-hydroxybenzoyl)amino]caprylate and one or more further excipients, as is described in the art. For example, the solid formulation may be as described in WO 2012/080471, WO2013/139694, WO 2013/189988, WO 2019/149880, WO2019/215063 or WO2021/219710.

Alternatively, the pharmaceutical composition may be a liquid formulation, such as an aqueous formulation. Such liquid composition may be suitable for oral administration or

for parenteral administration. Liquid compositions that are suitable for injection can be prepared using conventional techniques of the pharmaceutical industry which involve dissolving and mixing the ingredients as appropriate to give the desired end product. Thus, according to one procedure, the compound described herein is dissolved in a suitable buffer at a suitable pH. The composition may be sterilized, for example, by sterile filtration. Techniques and excipients which may be used to prepare liquid formulations are described in *Handbook of Pharmaceutical Excipients* (e.g. 8th edition, Sheskey et al., Eds., American Pharmaceuticals Association and Pharmaceutical Press, publications department of the Royal Pharmaceutical Society of Great Britain (2017) and later editions); and *Remington: The Science and Practice of Pharmacy* (e.g. 22nd edition, Remington and Allen, Eds., Pharmaceutical Press (2013), and later editions).

Pharmaceutical Indications

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may be used as a medicament.

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may be used for the following medical treatments:

(i) prevention and/or treatment of all forms of diabetes, such as hyperglycaemia, type 2 diabetes, impaired glucose tolerance, type 1 diabetes, non-insulin dependent diabetes, MODY (maturity onset diabetes of the young), gestational diabetes, and/or for reduction of HbA1c;

(ii) delaying or preventing diabetic disease progression, such as progression in type 2 diabetes, delaying the progression of impaired glucose tolerance (IGT) to insulin requiring type 2 diabetes, and/or delaying the progression of non-insulin requiring type 2 diabetes to insulin requiring type 2 diabetes;

(iii) prevention and/or treatment of eating disorders, such as obesity, e.g. by decreasing food intake, reducing body weight, suppressing appetite, inducing satiety; treating or preventing binge eating disorder, food cravings, bulimia nervosa and/or obesity induced by administration of an antipsychotic or a steroid; reduction of gastric motility; and/or delaying gastric emptying;

(iv) weight maintenance after successful weight loss (either drug induced or by diet and exercise) – i.e. prevention of weight gain after successful weight loss;

(v) prevention and/or treatment of cardiovascular diseases, such as delaying or reducing development of a major adverse cardiovascular event (MACE) selected from the group consisting of cardiovascular death, non-fatal myocardial infarction, non-fatal stroke,

revascularisation, hospitalisation for unstable angina pectoris, and hospitalisation for heart failure.

(vi) prevention and/or treatment of non-alcoholic steatohepatitis (NASH);

(vii) prevention and/or treatment of cognitive impairment, such as that caused by

5 Alzheimer's disease.

In some embodiments the indication is (i). In some embodiments the indication is (ii). In a still further particular aspect the indication is (iii). In some embodiments the indication is (iv). In some embodiments the indication is (v). In some embodiments the indication is (vi). In some embodiments the indication is (vii). In some embodiments the indication is type 2 diabetes and/or obesity.

10 The term "treatment", as used herein, refers to the medical therapy of any human or other vertebrate subject in need thereof. Said subject is expected to have undergone physical examination by a medical practitioner, or a veterinary medical practitioner, who has given a tentative or definitive diagnosis which would indicate that the use of said specific treatment is beneficial to the health of said human or other vertebrate. The timing and purpose of said treatment may vary from one individual to another, according to the status quo of the subject's health. Thus, said treatment may be prophylactic (preventative), palliative, symptomatic and/or curative.

15 In some embodiments the indication is (i) and (iii). In some embodiments the indication is (ii) and (iii).

In some embodiments the subject suffering from obesity is human, such as an adult human or a paediatric human (including infants, children, and adolescents).

20 Body mass index (BMI) is a measure of body fat based on height and weight. The formula for calculation is $BMI = \text{weight in kilograms} / \text{height in meters}^2$. A human subject suffering from obesity may have a BMI of ≥ 30 ; this subject may also be referred to as being obese. In some embodiments the human subject suffering from obesity may have a BMI of ≥ 35 or a BMI in the range of ≥ 30 to < 40 . In some embodiments the obesity is severe obesity or morbid obesity, wherein the human subject may have a BMI of ≥ 40 .

25 In some embodiments the invention relates to a method for treatment or prevention of overweight, optionally in the presence of at least one weight-related comorbidity. In some embodiments the invention relates to use of the formulation for treatment or prevention of overweight, optionally in the presence of at least one weight-related comorbidity. In some embodiments the subject suffering from overweight is human, such as an adult human or a paediatric human (including infants, children, and adolescents). In some embodiments a human subject suffering from overweight may have a BMI of ≥ 25 , such as a BMI of ≥ 27 ,

such as a BMI of ≥ 30 , such as a BMI of ≥ 35 or a BMI of ≥ 40 . In some embodiments a human subject suffering from overweight has a BMI in the range of 25 to <30 or in the range of 27 to <30 . In some embodiments the weight-related comorbidity is selected from the group consisting of hypertension, diabetes (such as type 2 diabetes), dyslipidaemia, high cholesterol and obstructive sleep apnoea.

The term "reduction of body weight" may include treatment or prevention of obesity and/or overweight.

Administration of the compound disclosed herein may be as an adjunct to a reduced-calorie diet and increased physical activity for chronic weight management in adult or paediatric patients with an initial body mass index (BMI) of 30 kg/m² or greater (obesity) or 27 kg/m² or greater (overweight) in the presence of at least one weight-related comorbidity (e.g. hypertension, type 2 diabetes mellitus, or dyslipidemia).

Dosing frequency

The GLP-1 receptor-amylin receptor co-agonist disclosed herein may be administered approximately once daily, such as once every 12-36 hours, such as once every 18-30 hours, such as approximately once every 24 hours.

The GLP-1 amylin receptor co-agonist disclosed herein may be administered approximately once every other day, such as once every 36-60 hours, such as once every 42-54 hours, such as approximately once every 48 hours.

The GLP-1 amylin receptor co-agonist disclosed herein may be administered approximately twice daily, such as once every 6-18 hours, such as once every 9-15 hours, such as approximately once every 12 hours.

Following is a non-limiting list of embodiments of the present invention.

EMBODIMENTS

1. A GLP-1 receptor-amylin receptor co-agonist comprising a polypeptide (R1) according to Formula I:

Z1-Z2-Z3,

comprising 1-3 lysine (Lys, K) residues and, optionally, no disulfide bridge; wherein:

- Z1 is a GLP-1 receptor agonist peptide comprising a maximum of 9 amino acid modifications, relative to SEQ ID NO: 1 (human GLP-1(7-37));
- Z2 is an optional peptide linker;

- Z3 is an amylin receptor agonist peptide comprising a C-terminal amide and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79.
2. A GLP-1 receptor-amylin receptor co-agonist comprising a polypeptide (R1)
5 according to Formula I:
Z1-Z2-Z3,
comprising 1-3 lysine (Lys, K) and/or cysteine (Cys, C) residues and no disulfide bridge;
wherein:
- Z1 is a GLP-1 receptor agonist peptide comprising a maximum of 9 amino acid
10 modifications, relative to SEQ ID NO: 1 (human GLP-1(7-37));
 - Z2 is an optional peptide linker;
 - Z3 is an amylin receptor agonist peptide comprising a C-terminal amide and a maximum of 7 amino acid modifications, relative to SEQ ID NO: 79.
- 15 3. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, further comprising 1-3 protraction moieties (R2-R3), attached to the polypeptide (R1) via said 1-3 lysine residues.
4. The GLP-1 receptor-amylin receptor co-agonist according to the preceding
20 embodiment, comprising 1-3 lysine residues.
5. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising 1-2 lysine residues.
- 25 6. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising 1 lysine residue.
7. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising 1-3 cysteine residues.
30
8. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising 1-2 cysteine residues.
9. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
35 preceding embodiments, comprising 1 cysteine residue.

10. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 0-3 lysine (Lys, K) residues at any one, or combination of, positions 9, 10, 12, 16, 17, 20, 21,
5 24, 25, 28, 29, 30 and/or 31, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

11. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 2 lysine (Lys, K) residues at positions:
10 • 12 and either 21, 30 or 31, or
• 21 and either 30 or 31,
relative to SEQ ID NO: 238 or SEQ ID NO: 255.

12. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 2 lysine (Lys, K) residues at positions:
15 • 12 and either 21 or 31, or
• 21 and 31,
relative to SEQ ID NO: 238 or SEQ ID NO: 255.

20 13. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 1 lysine residue (Lys, K) at any one of positions 12, 17, 20, 21, 28, 30 or 31.

25 14. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 1 lysine residue (Lys, K) at position 31.

15. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the peptide linker (Z2) comprises 0-3 lysine (K) residues.
30

16. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the peptide linker (Z2) comprises 1-2 lysine (Lys, K) residues.
35

17. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the peptide linker (Z2) comprises 1 lysine (Lys, K) residue.
- 5 18. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the peptide linker (Z2) comprises 0-3 cysteine (Cys, C) residues.
- 10 19. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the peptide linker (Z2) comprises 1-2 cysteine (Cys, C) residues.
- 15 20. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the peptide linker (Z2) comprises 1 cysteine (Cys, C) residue.
- 20 21. The GLP-1 receptor-amylin receptor agonist according to any one of the preceding claims, wherein the amylin receptor agonist peptide (Z3) comprises 0-3 lysine (Lys, K) residues.
22. The GLP-1 receptor-amylin receptor agonist according to any one of the preceding claims, wherein the amylin receptor agonist peptide (Z3) comprises 0-3 cysteine (Cys, C) residues.
- 25 23. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the amylin receptor agonist peptide (Z3) comprises 1-3 lysine (Lys, K) residues at any one of positions 1, 2, 3, 7, 14, 18, 20, 23 and/or 29, relative to SEQ ID NO: 240 or SEQ ID NO: 256.
- 30 24. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the amylin receptor agonist peptide (Z3) comprises 1 lysine (Lys, K) residue at any one of positions 14, 18, 20, 23, 29; relative to SEQ ID NO: 240 or SEQ ID NO: 256.

25. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the amylin receptor agonist peptide (Z3) comprises 1 lysine (Lys, K) residue at position 18, relative to SEQ ID NO: 240 or SEQ ID NO: 256.

- 5 26. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising a GLP-1 receptor agonist (Z1) according to Formula II (SEQ ID NO 238):

10 Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-Ser-Xaa12-Tyr-Leu-Glu-Xaa16-
Xaa17-Ala-Xaa19-Xaa20-Xaa21-Xaa22-Xaa23-Xaa24-Xaa25-Leu-Val-Xaa28-
Xaa29-Xaa30-Xaa31,

and wherein

Xaa1 is His (H) or Imp,
Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),
15 Xaa9 is Cys (C), Asp (D) or Lys (K),
Xaa10 is Cys (C), Lys (K) or Val (V),
Xaa12 is Cys (C), Lys (K), Arg (R) or Ser (S),
Xaa16 is Cys (C), Glu (E), Gly (G) or Lys (K),
Xaa17 is Cys (C), Gln (Q) or Lys (K),
20 Xaa19 is Ala (A) or Val (V),
Xaa20 is Cys (C), Lys (K) or Arg (R),
Xaa21 is Cys (C), Glu (E) or Lys (K),
Xaa22 is Phe (F), Trp (W) or Tyr (Y),
Xaa23 is Ile (I), Leu (L) or Val (V),
25 Xaa24 is Ala (A), Cys (C), Glu (E) or Lys (K),
Xaa25 is Cys (C), Lys (K) or Trp (W),
Xaa28 is Cys (C), Lys (K) or Arg (R),
Xaa29 is Cys (C), Lys (K) or Gly (G),
Xaa30 is Ala (A), Cys (C), Gly (G), Lys (K), Arg (R) or absent,
30 Xaa31 is Ala (A), Cys (C), Lys (K), Gly (G), Gln (Q) or absent.

27. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising a GLP-1 receptor agonist (Z1) according to Formula II (SEQ ID NO: 255):

Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-Ser-Xaa12-Tyr-Leu-Glu-Xaa16-
Xaa17-Ala-Xaa19-Xaa20-Xaa21-Xaa22-Xaa23-Xaa24-Xaa25-Leu-Val-Xaa28-
Xaa29-Xaa30-Xaa31,

and wherein

- 5 Xaa1 is His (H) or Imp,
 Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),
 Xaa9 is Asp (D) or Lys (K),
 Xaa10 is Lys (K) or Val (V),
 Xaa12 is Lys (K), Arg (R) or Ser (S),
10 Xaa16 is Glu (E), Gly (G) or Lys (K),
 Xaa17 is Gln (Q) or Lys (K),
 Xaa19 is Ala (A) or Val (V),
 Xaa20 is Lys (K) or Arg (R),
 Xaa21 is Glu (E) or Lys (K),
15 Xaa22 is Phe (F), Trp (W) or Tyr (Y),
 Xaa23 is Ile (I), Leu (L) or Val (V),
 Xaa24 is Ala (A), Glu (E) or Lys (K),
 Xaa25 is Lys (K) or Trp (W),
 Xaa28 is Lys (K) or Arg (R),
20 Xaa29 is Lys (K) or Gly (G),
 Xaa30 is Ala (A), Gly (G), Lys (K), Arg (R) or absent,
 Xaa31 is Ala (A), Lys (K), Gly (G), Gln (Q) or absent.

28. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
25 preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) does not
comprise an isoleucine (Ile, I) at position 22, relative to SEQ ID NO: 238 or SEQ ID NO:
255.

29. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
30 preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises a
phenylalanine (Phe, F), a tryptophan (W) or a tyrosine (Tyr, Y) at position 22, relative to
SEQ ID NO: 238 or SEQ ID NO: 255.

30. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises a phenylalanine (Phe, F) at position 22, relative to SEQ ID NO: 238 or SEQ ID NO: 255.
- 5 31. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises an isoleucine (Ile, I), a leucine (Leu, L) or a valine (Val, V) at position 23, relative to SEQ ID NO: 238 or SEQ ID NO: 255.
- 10 32. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises an isoleucine (Ile, I) at position 23, relative to SEQ ID NO: 238 or SEQ ID NO: 255.
33. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising a GLP-1 receptor agonist (Z1) according to Formula II (SEQ ID NO: 255):
- 15 Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-Ser-Xaa12-Tyr-Leu-Glu-Xaa16-Xaa17-Ala-Xaa19-Xaa20-Xaa21-Phe-Ile-Xaa24-Xaa25-Leu-Val-Xaa28-Xaa29-Xaa30-Xaa31,
- 20 wherein
- Xaa1 is His (H) or Imp,
- Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),
- Xaa9 is Asp (D) or Lys (K),
- Xaa10 is Lys (K) or Val (V),
- 25 Xaa12 is Lys (K), Arg (R) or Ser (S),
- Xaa16 is Glu (E), Gly (G) or Lys (K),
- Xaa17 is Gln (Q) or Lys (K)
- Xaa19 is Ala (A) or Val (V),
- Xaa20 is Lys (K) or Arg (R),
- 30 Xaa21 is Glu (E) or Lys (K),
- Xaa24 is Ala (A), Glu (E) or Lys (K),
- Xaa25 is Lys (K) or Trp (W),
- Xaa28 is Lys (K) or Arg (R)
- Xaa29 is Lys (K) or Gly (G),
- 35 Xaa30 is Ala (A), Gly (G), Lys (K), Arg (R) or absent,

Xaa31 is Ala (A), Lys (K), Gly (G), Gln (Q) or absent.

34. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments,

5 wherein Z1 is

Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-S-Xaa12-Tyr-Leu-Glu-Xaa16-
Xaa17-Ala-Xaa19-Xaa20-Xaa21-Phe-Ile-Xaa24-Xaa25-Leu-Val-Xaa28-Xaa29-
Xaa30-Xaa31,

and wherein

- 10 Xaa1 is His (H) or Imp,
Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),
Xaa9 is Cys (C) or Asp (D),
Xaa10 is Cys (C) or Val (V),
Xaa12 is Cys (C), Arg (R) or Ser (S),
15 Xaa16 is Cys (C), Glu (E) or Gly (G)
Xaa17 is Cys (C) or Gln (Q),
Xaa19 is Ala (A) or Val (V),
Xaa20 is Cys (C) or Arg (R),
Xaa21 is Cys (C) or Glu (E),
20 Xaa24 is Ala (A), Cys (C) or Glu (E)
Xaa25 is Cys (C) or Trp (W),
Xaa28 is Cys (C) or Arg (R),
Xaa29 is Cys (C) or Gly (G),
Xaa30 is Cys (C), Ala (A), Gly (G), Arg (R) or absent,
25 Xaa31 is Ala (A), Cys (C), Gly (G), Gln (Q) or absent.

35. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the amylin receptor agonist peptide (Z3) does not comprise a proline (Pro, P) at position 12, relative to SEQ ID NO: 240 or SEQ ID NO: 256.

- 30 36. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises a leucine (Leu, L) at position 12, relative to SEQ ID NO: 240 or SEQ ID NO: 256.

37. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises a leucine at position 12 (Leu12), relative to SEQ ID NO: 240 or SEQ ID NO: 256.

5 38. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein optional linker Z2 is absent or comprises 1-30, 1-25 or 1-20 naturally occurring amino acid residues (SEQ ID NO 239).

39. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
10 preceding embodiments, wherein optional linker Z2 comprises 0-20 amino acid residues selected from the group consisting of Ala (A), Glu (E), Gly (G), Lys (K), Pro (P) and Gln (Q).

40. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein optional linker Z2 is any one of those listed in Table 1.
15

41. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising an amylin receptor agonist peptide (Z3) according to Formula III (SEQ ID NO 240):

20 Xaa1-Xaa2-Xaa3-Leu-Ser Thr-Xaa7-Ala-Leu-Gly-Arg-Leu-Ser-Xaa14-Glu-Leu-His-
Xaa18-Leu-Xaa20-Thr-Leu-Xaa23-Arg-Thr-Glu-Thr-Gly-Xaa29-Gly-Ser-Xaa32,
wherein

Xaa1 is Ala (A), Cys (C), Lys (K) or is absent,
Xaa2 is Cys (C), Lys (K) or Ser (S),
Xaa3 is Cys (C), Glu (E), Lys (K) or Arg (R),
25 Xaa7 is Ala (A), Cys (C), Glu (E) or Lys (K),
Xaa14 is Ala (A), Cys (C) or Lys (K),
Xaa18 is Cys (C), Glu (E), Lys (K) or Gln (Q),
Xaa20 is Ala (A), Cys (C) or Lys (K),
Xaa23 is Cys (C), Lys (K) or Pro (P),
30 Xaa29 is Cys (C), Ser (S) or Lys (K) and
Xaa32 is Pro (P) or Tyr (Y).

42. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising an amylin receptor agonist (Z3) according to Formula
35 III (SEQ ID NO 256):

Xaa₁-Xaa₂-Xaa₃-Leu-Ser-Thr-Xaa₇-Ala-Leu-Gly-Arg-Leu-Ser-Xaa₁₄-Glu-Leu-His-
Xaa₁₈-Leu-Xaa₂₀-Thr-Leu-Xaa₂₃-Arg-Thr-Glu-Thr-Gly-Xaa₂₉-Gly-Ser-Xaa₃₂,

wherein

- 5 Xaa1 is Ala (A) or Lys (K) or is absent,
Xaa2 is Lys (K) or Ser (S),
Xaa3 is Glu (E), Lys (K) or Arg (R),
Xaa7 is Ala (A), Glu (E) or Lys (K),
Xaa14 is Ala (A) or Lys (K),
Xaa18 is Glu (E), Lys (K) or Gln (Q),
10 Xaa20 is Ala (A) or Lys (K),
Xaa23 is Lys (K) or Pro (P),
Xaa29 is Ser (S) or Lys (K) and
Xaa32 is Pro (P) or Tyr (Y).
- 15 43. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 2, 3,
4, 5, 6, 7, 8 or 9 amino acid modifications relative to human GLP-1(7-37).
- 20 44. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises 1 or 2
substitutions of a naturally occurring amino acid residue with 1 or 2 unnatural amino acids.
- 25 45. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises an
imidazole propionyl (Imp) at position 7, relative to human GLP-1(7-37), or at position 1,
relative to SEQ ID NO: 238 or SEQ ID NO: 255.
- 30 46. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises an alpha-
aminoisobutyryl (Aib) at position 8, relative to human GLP-1(7-37), or at position 2, relative
to SEQ ID NO: 238 or SEQ ID NO: 255.
47. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the GLP-1 receptor agonist peptide (Z1) comprises an

alanine (Ala, A) at any one of positions 8, 25, 30 or 37, relative to human GLP-1(7-37), or at any one of positions 2, 19, 24 or 31, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

48. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises an arginine residue (Arg, R) at any one of positions 26 or 34, relative to human GLP-1(7-37), or at any one of positions 18 or 26, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

49. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises an aspartic acid residue (Asp, D) at position 15, relative to human GLP-1(7-37), or at position 9, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

50. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises a glutamic acid residue (Glu, E) at any one of positions 22, 27 or 30, relative to human GLP-1(7-37), or at any one of positions 16, 21 or 24, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

51. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises a glycine residue (Gly, G) at any one of positions 8 or 36, relative to human GLP-1(7-37), or at any one of positions 2 or 30, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

52. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises a histidine residue (His, H) at position 7, relative to human GLP-1(7-37), or at position 1, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

53. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises a serine residue (Ser, S) at position 18, relative to human GLP-1(7-37), or at positions 12, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

54. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises a tryptophan

residue (Trp, W) at any one of positions 8 or 31, relative to human GLP-1(7-37), or at any one of positions 2 or 25, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

55. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
5 preceding embodiments, wherein the GLP-1 receptor agonist (Z1) comprises a valine residue (Val, V) at any one of positions 16 or 25, relative to human GLP-1(7-37), or at any one of positions 10 or 19, relative to SEQ ID NO: 238 or SEQ ID NO: 255.

56. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
10 preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises 0, 1, 2, 3 or 4 amino acid modifications, relative to SEQ ID NO: 79.

57. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises 0, 1, 2
15 or 3 amino acid modifications, relative to SEQ ID NO: 79.

58. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises 0, 1
or 2 amino acid modifications, relative to SEQ ID NO: 79.
20

59. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises 0 or 1
amino acid modifications, relative to SEQ ID NO: 79.

25 60. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises a
lysine (Lys, K) residue at any one of positions 1, 2, 3, 7, 10, 14, 18, 20, 23 or 29 relative to
SEQ ID NO: 240 or SEQ ID NO: 256.

30 61. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises a
lysine (Lys, K) residue at any one of positions 2, 14, 18, 20, 23 or 29 relative to SEQ ID NO:
240 or SEQ ID NO: 256.

62. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises an arginine (Arg, R) residue at position 3, relative to SEQ ID NO: 240 or SEQ ID NO: 256.
- 5 63. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises a Glu (E) residue at position 7, relative to SEQ ID NO: 240 or SEQ ID NO: 256.
64. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
10 preceding embodiments, wherein the amylin receptor agonist peptide (Z3) comprises a Leu (L) at position 12, relative to SEQ ID NO 79.
65. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein Z1 is a GLP-1 receptor agonist selected from the group
15 consisting of SEQ ID NOs 2-78.
66. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein Z2 is any amino acid residue or peptide presented in Table 1.
20
67. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein Z3 is an amylin receptor agonist peptide (Z3) selected from the group consisting of SEQ ID NOs 79-88.
- 25 68. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, further comprising a protraction moiety attached to each lysine residue.
69. The GLP-1 receptor-amylin receptor co-agonist according to any one of the
30 preceding embodiments, wherein each lysine residue is acylated.
70. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein said protraction moiety comprises a C14-C20 diacid.

71. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the protraction moiety comprises a C14 diacid, a C16 diacid, a C18 diacid, a C20 diacid, a C18-tetrazole, a C14-sulfonic acid, 4-(9-carboxynonyloxy) benzoic acid, 4-(10-carboxydecyloxy) benzoic acid or 3-(9-carboxynonyloxy) benzoic acid.
72. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising two C14 diacids, C16 diacids or C18 diacids.
73. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, comprising three C14 diacids.
74. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments, wherein the lysine is attached to any one of the optional linkers ("R2") presented in Table 3.
75. A GLP-1 receptor-amylin receptor co-agonist comprising a polypeptide selected from any one of SEQ ID NOs 117-236.
76. A GLP-1 receptor-amylin receptor co-agonist selected from any one of those identified in Example 1a.
77. A pharmaceutically acceptable salt of the GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments.
78. A pharmaceutical formulation comprising a GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding embodiments.
79. The pharmaceutical formulation according to the preceding embodiment, which is for oral administration.
80. The pharmaceutical formulation according to the preceding embodiment, which is a solid pharmaceutical formulation.

81. The solid pharmaceutical formulation according to the preceding embodiment, which is a tablet.
82. The tablet according to the preceding embodiment, comprising sodium N-(8-(2-hydroxybenzoyl)amino)caprylate and magnesium stearate.
83. The tablet according to the preceding embodiment, comprising 75-600 mg sodium N-(8-(2-hydroxybenzoyl)amino)caprylate and 7-8.5 mg magnesium stearate.
84. The pharmaceutical formulation according to any one of embodiments 78-83, which is for dosing approximately once daily, such as once every 12-36 hours, such as once every 18-30 hours, such as approximately once every 24 hours.
85. A GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84 for use as a medicament.
86. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use in the treatment of subjects with an initial body mass index (BMI) of 27 or more, such as 30 or more.
87. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use in the treatment of subjects with an initial body mass index (BMI) of 27 or more and at least one weight-related co-morbidity.
88. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use as an adjunct to a reduced-calorie diet and increased physical activity for chronic weight management in adult subjects with an initial body mass index (BMI) of 30 kg/m² or greater (obesity) or 27 kg/m² or greater (overweight) in the presence of at least one weight-related co-morbidity.

89. Use according to the preceding embodiments, wherein said co-morbidity is diabetes and/or a cardiovascular disease.

5 90. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use in the treatment of subjects with diabetes, such as type II diabetes.

10 91. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use in the treatment and/or prevention of cardiovascular disease.

15 92. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use in the treatment of NASH.

20 93. The GLP-1 receptor-amylin receptor co-agonist according to any one of embodiments 1-77 or the formulation according to any one of embodiments 78-84, for use in the treatment and/or prevention of cognitive impairment, such as that caused by Alzheimer's disease.

EXAMPLES

25 **Example 1: Synthesis of GLP-1 receptor-amylin receptor co-agonists and comparator compounds**

This example provides the identity, materials for making and methods of synthesis of many compounds according to the current invention.

30 Also provided are the identity, materials for making and methods of synthesis of the comparator compounds described herein.

Materials and Methods

List of Abbreviations

35 The following abbreviations are used in the following, in alphabetical order:

- Ac: acetyl
Aib: alpha-aminoisobutyric acid
AUC: Area Under the Curve
Boc: *t*-butyloxycarbonyl
5 DCM: dichloromethane
DIC: diisopropylcarbodiimide
DIPEA: *N,N*-diisopropylethylamine or Hünig's base
DMF: dimethyl formamide
DODT: 3,6-dioxa-1,8-octanedithiol
10 DTT: dithiothreitol
EDTA: ethylenediaminetetraacetic acid
ELISA: Enzyme Linked Immuno Sorbent Assay
Fmoc: 9-fluorenylmethyloxycarbonyl
HFIP: 1,1,1,3,3,3-hexafluoro-2-propanol or hexafluoroisopropanol
15 HOBt: 1-hydroxybenzotriazole
HPLC: High Performance Liquid Chromatography
HSA: Human Serum Albumin
Imp: imidazolepropionyl
i.v.: intravenously
20 LCMS or LC-MS: Liquid Chromatography Mass Spectroscopy
MeCN: acetonitrile
Mtt: 4-methyltrityl
NHS: N-hydroxysuccinimide
NMP: N-methyl pyrrolidone
25 Oxyma Pure®: cyano-hydroxyimino-acetic acid ethyl ester
PK: pharmacokinetic
QTof: Quantitative Time of Flight
s.c.: subcutaneously
SD: Standard Deviation
30 SEC-HPLC: Size Exclusion High Performance Liquid Chromatography
SEM: Standard Error of Mean
tBu: *t*-butyl
TFA: trifluoroacetic acid
TIS: triisopropylsilane
35 Trt: triphenylmethyl or trityl

Trx: tranexamic acid

UPLC: Ultra Performance Liquid Chromatography

General Methods of Preparation

5 This section relates to general methods for solid phase peptide synthesis (SPPS) methods, including methods for cleaving the peptide from the resin and removal of the protecting groups and for its purification) Included as well are LCMS methods for detecting and characterising the resulting peptide.

Resins used for the preparation of C-terminal peptide amides were PAL Amide AM
10 resin (loading e.g. 0.6 mmol/g) or H-Rink Amide-ChemMatrix resin (loading e.g. 0.5 mmol/g) or Rink Amide AM polystyrene resin (loading e.g. 0.3-0.7 mmol/g) or Tentagel® rink amide resin (loading e.g. 0.2-0.3 mmol/g) or similar resins suitable for SPPS .

The Fmoc-protected amino acid derivatives used comprised amongst others the standard recommended: Fmoc-Ala-OH, Fmoc-Arg(Pbf)-OH, Fmoc-Asn(Trt)-OH, Fmoc-
15 Asp(OtBu)-OH, Fmoc-Cys(Trt)-OH, Fmoc-Gln(Trt)-OH, Fmoc-Glu(OtBu)-OH, Fmoc-Gly-OH, Fmoc-His(Trt)-OH, Fmoc-Ile-OH, Fmoc-Leu-OH, Fmoc-Lys(Boc)-OH, Fmoc-Met-OH, Fmoc-Phe-OH, Fmoc-Pro-OH, Fmoc-Ser(tBu)-OH, Fmoc-Thr(tBu)-OH, Fmoc-Trp(Boc)-OH or Fmoc-Trp-OH, Fmoc-Tyr(tBu)-OH, Fmoc-Val-OH, Fmoc-Lys(Mtt)-OH, Fmoc-Aib-OH, etc. supplied from e.g. AAPPTEC, Anaspec, Bachem, ChemImpex, Iris Biotech, Midwest
20 Biotech, Gyros Protein Technologies or Novabiochem. Other building blocks such as Fmoc-pseudoprolines and similar derivatives were employed in some difficult sequences. Where nothing else is specified the natural L-form of the amino acids are used. The N-terminal amino acid was Boc protected at the alpha amino group, either by using a reagent with the Boc group pre-installed (e.g. Boc-His(Trt)-OH for peptides with His at the N-terminus) or by
25 exchanging the N-terminal Fmoc protective group for the Boc protective group after installation of the amino acid at the peptide N-terminus. Fmoc-Lys(Mtt)-OH and similar derivatives, such as, but not limited to Fmoc-Lys(ivDDE) etc. bearing an orthogonal protecting group at the epsilon amino position, were used in the cases where side chain instalment was relevant. The epsilon amino derivative would be liberated using a suitable
30 orthogonal deprotecting agent, such as HFIP in DCM, and the side chain installed either stepwise with SPPS or by coupling the epsilon amino functionality directly to the activated ester (e.g. NHS ester) sidechain building block.

In case of stepwise side chain attachment using SPPS, the following suitably protected building blocks such as, but not limited to, Fmoc-8-amino-3,6-dioxaoctanoic acid
35 (Fmoc-OEG-OH), Fmoc-tranexamic acid (Fmoc-Trx-OH), Fmoc-Glu-OtBu, octadecanedioic

acid mono-*tert*-butyl ester, nonadecanedioic acid mono-*tert*-butyl ester, eicosanedioic acid mono-*tert*-butyl ester, hexadecanedioic acid mono-*tert*-butyl ester, tetradecanedioic acid mono-*tert*-butyl ester, or 4-(9-carboxynonyloxy) benzoic acid *tert*-butyl ester were used. All operations stated below were performed within a 50-450 μ mol synthesis scale range.

5

SPPS of the peptidyl backbone:

SPPS was performed using Fmoc based chemistry on a SymphonyX Solid Phase Peptide Synthesizer from Protein Technologies (Tucson, AZ 85714 U.S.A.). Fmoc-deprotection was achieved with 20% piperidine in DMF, containing between 0 and 0.2 M Oxyma. Peptide couplings were performed using DIC/Oxyma Pure®. Amino acid/Oxyma Pure® solutions (0.3 M/0.3 M in DMF at a molar excess of 3-12 fold) were added to the resin followed by the same molar equivalent of DIC (as a 0.6-1.5M solution in DMF) and collidine (1.5M in DMF). The step-wise assembly was done using the following steps: 1) pre-swelling of resin with DMF; 2) Fmoc-deprotection by the use of 20% piperidine in DMF containing between 0 and 0.2M Oxyma Pure® for 1-5 treatments of 5-30 min each; 3) washes with DMF to remove traces of piperidine; 4) coupling of Fmoc-amino acid with 3-12 eq. of Fmoc-amino acid as a 0.3M solution in 0.3M Oxyma Pure® in DMF mixed with an equimolar volume of DIC and collidine for 1-12 hours. In the case of sterically hindered amino acids, this coupling step was repeated once or twice; 5) washes with DMF to remove excess reagents; 6) final wash with DCM at the completion of the assembly which made the resin ready for attachment of a modifying group on lysine side chain.

Alternatively, SPPS was performed using Fmoc based chemistry on a SymphonyX Solid Phase Peptide Synthesizer from Protein Technologies (Tucson, AZ 85714 U.S.A.). Fmoc-deprotection was achieved with 20% piperidine in DMF, containing between 0 and 0.2 M Oxyma. Peptide couplings were performed using DIC/Oxyma Pure®. Amino acid/Oxyma Pure® solutions (0.3 M/0.3 M in DMF at a molar excess of 3-12 fold) were added to the resin followed by the same molar equivalent of DIC (as a 0.6M solution in DMF). The step-wise assembly was done using the following steps: 1) pre-swelling of resin with DMF; 2) Fmoc-deprotection by the use of 20% piperidine in DMF containing between 0 and 0.2M Oxyma Pure® for 1-5 treatments of 5-30 min each; 3) washes with DMF to remove traces of piperidine; 4) coupling of Fmoc-amino acid with 3-12 eq. of Fmoc-amino acid as a 0.3M solution in 0.3M Oxyma Pure® in DMF mixed with an equimolar volume of DIC for 1-12 hours. In the case of sterically hindered amino acids, this coupling step was repeated once or twice; 5) washes with DMF to remove excess reagents; 6) final wash with

DCM at the completion of the assembly which made the resin ready for attachment of a modifying group on lysine side chain.

Alternatively, the protected peptidyl resin was synthesized according to the Fmoc strategy on a Prelude solid phase peptide synthesiser (Protein Technologies, Tucson, USA) using the manufacturer supplied machine protocols. Coupling was done by the use of DIC (dicyclohexylcarbodiimide) and Oxyma Pure (ethyl 2-cyano-2-(hydroxyimino)-acetate, Merck, Novabiochem, Switzerland) mediated couplings in DMF. The coupling of the Fmoc-amino acid was done as described above using 4-8 time excess of amino acid relative to resin substitution (4-8 eq). Coupling time ranged from 1 hour up to 6 hours. The Fmoc-Arg(pbf)-OH was coupled using a double coupling procedure (1 hour + 1hour). The step-wise solid phase assembly on the Prelude was done using the following steps: 1) deprotection (removal of Fmoc) by the use of 20-25 % piperidine in DMF for 2x4-10 min. , step 2) Wash (removal of piperidine) with DMF and DCM, step 3) Coupling of Fmoc-amino acid (0.3M Fmoc-amino acid in 0.3M Oxyma Pure in DMF) 3-10 eq excess for 1-4 hours coupling initiated by adding 1/10 volume of 3M DIC in DMF and 1/10 volume collidine in DMF. Mixing was done by occasional bubbling with nitrogen, step 4) Wash (removal of excess amino acid and reagents by the use of DMF and DCM). The last step included washing with DCM which made the resin ready for attachment of a modifying group on lysine side chain.

20

Attachment of side chains to resin bound protected peptide backbone:

The N-epsilon-lysine Mtt protection group was removed by washing the resin with a suitable orthogonally deprotection mixture such as, but not limited to, 20-30% HFIP in DCM, containing 0-5% TIS in multiple cycles (e.g. 1 x 5 min and 2 x 20 min) before washing with piperidine, DMF and DCM.

Acylation was performed either manually, or on a solid phase peptide synthesizer, such as, but not limited to the SymphonyX Solid Phase Peptide Synthesizer from Protein Technologies (Tucson, AZ 85714 U.S.A.) as described in this method's section "SPPS of the peptidyl backbone" using stepwise addition of building blocks such as, but not limited to, Fmoc-8-amino-3,6-dioxaoctanoic acid (Fmoc-OEG-OH), Fmoc-Glu-OtBu, Fmoc-tranexamic acid (Fmoc-Trx-OH). Introduction of the fatty acid moiety was achieved using a suitable building block, such as, but not limited to, octadecanedioic acid mono-*tert*-butyl ester, eicosanedioic acid mono-*tert*-butyl ester, hexadecanedioic acid mono-*tert*-butyl ester, tetradecanedioic acid mono-*tert*-butyl ester, 4-(9-carboxynonyloxy) benzoic acid *tert*-butyl ester.

35

Alternatively the side chain moiety could be installed in 1 step, directly after removal of the Mtt group, as described in the first part of this section, by using the a suitable side chain building block equipped with an activated ester, e.g. NHS.

5 *Cleavage of resin bound peptide and purification*

After synthesis the resin was washed with DCM, and the peptidyl resin subject to treatment with TFA mixtures in presence of 1-20 volume % scavenger agents such as, but not limited to, water, TIS, DTT and DODT. The cleavage reactions would typically be carried out at room temperature and a duration of 1-3 hours. Alternatively, the cleavage could be carried out at elevated temperatures (e.g. 50 degrees Celsius), for shorter times (15-60 minutes). The cleavage reaction was followed by precipitation of the crude peptide using cold (e.g. 5 degrees Celsius) diethyl ether. The precipitate was washed with diethylether and dissolved in a suitable mixture of water and MeCN. Optionally, suitable water-miscible co-solvents such as acetic acid were used.

15 The crude peptide solution was purified by reversed-phase preparative HPLC (Waters Deltaprep 4000) on a column containing C18-silica gel. Elution was performed with an increasing gradient of MeCN in water containing 0.1 % TFA. Relevant fractions were analysed by analytical UPLC. Fractions containing the pure target peptide were pooled and freeze dried.

20 Alternatively, or when further purification was necessary, the crude peptide or lyophilized peptide TFA salt isolated as described above was dissolved in a neutral (e.g. pH 7-8) aqueous buffer based on common buffer salts such as, but not limited to, sodium hydrogen phosphate or ammonium bicarbonate and purified with reversed-phase preparative HPLC (Waters Deltaprep 4000) on a column containing C18-silica gel. Elution was performed with an increasing gradient of MeCN in aqueous buffer (such as, but not limited to, sodium phosphate (5-100 mM, pH 7-8, or 2-40 g/l ammonium bicarbonate). Relevant fractions were analysed by analytical UPLC. Fractions containing the pure target peptide were pooled and acidified with TFA until pH 2 and diluted with water until the total concentration of MeCN was <20%. Optionally, the resulting mixture was degassed by

30 filtration using vacuum. The solution was then purified by reversed-phase preparative HPLC (Waters Deltaprep 4000) on a column containing C18-silica gel. Elution was performed with an increasing gradient of MeCN in water containing 0.1 % TFA. Relevant fractions were analysed by analytical UPLC. Fractions containing the pure target peptide were pooled and freeze dried

35

LCMS characterization methods:

LCMS34:

LCMS34 was performed on a set up consisting of Waters Acquity UPLC H Class system and Waters Xevo G2-XS QToF. Eluents: A: 0.1% formic acid in MQ water; B: 0.1% formic acid in MeCN.

The analysis was performed at RT (column temp 40 °C) by injecting an appropriate volume of the sample onto the column which was eluted with a gradient of A and B. The UPLC conditions, detector settings, and mass spectrometer settings were: Column: Waters Acquity BEH, C-18, 1.7µm, 2.1mm x 50mm. Gradient: Linear 5% - 95% B during 4.0 min at 0.4 ml/min. Detection: MS resolution mode, ionisation method: ES. Scan: 50-4000 amu.

LCMS36:

LCMS36 was performed on a set up consisting of Waters Acquity UPLC H Class system and Waters Xevo G2-XS QToF. Eluents: A: 0.1% formic acid in MQ water; B: 0.1% formic acid in MeCN.

The analysis was performed at RT (column temp 60C) by injecting an appropriate volume of the sample onto the column which was eluted with a gradient of A and B. The UPLC conditions, detector settings, and mass spectrometer settings were: Column: Phenomenex Aeris , C-4, 3.6 µm widepore, 2.1mm x 50mm. Gradient: stepwise over 8 minutes; 5% - 25% B during 1.0 min, 25% - 65% B, during 6 minutes, 65-95% B during 0.5 minutes, 95% B isocratic for 0.5 minutes, at 0.4 ml/min. Detection: MS resolution mode, ionisation method: ES. Scan: 50-4000 amu.

LCMS_ZQ:

LCMS_ZQ was performed on a LCMS instrument consisting of Waters Acquity UPLC system coupled with Waters Acquity TUV detector and Waters Micromass ZQ 2000 detector. Eluents: A – 0.05% TFA in MQ-water, B – 0.05% TFA in acetonitrile.

The analysis was performed at RT (column temperature 40 °C) by injecting appropriate volume (0.2 – 10 µl) of the sample onto the column which was eluted with a gradient of A and B. Column: Waters Acquity UPLC BEH, C-18, 1.7µm, 2.1mm x 50mm. Gradient run time: Linear 5-95% B over 4.5 min, then 95% B for 0.5 min, 95-5% B for 0.5 min, 5% B for 0.5 min at a flow rate of 0.45 ml/min. Detection: Ionization method: Electron spray positive, Scanning range: 200 – 2048 (m/z), Capillary voltage: 3.0 kV, Cone Voltage: 20 V, Scan time: 0.9 s, Interscan delay: 0.1 s, Detection method: quadrupole.

LCMS01:

LCMS_01 was performed on a setup consisting of Waters Acquity UPLC system and LCT Premier XE mass spectrometer from Micromass. Eluents: A: 0.1% Formic acid in MQ water; B: 0.1% Formic acid in MeCN. The analysis was performed at RT (column temp

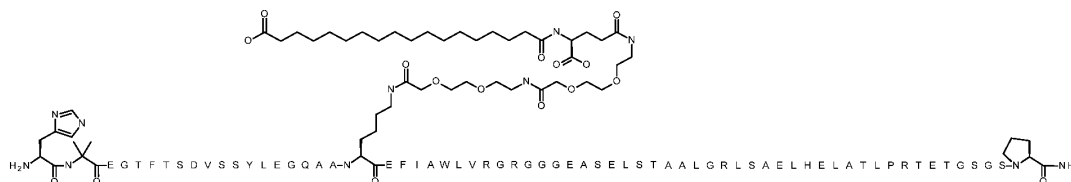
- 5 40C) by injecting an appropriate volume of the sample onto the column which was eluted with a gradient of A and B. The UPLC conditions, detector settings and mass spectrometer settings were: Column: Waters Acquity UPLC BEH, C-18, 1.7 μ m, 2.1mm x 50mm. Gradient: Linear 5% - 95% B during 4.0 min at 0.4ml/min. Detection: 214 nm (analogue output from TUV (Tunable UV detector)) MS ionisation mode: API-ES. Scan: 500-2000
- 10 atomic mass units (amu).

Manufactured compounds

Example 1a: Compounds according to the invention

Compound 0007

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-EFIAWLVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₃₂ H₅₃₁ N₈₉ O₁₁₂

Molecular weight (average) calculated: 7561.3 g/mol

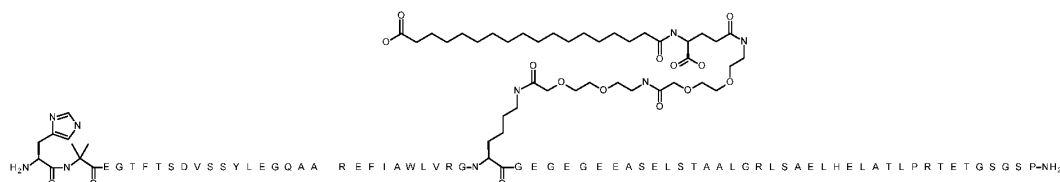
mono isotopic mass: 7556.9 g/mol

LCMS34: found (M + 5H)⁵⁺ 1513.3 (most abundant)

- 25 The amino acid sequence of
HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 117

Compound 0009

H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRG-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GEGEGEEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



5 C₃₄₇ H₅₅₂ N₉₂ O₁₂₁

Molecular weight (average) calculated: 7948.6 g/mol

mono isotopic mass: 7944.0 g/mol

LCMS34: found (M + 5H)⁵⁺ 1591 (most abundant)

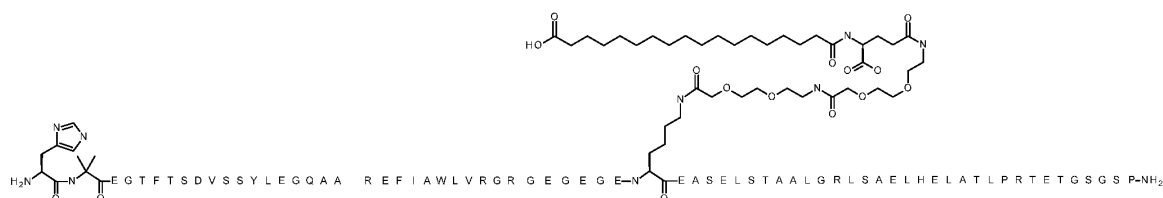
The amino acid sequence of

10 HXEGTFTSDVSSYLEGQAAREFIAWLVRGKGE GEGEGEEASELSTAALGRLSAELHELATLP
RTETGSGSP has SEQ ID NO: 118

Compound 0010

H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRGRGEGEGE-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-

15 carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₃ H₅₆₄ N₉₆ O₁₂₂

20 Molecular weight (average) calculated: 8104.8 g/mol

mono isotopic mass: 8100.1 g/mol

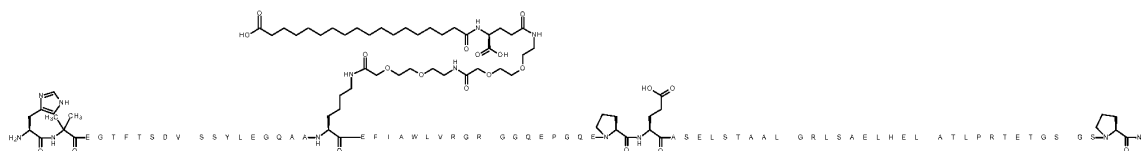
LCMS34: found (M + 5H)⁵⁺ 1622 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEGQAAREFIAWLVRGRGEGEGEKEASELSTAALGRLSAELHELATL
25 PRTETGSGSP has SEQ ID NO: 119

Compound 0019

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFI A W L V R G R G G Q E P G Q E P E A S E L S T A A L G R L S A E L H E L A T L P R T E T G S G S P - a m i d e



5 C₃₆₂ H₅₇₅ N₉₇ O₁₂₄

Molecular weight (average) calculated: 8270.0144 g/mol

mono isotopic mass: 8265.1670 g/mol

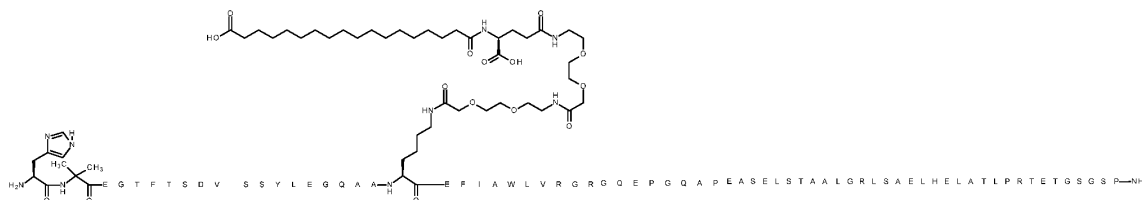
LCMS36: found (M + 5H)⁵⁺ 1654.85 (most abundant)

The amino acid sequence of

10 HXEGTFTSDVSSYLEGQAAKEFI A W L V R G R G G Q E P G Q E P E A S E L S T A A L G R L S A E L H E L A T L P R T E T G S G S P has SEQ ID NO: 120

Compound 0026

15 H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFI A W L V R G R G G Q E P G Q A P E A S E L S T A A L G R L S A E L H E L A T L P R T E T G S G S P - a m i d e



C₃₅₈ H₅₇₀ N₉₆ O₁₂₁

Molecular weight (average) calculated: 8154.9270 g/mol

20 mono isotopic mass: 8150.1400 g/mol

LCMS34: found (M + 5H)⁵⁺ 1631.04 (most abundant)

The amino acid sequence of

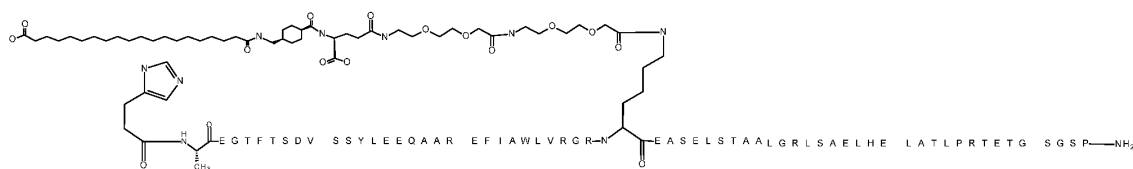
HXEGTFTSDVSSYLEGQAAKEFI A W L V R G R G G Q E P G Q A P E A S E L S T A A L G R L S A E L H E L A T L P R T E T G S G S P has SEQ ID NO: 121

25

Compound 0035

Imp-AEGTFTSDVSSYLEEQAAREFI A W L V R G R -K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[(19-

carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-EASELSTAALGRLSAELHELATLPRTETGSGSP-amide



5 C₃₄₄ H₅₅₂ N₉₀ O₁₁₃

Molecular weight (average) calculated: 7756.5989 g/mol

mono isotopic mass: 7752.0214 g/mol

LCMS01: found (M + 5H)⁵⁺ 1552 (most abundant)

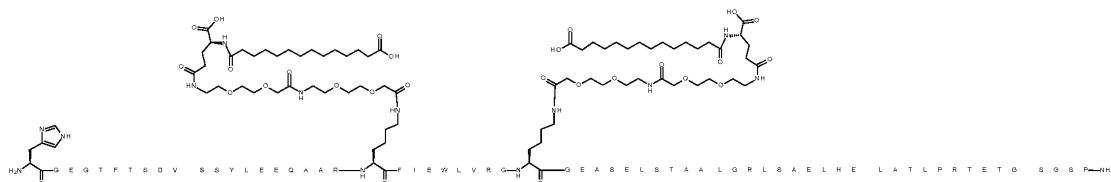
The amino acid sequence of

10 XAEGTFTSDVSSYLEEQAAREFIAWLVRGRKEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 122

Compound 0039

HGEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[[[4S)-4-carboxy-4-(13-

15 carboxytridecanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])
-FIEWLVRG-K([2-[2-[2-[2-[2-[2-[[[4S)-4-carboxy-4-(13-
carboxytridecanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])
-GEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



20 C₃₅₉ H₅₇₇ N₉₁ O₁₂₄

Molecular weight (average) calculated: 8151.9580 g/mol

mono isotopic mass: 8147.1642 g/mol

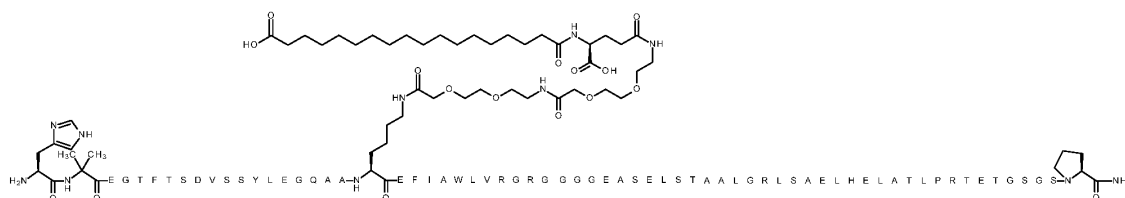
LCMS01: found (M + 5H)⁵⁺ 1631.6 (most abundant)

The amino acid sequence of

25 HGEGTFTSDVSSYLEEQAARKFIEWLVRGKGEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 123

Compound 0040

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFI A WLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



5 C₃₃₄ H₅₃₄ N₉₀ O₁₁₃

Molecular weight (average) calculated: 7618.3490 g/mol

mono isotopic mass: 7613.8806 g/mol

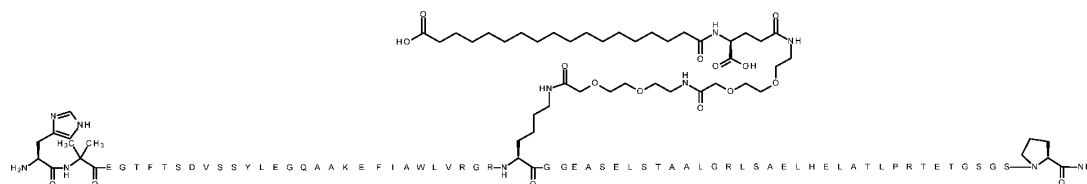
LCMS34: found (M + 5H)⁵⁺ 1524.92 (most abundant)

The amino acid sequence of

10 HXEGTFTSDVSSYLEGQAAKEFI A WLVRGRGGGGGEASELSTAALGRLSAELHELATLPRT
ETGSGSP has SEQ ID NO: 124

Compound 0042

15 H-Aib-EGTFTSDVSSYLEGQAAKEFI A WLVRGR-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₃₆ H₅₄₀ N₉₀ O₁₁₂

Molecular weight (average) calculated: 7632.4186 g/mol

20 mono isotopic mass: 7627.9326 g/mol

LCMS34: found (M + 5H)⁵⁺ 1527.76 (most abundant)

The amino acid sequence of

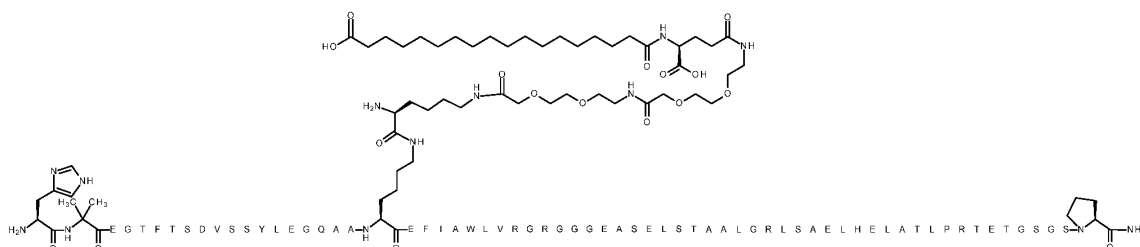
HXEGTFTSDVSSYLEGQAAKEFI A WLVRGRKGGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 125

25

Compound 0044

H-Aib-EGTFTSDVSSYLEGQAA-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl]-EFIAWLVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



5 C₃₃₈ H₅₄₃ N₉₁ O₁₁₃

Molecular weight (average) calculated: 7689.4699 g/mol

mono isotopic mass: 7684.9541 g/mol

LCMS34: found (M + 5H)⁵⁺ 1538.8 (most abundant)

The amino acid sequence of

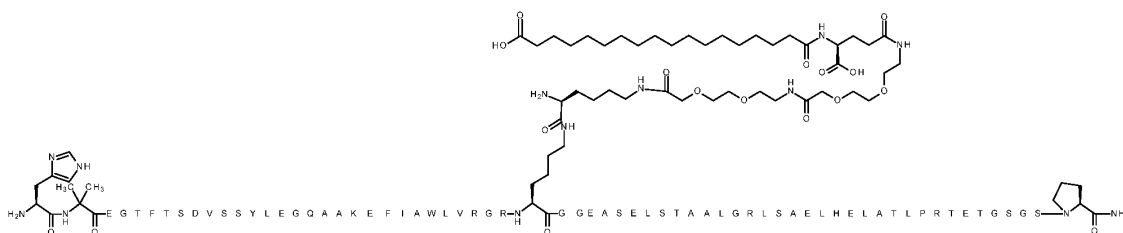
10 HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 117

Compound 0045

H-Aib-EGTFTSDVSSYLEGQAAKEFIAWLVRGR-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-

15 4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl]-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₂ H₅₅₂ N₉₂ O₁₁₃

20 Molecular weight (average) calculated: 7760.5909 g/mol

mono isotopic mass: 7756.0276 g/mol

LCMS34: found (M + 5H)⁵⁺ 1553.22 (most abundant)

The amino acid sequence of

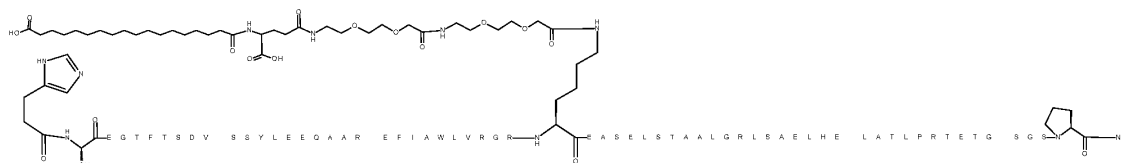
HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRKGGEASELSTAALGRLSAELHELATLPRTET

25 GSGSP has SEQ ID NO: 125

Compound 0051

Imp-AEGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-amide

5



C₃₃₄ H₅₃₅ N₈₉ O₁₁₂

Molecular weight (average) calculated 7589.3508 g/mol

mono isotopic mass: 7584.8904 g/mol

10 LCMS01: found (M + 5H)⁵⁺ 1519.1 (most abundant)

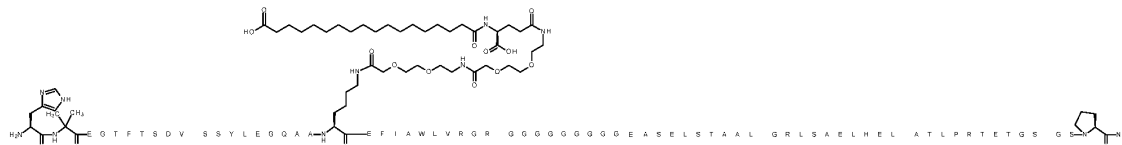
The amino acid sequence of

XAEGTFTSDVSSYLEEQAAREFIAWLVRGRKEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 122

15 Compound 0052

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVRGRGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



20 C₃₄₄ H₅₄₉ N₉₅ O₁₁₈

Molecular weight (average) calculated: 7903.6056 g/mol

mono isotopic mass: 7898.9879 g/mol

LCMS34: found (M + 5H)⁵⁺ 1581.7 (most abundant)

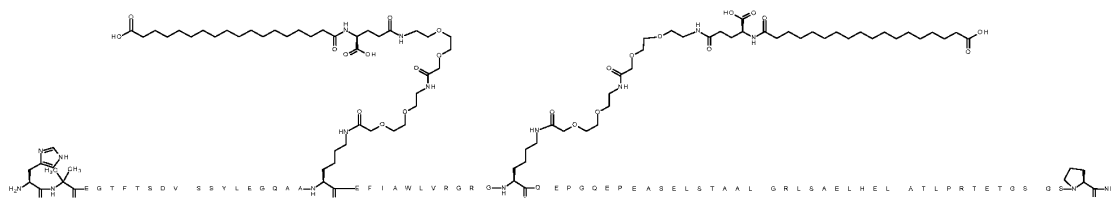
The amino acid sequence of

25 HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGGGGGGGGEASELSTAALGRLSAELHEL

ATLPRTETGSGSP has SEQ ID NO: 126

Compound 0056

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-EFIWLVRGRG-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-QEPGQEPEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₄₀₁ H₆₄₅ N₁₀₁ O₁₃₆

Molecular weight (average) calculated: 9057.0071 g/mol

mono isotopic mass: 9051.6660 g/mol

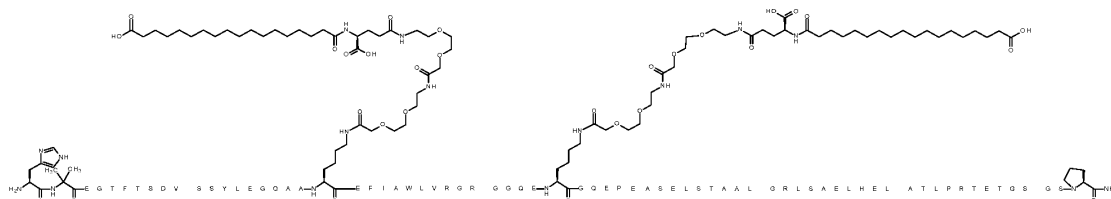
10 LCMS34: found (M + 5H)⁵⁺ 1812.4 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEGQAAKEFIWLVRGRGKQEPGQEPEASELSTAALGRLSAELHELA
TLPRRTETGSGSP has SEQ ID NO: 127

15 Compound 0057

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-EFIWLVRGRGGQE-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GQEPEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₉₈ H₆₄₁ N₁₀₁ O₁₃₆

Molecular weight (average) calculated: 9016.9432 g/mol

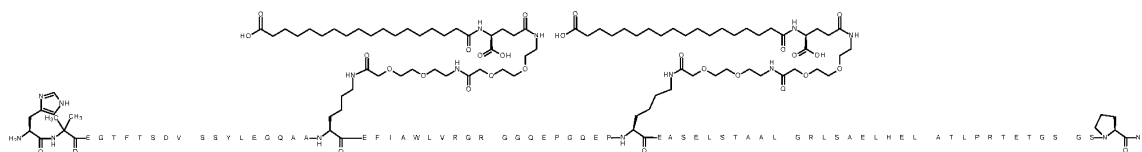
mono isotopic mass: 9011.6347 g/mol

25 LCMS34: found (M + 5H)⁵⁺ 1804.1 (most abundant)

The amino acid sequence of
HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGQEKGEPEASELSTAALGRLSAELHELA
TLPRRTETGSGSP has SEQ ID NO: 128

5 Compound 0071

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVRGRGGQEPGQEP-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₄₀₃ H₆₄₈ N₁₀₂ O₁₃₇

Molecular weight (average) calculated: 9114.0584 g/mol

mono isotopic mass: 9108.6874 g/mol

15 LCMS34: found (M + 5H)⁵⁺ 1823.7 (most abundant)

The amino acid sequence of
HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGQEPGQEPKEASELSTAALGRLSAELHEL
ATLPRRTETGSGSP has SEQ ID NO: 129

20 Compound 0072

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVRGRGG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₇ H₅₉₃ N₉₃ O₁₂₄

Molecular weight (average) calculated: 8292.1840 g/mol

mono isotopic mass: 8287.2955 g/mol

LCMS34: found (M + 5H)⁵⁺ 1659.27 (most abundant)

The amino acid sequence of

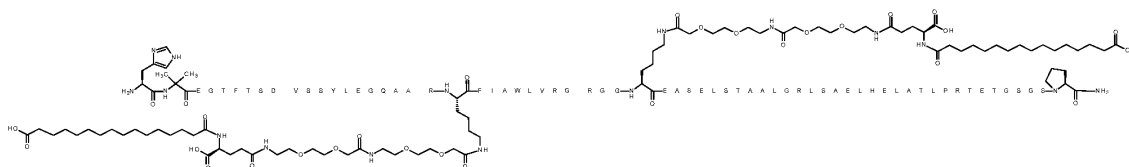
HXEGTFTSDVSSYLEGQAARKEFIAWLVRGRGGKEASELSTAALGRLSAELHELATLPRTET

GSGSP has SEQ ID NO: 130

5

Compound 0073

H-Aib-EGTFTSDVSSYLEGQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGRGG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₈ H₅₉₈ N₉₆ O₁₂₂

Molecular weight (average) calculated: 8319.2557 g/mol

15 mono isotopic mass: 8314.3540 g/mol

LCMS34: found (M + 5H)⁵⁺ 1664.69 (most abundant)

The amino acid sequence of

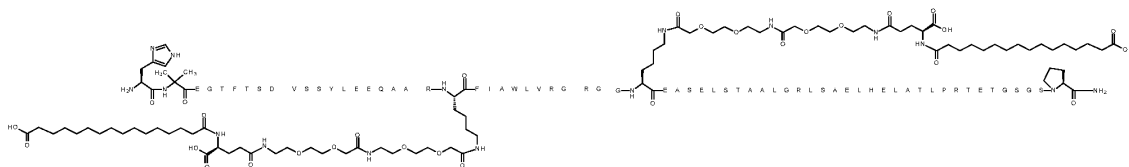
HXEGTFTSDVSSYLEGQAARKFIAWLVRGRGGKEASELSTAALGRLSAELHELATLPRTET

GSGSP has SEQ ID NO: 131

20

Compound 0074

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGRGG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₁ H₆₀₂ N₉₆ O₁₂₄

Molecular weight (average) calculated: 8391.3184 g/mol

C₃₆₇ H₅₉₃ N₉₃ O₁₂₄

Molecular weight (average) calculated: 8292.1840 g/mol

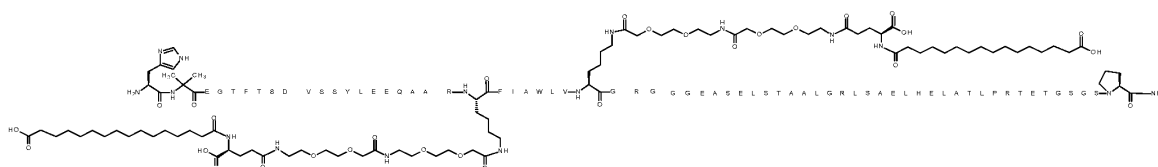
mono isotopic mass: 8287.2955 g/mol

LCMS34: found (M + 5H)⁵⁺ 1659.48 (most abundant)

- 5 The amino acid sequence of
HXEGTFTSDVSSYLEEQAARKFIAWLVRGKGGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 134

Compound 0077

- 10 H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIAWL-V-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



15

C₃₆₇ H₅₉₃ N₉₃ O₁₂₄

Molecular weight (average) calculated: 8292.1840 g/mol

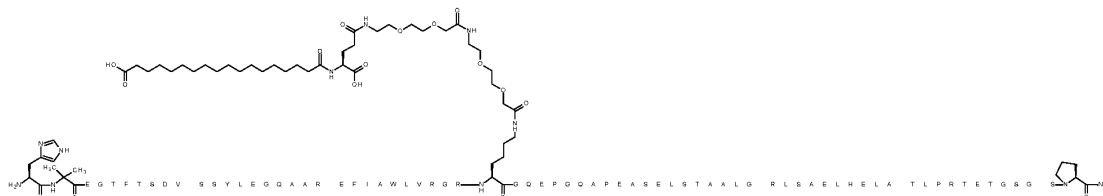
mono isotopic mass: 8287.2955 g/mol

LCMS34: found (M + 5H)⁵⁺ 1659.48 (most abundant)

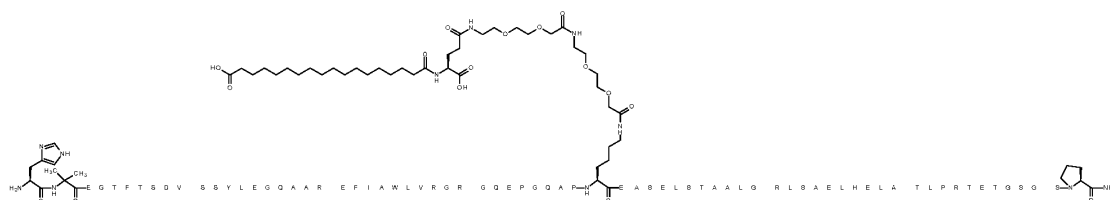
- 20 The amino acid sequence of
HXEGTFTSDVSSYLEEQAARKFIAWLVRGKGGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 135

Compound 0083

- 25 H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GQEPGQAPEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



- 25 H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRGRGQEPGQAP-K([2-[2-[2-[2-[2-[[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-EASELSTAALGRLSAELHELATLPRTTETGSGSP-amide



C₃₆₄ H₅₈₂ N₁₀₀ O₁₂₂

Molecular weight (average) calculated: 8311.1127 g/mol

mono isotopic mass: 8306.2411 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1662.24 (mono isotopic)

The amino acid sequence of

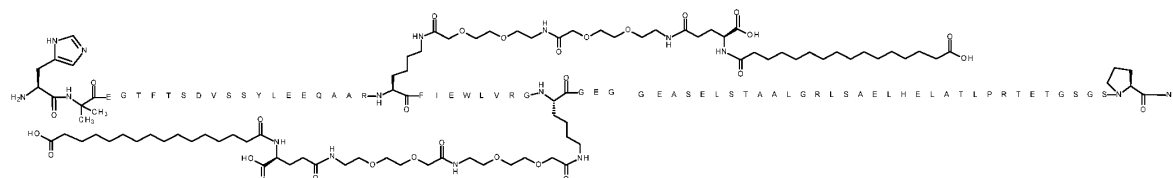
HXEGTFTSDVSSYLEGQAAREFIAWLVRGRGQEPGQAPKEASELSTAALGRLSAELHELA

TLPRTEGSGSP has SEQ ID NO: 138

- 10 Compound 0086

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GEGGEASELSTAALGRLSAELHELATLPRTEGSGSP-amide

- 15 tyl])-GEGGEASELSTAALGRLSAELHELATLPRTEGSGSP-amide



C₃₇₄ H₆₀₂ N₉₄ O₁₂₉

Molecular weight (average) calculated: 8479.3341 g/mol

mono isotopic mass: 8474.3436 g/mol

- 20 LCMS34: found (M + 5H)⁵⁺ 1696.69 (most abundant)

The amino acid sequence of

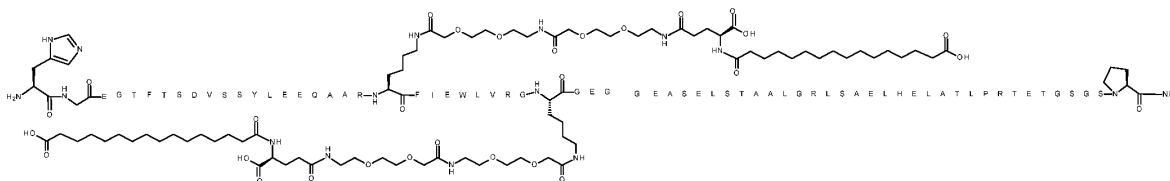
HXEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGGEASELSTAALGRLSAELHELATLPRTE

TGSGSP has SEQ ID NO: 139

- 25 Compound 0087

HGEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-

carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GEGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₂ H₅₉₈ N₉₄ O₁₂₉

5 Molecular weight (average) calculated: 8451.2809 g/mol

mono isotopic mass: 8446.3123 g/mol

LCMS34: found (M + 5H)⁵⁺ 1691.08 (most abundant)

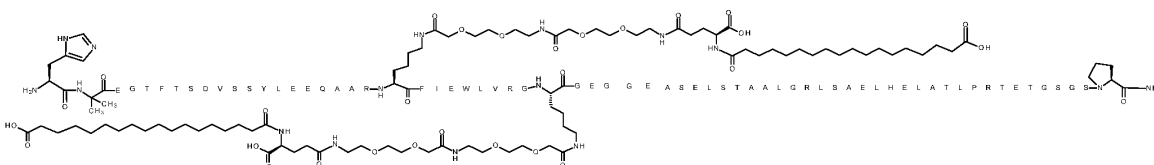
The amino acid sequence of

HGEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGGEASELSTAALGRLSAELHELATLPRTET

10 TGSGSP has SEQ ID NO: 140

Compound 0089

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GEGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₈ H₆₁₀ N₉₄ O₁₂₉

20 Molecular weight (average) calculated: 8535.4404 g/mol

mono isotopic mass: 8530.4062 g/mol

LCMS34: found (M + 5H)⁵⁺ 1707.9 (most abundant)

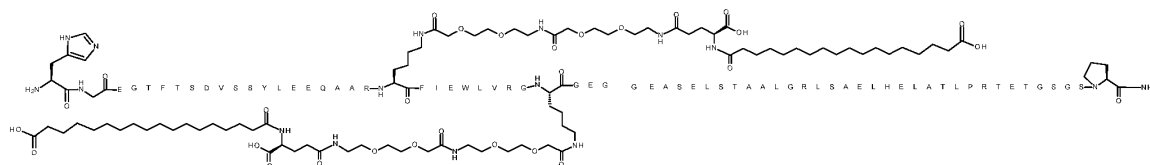
The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGGEASELSTAALGRLSAELHELATLPRTET

25 TGSGSP has SEQ ID NO: 139

Compound 0090

HGEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GEGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₆ H₆₀₆ N₉₄ O₁₂₉

Molecular weight (average) calculated: 8507.3872 g/mol

mono isotopic mass: 8502.3749 g/mol

10 LCMS34: found (M + 5H)⁵⁺ 1702.49 (most abundant)

The amino acid sequence of

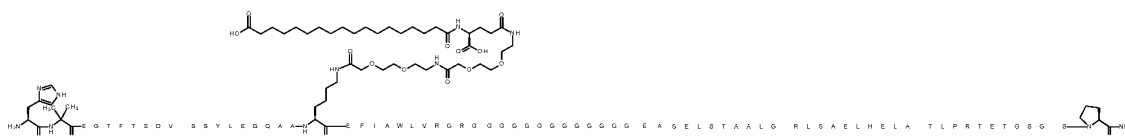
HGEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 140

15 Compound 0092

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-

EFIAWLVRGRGGGGGGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-

20 amide



C₃₅₂ H₅₆₁ N₉₉ O₁₂₂

Molecular weight (average) calculated: 8131.8108 g/mol

mono isotopic mass: 8127.0737 g/mol

25 LCMS34: found (M + 5H)⁵⁺ 1627.5 (most abundant)

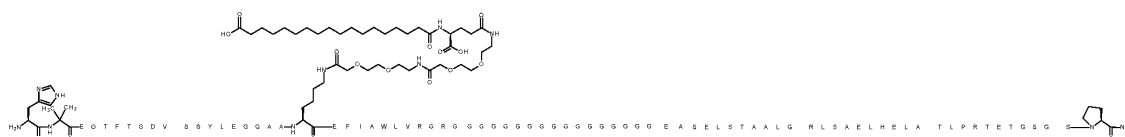
The amino acid sequence of

HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGGGGGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 141

Compound 0093

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[[[4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

- 5 E F I A W L V R G R G G G G G G G G G G G G G G G G G G E A S E L S T A A L G R L S A E L H E L A T L P R T E T G S G
SP-amide



C₃₆₀ H₅₇₃ N₁₀₃ O₁₂₆

Molecular weight (average) calculated: 8360.0161 g/mol

- 10 mono isotopic mass: 8355.1596 g/mol

LCMS34: found (M + 5H)⁵⁺ 1672.8 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEGQAAKEFI A W L V R G R G G G G G G G G G G G G G G G G G E A S E L S T A A L G
R L S A E L H E L A T L P R T E T G S G S P has SEQ ID NO: 142

- 15

Compound 0094

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[[[4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-E F I A W L V R G R G G G G G G G G G G G G G G G G G E A S E L S T A A L G R L S A E L H E L A T L P R T E T G S G S P-amide



- 20

C₃₄₆ H₅₅₃ N₉₅ O₁₁₈

Molecular weight (average) calculated: 7931.6587 g/mol

mono isotopic mass: 7927.0192 g/mol

LCMS34: found (M + 5H)⁵⁺ 1587.2 (most abundant)

- 25 The amino acid sequence of

HXEGTFTSDVSSYLEGQAAKEFI A W L V R G R G G G G G G G G G G G G E A S E L S T A A L G R L S A E L H E L
A T L P R T E T G S G S P has SEQ ID NO: 126

Compound 0095

- 30 H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[[[4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]

oxy]acetyl]amino]ethoxy]ethoxy]acetyl))-

EFIWLVRGRGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₄ H₅₆₆ N₉₆ O₁₁₉

5 Molecular weight (average) calculated: 8070.8536 g/mol

mono isotopic mass: 8066.1189 g/mol

LCMS34: found (M + 5H)⁵⁺ 1615.0 (most abundant)

The amino acid sequence of

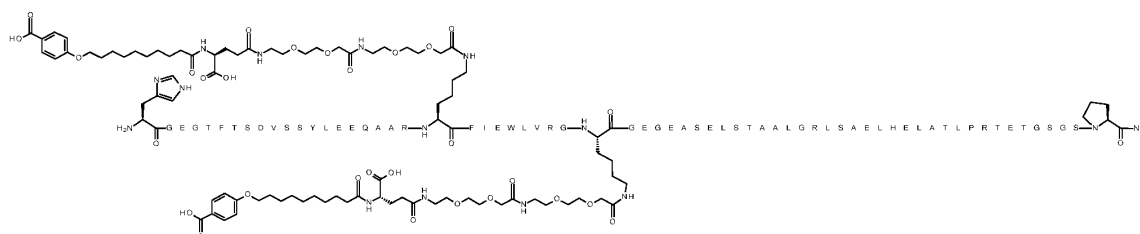
HXEGTFTSDVSSYLEGQAAKEFIWLVRGRGGGGGGGGGEASELSTAALGRLSAELHEL

10 ATLPRTETGSGSP has SEQ ID NO: 126

Compound 0096

HGEGTFTSDVSSYLEEQAAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-

15 acetyl])-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-
carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]
acetyl))-GEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₂ H₅₈₃ N₉₃ O₁₃₀

20 Molecular weight (average) calculated: 8438.1545 g/mol

mono isotopic mass: 8433.1868 g/mol

LCMS34: found (M + 5H)⁵⁺ 1688.65 (most abundant)

The amino acid sequence of

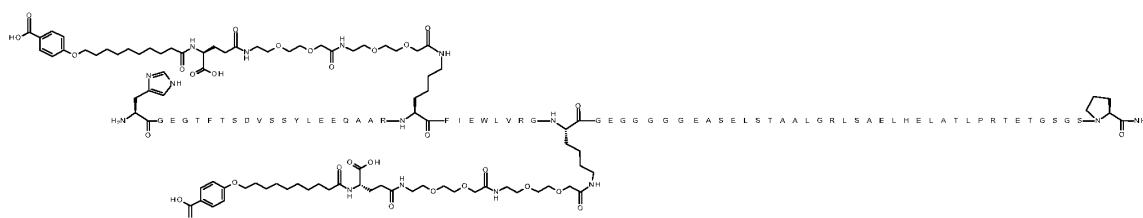
HGEGTFTSDVSSYLEEQAAARKFIEWLVRGKGEGEASELSTAALGRLSAELHELATLPRTET

25 GSGSP has SEQ ID NO: 143

Compound 0097

HGEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GEGGGGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide

5



C₃₈₀ H₅₉₅ N₉₇ O₁₃₄

Molecular weight (average) calculated: 8666.3598 g/mol

mono isotopic mass: 8661.2726 g/mol

10 LCMS34: found (M + 5H)⁵⁺ 1734.26 (most abundant)

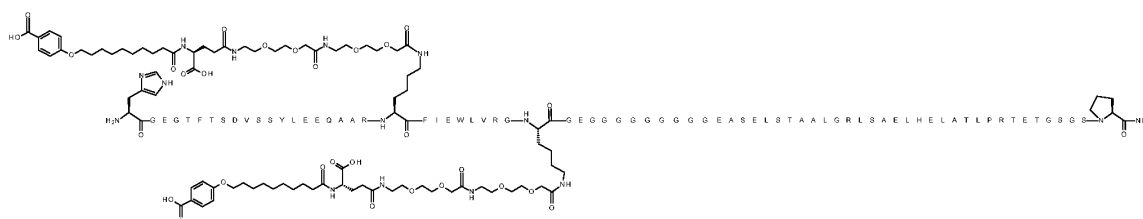
The amino acid sequence of

HGEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGGGGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP has SEQ ID NO: 144

15 Compound 0098

HGEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GEGGGGGGGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide

20



C₃₈₈ H₆₀₇ N₁₀₁ O₁₃₈

Molecular weight (average) calculated: 8894.5651 g/mol

mono isotopic mass: 8889.3585 g/mol

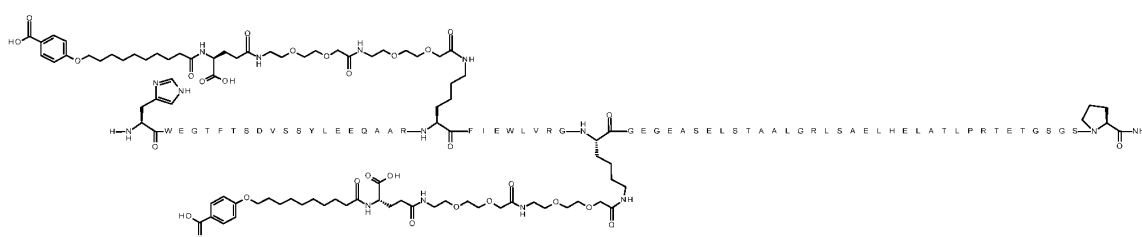
25 LCMS34: found (M + 5H)⁵⁺ 1779.88 (most abundant)

The amino acid sequence of

HGEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGGGGGGGGGEASELSTAALGRLSAELH
ELATLPRTETGSGSP has SEQ ID NO: 145

5 Compound 0099

HWEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GEGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₈₁ H₅₉₀ N₉₄ O₁₃₀

Molecular weight (average) calculated: 8567.3131 g/mol

mono isotopic mass: 8562.2446 g/mol

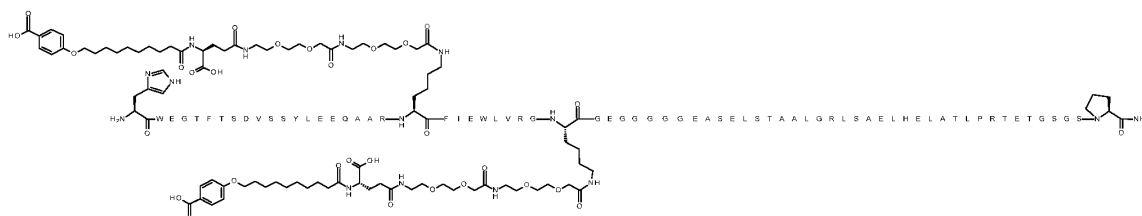
15 LCMS34: found (M + 5H)⁵⁺ 1714.46 (most abundant)

The amino acid sequence of

HWEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 146

20 Compound 0100

HWEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[[[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GEGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₈₉ H₆₀₂ N₉₈ O₁₃₄

Molecular weight (average) calculated: 8795.5184 g/mol

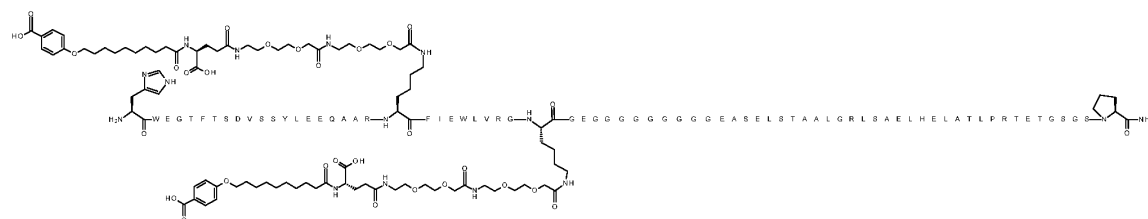
mono isotopic mass: 8790.3305 g/mol

LCMS34: found (M + 5H)⁵⁺ 1760.08 (most abundant)

- 5 The amino acid sequence of
HWEFTFTSDVSSYLEEQAARKFIEWLVRGKGEGGGGGGEASELSTAALGRLSAELHELATL
PRTETGSGSP has SEQ ID NO: 147

Compound 0101

- 10 HWEFTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GEGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



15

C₃₉₇ H₆₁₄ N₁₀₂ O₁₃₈

Molecular weight (average) calculated: 9023.7237 g/mol

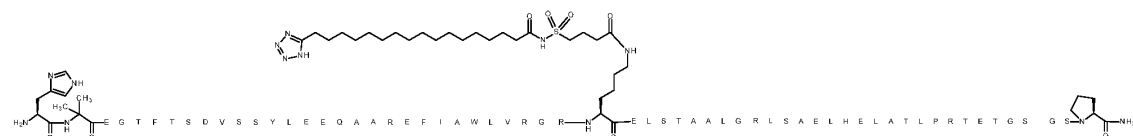
mono isotopic mass: 9018.4163 g/mol

LCMS34: found (M + 5H)⁵⁺ 1805.49 (most abundant)

- 20 The amino acid sequence of
HWEFTFTSDVSSYLEEQAARKFIEWLVRGKGEGGGGGGGGGEASELSTAALGRLSAELH
ELATLPRTETGSGSP has SEQ ID NO: 148

Compound 0102

- 25 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K(4-[17-(1H-tetrazol-5-yl)heptadecanoylsulfamoyl]butanoyl)-ELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₁₁ H₄₉₉ N₈₉ O₉₈ S

Molecular weight (average) calculated: 7084.8923 g/mol

mono isotopic mass: 7080.6520 g/mol

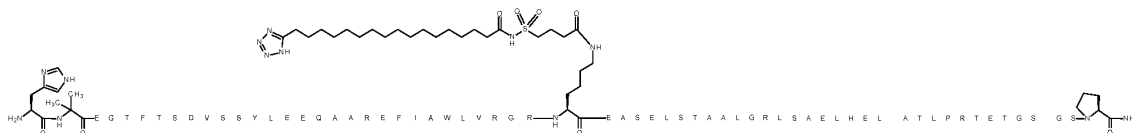
LCMS01: found (M + 5H)⁵⁺ 1418 (most abundant)

The amino acid sequence of

- 5 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKELSTAALGRLSAELHELATLPRRTETGSGSP
has SEQ ID NO: 149

Compound 0103

- 10 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K(4-[17-(1H-tetrazol-5-
yl)heptadecanoylsulfamoyl]butanoyl)-EASELSTAALGRLSAELHELATLPRRTETGSGSP-
amide



C₃₂₂ H₅₁₆ N₉₂ O₁₀₄ S

Molecular weight (average) calculated: 7372.1614 g/mol

- 15 mono isotopic mass: 7367.7637 g/mol

LCMS01: found (M + 5H)⁵⁺ 1475 (most abundant)

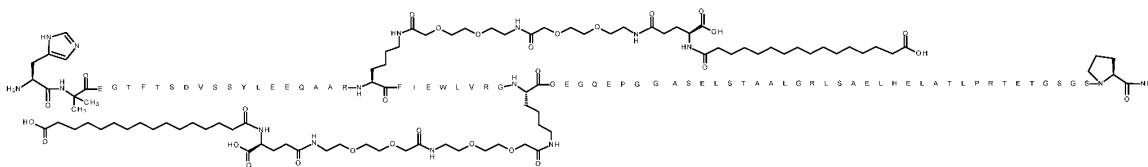
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKEASELSTAALGRLSAELHELATLPRRTETGS
GSP has SEQ ID NO: 150

20

Compound 0105

- H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-
carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace-
tyl])-FIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-
25 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace-
tyl])-GEGQEPGGASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₈₆ H₆₂₀ N₉₈ O₁₃₃

Molecular weight (average) calculated: 8761.6298 g/mol

mono isotopic mass: 8756.4764 g/mol

LCMS34: found (M + 5H)⁵⁺ 1753.31 (most abundant)

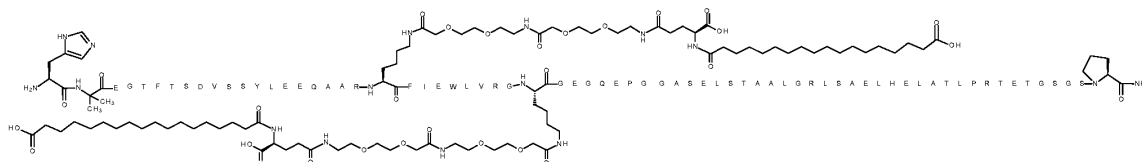
The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGQEPGGASELSTAALGRLSAELHELATL

5 PR TetGSGSP has SEQ ID NO: 151

Compound 0106

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GEGQEPGGASELSTAALGRLSAELHELATLPR TetGSGSP-amide



C₃₉₀ H₆₂₈ N₉₈ O₁₃₃

15 Molecular weight (average) calculated: 8817.7361 g/mol

mono isotopic mass: 8812.5390 g/mol

LCMS34: found (M + 5H)⁵⁺ 1764.33 (most abundant)

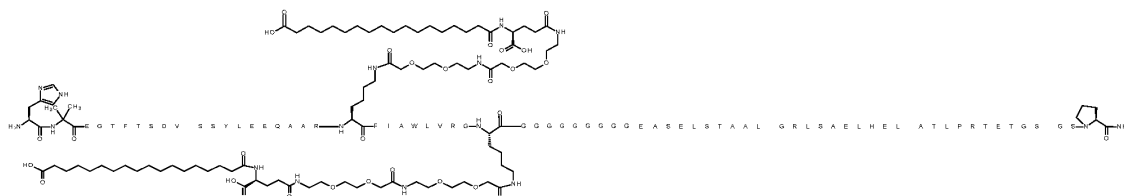
The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIEWLVRGKGEGQEPGGASELSTAALGRLSAELHELATL

20 PR TetGSGSP has SEQ ID NO: 151

Compound 0109

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GGGGGGGGGEASELSTAALGRLSAELHELATLPR TetGSGSP-amide



C₃₈₃ H₆₁₉ N₉₉ O₁₃₀

Molecular weight (average) calculated: 8690.5983 g/mol

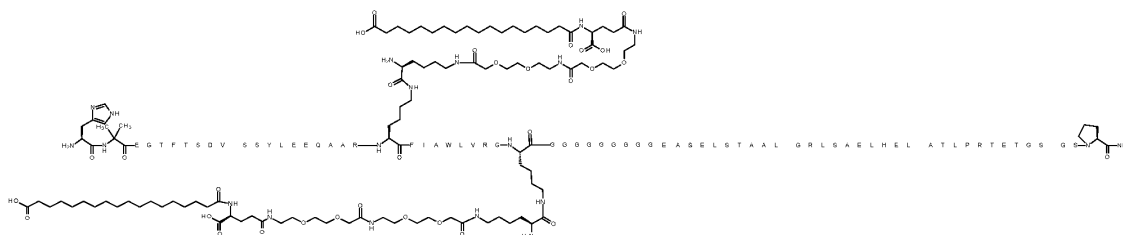
mono isotopic mass: 8685.4869 g/mol

LCMS34: found (M + 5H)⁵⁺ 1738.9 (most abundant)

- 5 The amino acid sequence of
HXEGTFTSDVSSYLEEQAARKFIAWLVRGKGGGGGGGGGEASELSTAALGRLSAELHEL
ATLPRRTETGSGSP has SEQ ID NO: 152

Compound 0110

- 10 H-Aib-EGTFTSDVSSYLEEQAAR-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-FIAWLVRG-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-GGGGGGGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₉₅ H₆₄₃ N₁₀₃ O₁₃₂

- 20 Molecular weight (average) calculated: 8946.9428 g/mol

mono isotopic mass: 8941.6768 g/mol

LCMS34: found (M + 5H)⁵⁺ 1790.3 (most abundant)

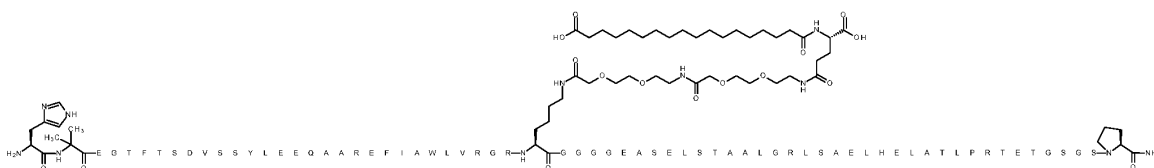
The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIAWLVRGKGGGGGGGGGEASELSTAALGRLSAELHEL

- 25 ATLPRRTETGSGSP has SEQ ID NO: 152

Compound 0111

- 30 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([(2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₄₃ H₅₅₀ N₉₄ O₁₁₆

Molecular weight (average) calculated: 7846.5973 g/mol

mono isotopic mass: 7842.0028 g/mol

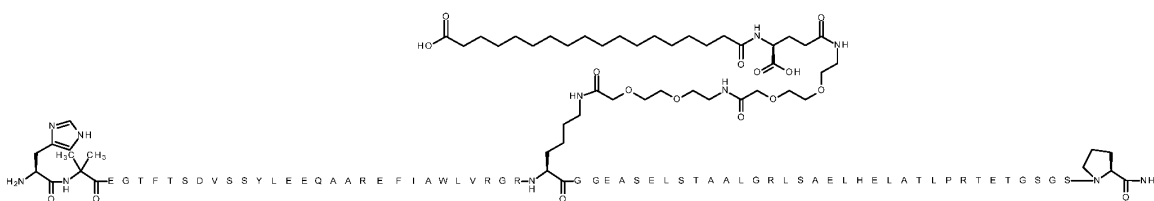
- 5 LCMS34: found (M + 5H)⁵⁺ 1570.22 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 153

10 Compound 0114

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]acetyl])-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



15 Compound 0115

Molecular weight (average) calculated: 7732.4947 g/mol

mono isotopic mass: 7727.9599 g/mol

LCMS34: found (M + 5H)⁵⁺ 1547.4 (most abundant)

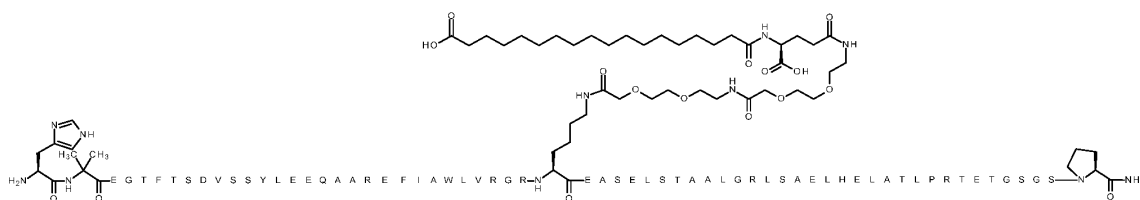
The amino acid sequence of

- 20 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 154

Compound 0115

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]acetyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-amide

- 25



C₃₃₅ H₅₃₈ N₉₀ O₁₁₂

Molecular weight (average) calculated: 7618.3920 g/mol

mono isotopic mass: 7613.9169 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1524.6 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKEASELSTAALGRLSAELHELATLPRTETGS

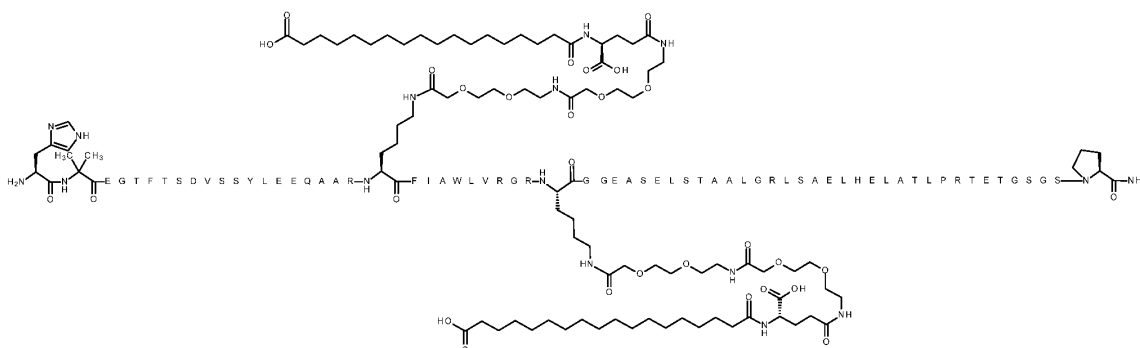
GSP has SEQ ID NO: 150

- 10 Compound 0116

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide

- 15 ty])-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₅ H₆₁₀ N₉₆ O₁₂₄

Molecular weight (average) calculated: 8447.4247 g/mol

mono isotopic mass: 8442.4378 g/mol

- 20 LCMS34: found (M + 5H)⁵⁺ 1690.3 (most abundant)

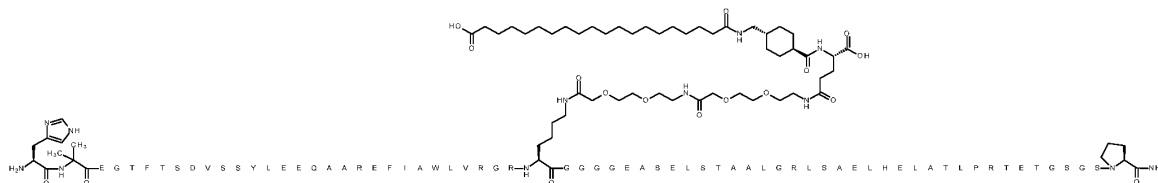
The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIAWLVRGRKGGEASELSTAALGRLSAELHELATLPRTET

GSGSP has SEQ ID NO: 155

Compound 0120

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVGRGR-K([2-[2-[2-[[2-[2-[2-[[4S)-4-carboxy-4-[[4-
 [(19-
 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth
 5 oxy]acetyl]amino]ethoxy]ethoxy]acetyl])-
 GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₃ H₅₆₇ N₉₅ O₁₁₇

Molecular weight (average) calculated: 8013.8454 g/mol

10 mono isotopic mass: 8009.1338 g/mol

LCMS34: found (M + 5H)⁵⁺ 1603.63 (most abundant)

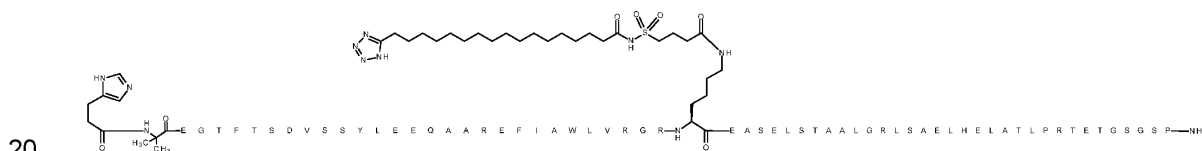
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVGRGRKGGGGEASELSTAALGRLSAELHELATLPR
 TETGSGSP has SEQ ID NO: 153

15

Compound 0124

Imp-Aib-EGTFTSDVSSYLEEQAAREFIAWLVGRGR-K(4-[17-(1H-tetrazol-5-
 yl)heptadecanoylsulfamoyl]butanoyl)-EASELSTAALGRLSAELHELATLPRRTETGSGSP-
 amide



20

C₃₂₂ H₅₁₅ N₉₁ O₁₀₄ S

Molecular weight (average) calculated: 7357.1468 g/mol

mono isotopic mass: 7352.7528 g/mol

LCMS01: found (M + 5H)⁵⁺ 1472 (most abundant)

25 The amino acid sequence of

XXEGTFTSDVSSYLEEQAAREFIAWLVGRGRKEASELSTAALGRLSAELHELATLPRRTETGS
 GSP has SEQ ID NO: 156

Compound 0125

H-Aib-EGTFTSDVSSYLEEQAAR-K(4-[17-(1H-tetrazol-5-yl)heptadecanoylsulfamoyl]butanoyl)-
FIAWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



5 C₃₁₉ H₅₁₂ N₉₂ O₁₀₂ S

Molecular weight (average) calculated: 7300.0988 g/mol

mono isotopic mass: 7295.7426 g/mol

LCMS01: found (M + 5H)⁵⁺ 1461 (most abundant)

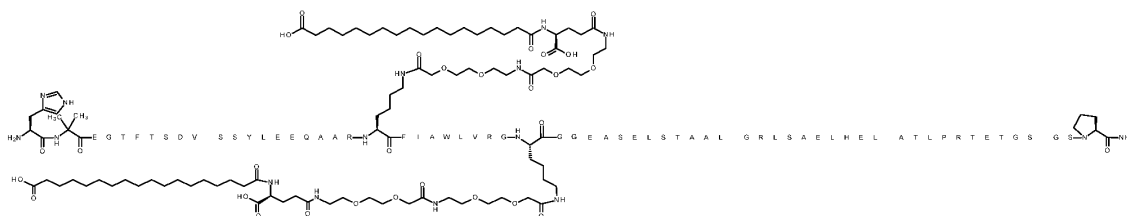
The amino acid sequence of

10 HXEGTFTSDVSSYLEEQAARKFIAWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGS
GSP has SEQ ID NO: 157

Compound 0127

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

15 carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]-FIAWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



20 C₃₆₉ H₅₉₈ N₉₂ O₁₂₃

Molecular weight (average) calculated: 8291.2390 g/mol

mono isotopic mass: 8286.3367 g/mol

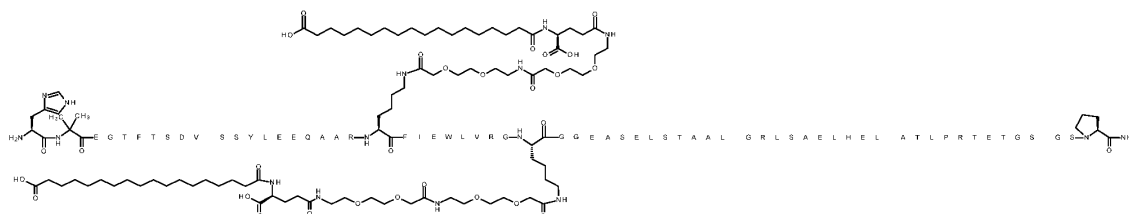
LCMS34: found (M + 5H)⁵⁺ 1659.1 (most abundant)

The amino acid sequence of

25 HXEGTFTSDVSSYLEEQAARKFIAWLVRGKGGEASELSTAALGRLSAELHELATLPRRTETG
SGSP has SEQ ID NO: 158

Compound 0128

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₇₁ H₆₀₀ N₉₂ O₁₂₅

Molecular weight (average) calculated: 8349.2751 g/mol

mono isotopic mass: 8344.3421 g/mol

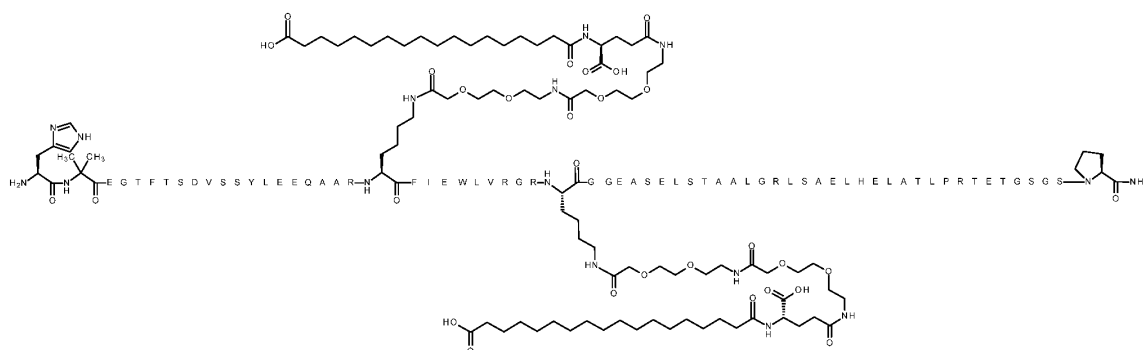
10 LCMS34: found (M + 5H)⁵⁺ 1670.7 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIEWLVRGKGGEASELSTAALGRLSAELHELATLPRRTETGSGSP has SEQ ID NO: 159

15 Compound 0129

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₇₇ H₆₁₂ N₉₆ O₁₂₆

Molecular weight (average) calculated: 8505.4608 g/mol

mono isotopic mass: 8500.4432 g/mol

LCMS_ZQ: found $(M + 5H)^{5+}$ 1702.0 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIEWLVRGRKGGEASELSTAALGRLSAELHELATLPRTET

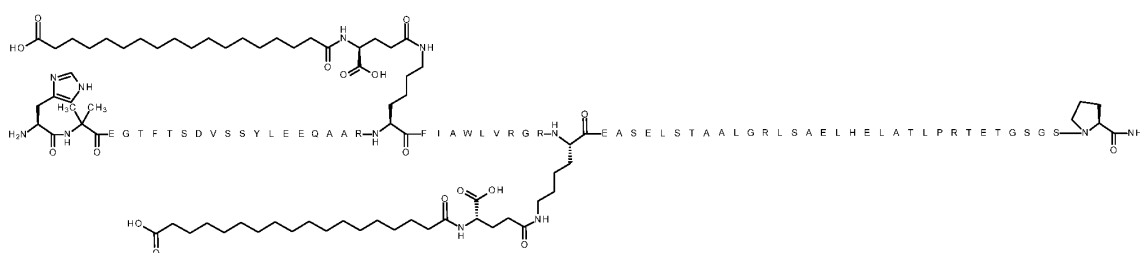
GSGSP has SEQ ID NO: 160

5

Compound 0131

H-Aib-EGTFTSDVSSYLEEQAAR-K([(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl])-FIAWLVRGR-K([(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-

10 amide



C₃₄₇ H₅₆₀ N₉₀ O₁₁₀

Molecular weight (average) calculated: 7752.6963 g/mol

mono isotopic mass: 7748.0993 g/mol

15 LCMS34: found $(M + 5H)^{5+}$ 1551.6 (most abundant)

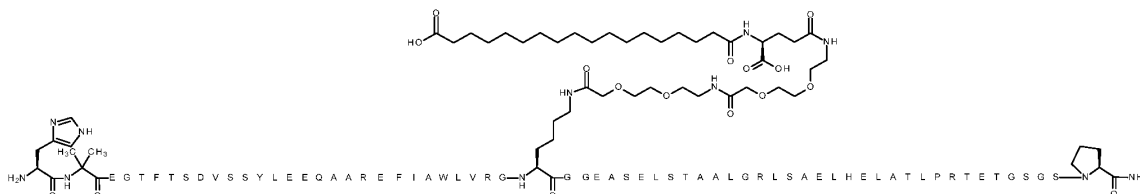
The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIAWLVRGRKEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 161

20 Compound 0132

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRG-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



25 C₃₃₃ H₅₃₂ N₈₈ O₁₁₃

Molecular weight (average) calculated: 7576.3090 g/mol

mono isotopic mass: 7571.8588 g/mol

LCMS34: found (M + 5H)⁵⁺ 1516.11 (most abundant)

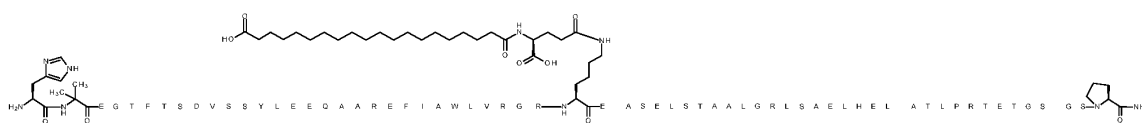
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAAREFIAWLVRGKGGEASELSTAALGRLSAELHELATLPRTETG

5 SGSP has SEQ ID NO: 162

Compound 0141

H-Aib-EGTFTSDVSSYLEEQAAAREFIAWLVRGR-K([(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-
10 amide



C₃₂₅ H₅₂₀ N₈₈ O₁₀₆

Molecular weight (average) calculated: 7356.1323 g/mol

mono isotopic mass: 7351.8005 g/mol

15 LCMS01: found (M + 5H)⁵⁺ 1472 (most abundant)

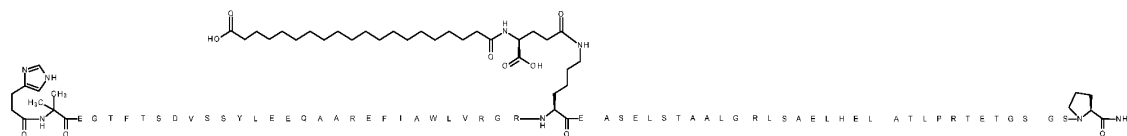
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAAREFIAWLVRGRKEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 150

Compound 0142

Imp-Aib-EGTFTSDVSSYLEEQAAAREFIAWLVRGR-K([(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl])-EASELSTAALGRLSAELHELATLPRTETGSGSP-
20 amide



25 C₃₂₅ H₅₁₉ N₈₇ O₁₀₆

Molecular weight (average) calculated: 7341.1177 g/mol

mono isotopic mass: 7336.7896 g/mol

LCMS01: found (M + 5H)⁵⁺ 1469 (most abundant)

The amino acid sequence of

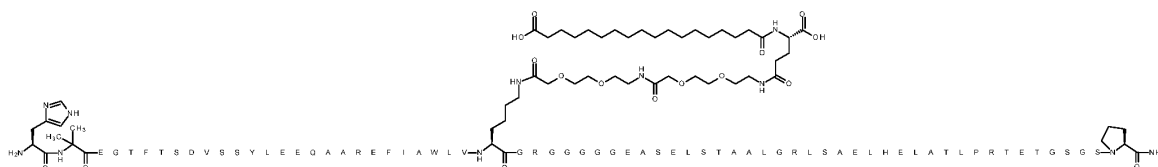
30 XXEGTFTSDVSSYLEEQAAAREFIAWLVRGRKEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 156.

Compound 0144

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLK-([2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide

5



C₃₃₉ H₅₄₁ N₉₁ O₁₁₆

Molecular weight (average) calculated: 7747.4629 g/mol

mono isotopic mass: 7742.9232 g/mol

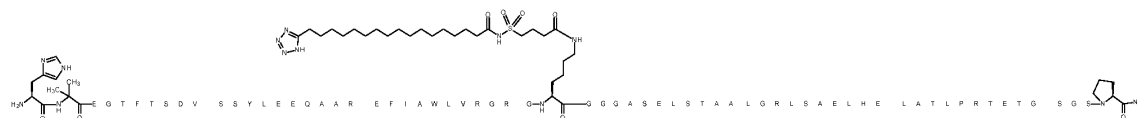
10 LCMS34: found (M + 5H)⁵⁺ 1550.39 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVKGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 163

15 Compound 0145

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGRG-K(4-[17-(1H-tetrazol-5-yl)heptadecanoylsulfamoyl]butanoyl)-GGGASELSTAALGRLSAELHELATLPRTETGSGSP-amide



20 C₃₂₅ H₅₂₁ N₉₅ O₁₀₅ S

Molecular weight (average) calculated: 7471.2527 g/mol

mono isotopic mass: 7466.8070 g/mol

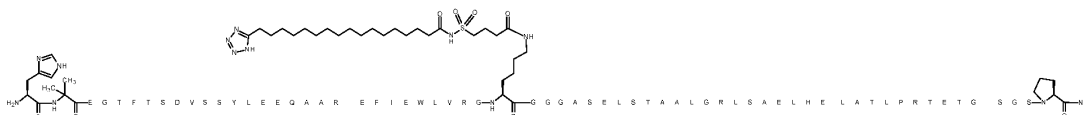
LCMS01: found (M + 5H)⁵⁺ 1495 (most abundant)

The amino acid sequence of

25 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRGKGGGASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 164

Compound 0146

H-Aib-EGTFTSDVSSYLEEQAAAREFIEWLVGRG-K(4-[17-(1H-tetrazol-5-yl)heptadecanoylsulfamoyl]butanoyl)-GGGASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



5 C₃₁₉ H₅₀₈ N₉₀ O₁₀₅ S

Molecular weight (average) calculated: 7316.0518 g/mol

mono isotopic mass: 7311.6899 g/mol

LCMS01: found (M + 5H)⁵⁺ 1464 (most abundant)

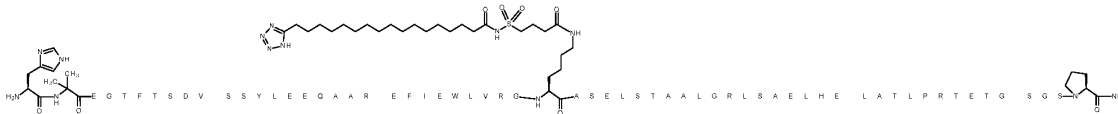
The amino acid sequence of

10 HXEGTFTSDVSSYLEEQAAAREFIEWLVGRGKGGGASELSTAALGRLSAELHELATLPRRTETGSGSP has SEQ ID NO: 165

Compound 0147

H-Aib-EGTFTSDVSSYLEEQAAAREFIEWLVGRG-K(4-[17-(1H-tetrazol-5-

15 yl)heptadecanoylsulfamoyl]butanoyl)-ASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₁₃ H₄₉₉ N₈₇ O₁₀₂ S

Molecular weight (average) calculated: 7144.8979g/mol

mono isotopic mass: 7140.6255 g/mol

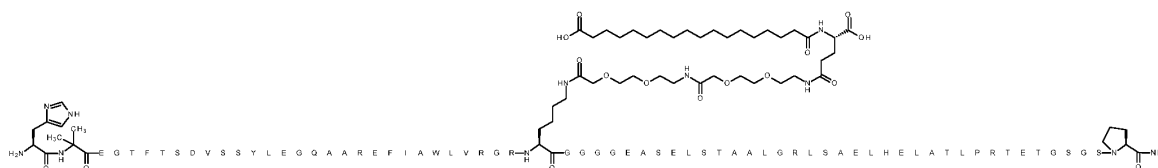
20 LCMS01: found (M + 5H)⁵⁺ 1430 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAAREFIEWLVGRGKASELSTAALGRLSAELHELATLPRRTETGSGS
P has SEQ ID NO: 166.

25 Compound 0151

H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVGRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₄₀ H₅₄₆ N₉₄ O₁₁₄

Molecular weight (average) calculated: 7774.5346 g/mol

mono isotopic mass: 7769.9817 g/mol

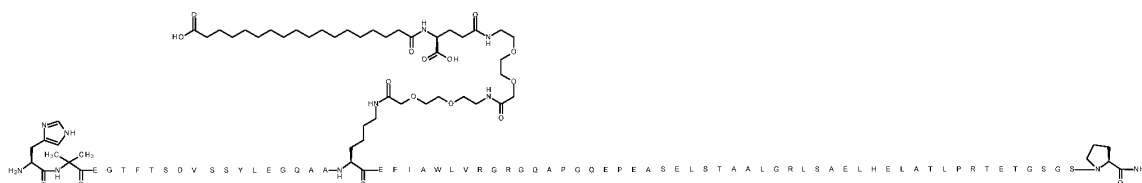
- 5 LCMS34: found (M + 5H)⁵⁺ 1555.81 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEGQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 167.

10 Compound 0156

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]acetyl])-EFIAWLVRGRGQAPGQEPEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



- 15 C₃₅₈ H₅₇₀ N₉₆ O₁₂₁

Molecular weight (average) calculated: 8154.9270 g/mol

mono isotopic mass: 8150.1400 g/mol

LCMS34: found (M + 5H)⁵⁺ 1631.04 (mono isotopic)

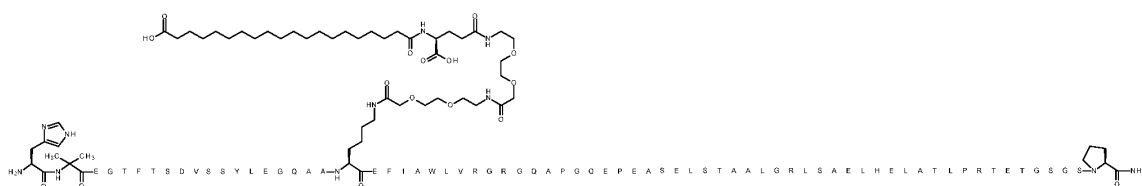
The amino acid sequence of

- 20 HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGQAPGQEPEASELSTAALGRLSAELHELAT
LPRRTETGSGSP has SEQ ID NO: 168

Compound 0157

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-

- 25 carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVRGRGQAPGQEPEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₀ H₅₇₄ N₉₆ O₁₂₁

Molecular weight (average) calculated: 8182.9802 g/mol

mono isotopic mass: 8178.1713 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1636.65 (mono isotopic)

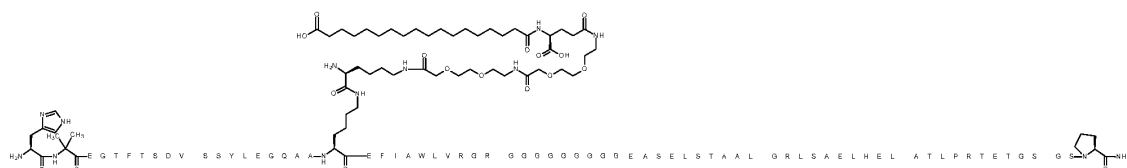
The amino acid sequence of

HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGQAPGQEPEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 168

- 10 Compound 0159

H-Aib-EGTFTSDVSSYLEGQAA-K([[(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl])-

- 15 E FIAWLVRGRGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₀ H₅₆₁ N₉₇ O₁₁₉

Molecular weight (average) calculated: 8031.7778 g/mol

mono isotopic mass: 8027.0828 g/mol

- 20 LCMS34: found (M + 5H)⁵⁺ 1607.2 (most abundant)

The amino acid sequence of

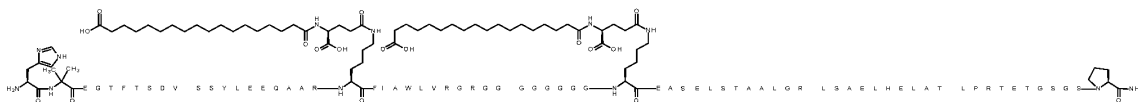
HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGGGGGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 126

- 25 Compound 0160

H-Aib-EGTFTSDVSSYLEEQAAAR-K([[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl])-FIAWLVRGRGGGGGGGGG-K([[(4S)-4-carboxy-4-

(17-carboxyheptadecanoylamino)butanoyl]-

EASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₃ H₅₈₄ N₉₈ O₁₁₈

5 Molecular weight (average) calculated: 8209.1069 g/mol

mono isotopic mass: 8204.2710 g/mol

LCMS34: found (M + 5H)⁵⁺ 1642.7 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIAWLVRGRGGGGGGGGKEASELSTAALGRLSAELHELA

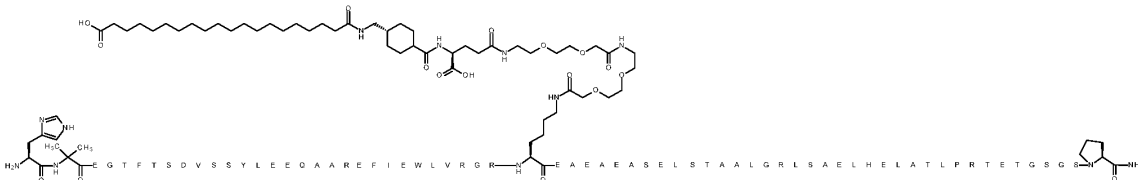
10 TLPRTETGSGSP has SEQ ID NO: 169

Compound 0179

H-Aib-EGTFTSDVSSYLEEQAAREFIEWLVRGR-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[(19-

15 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]-

EAEAEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₃ H₅₈₁ N₉₅ O₁₂₃

20 Molecular weight (average) calculated: 8244.0599 g/mol

mono isotopic mass: 8239.2129 g/mol

LCMS34: found (M + 5H)⁵⁺ 1648.86 (mono isotopic)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIEWLVRGRKEAEAEASELSTAALGRLSAELHELATLPRT

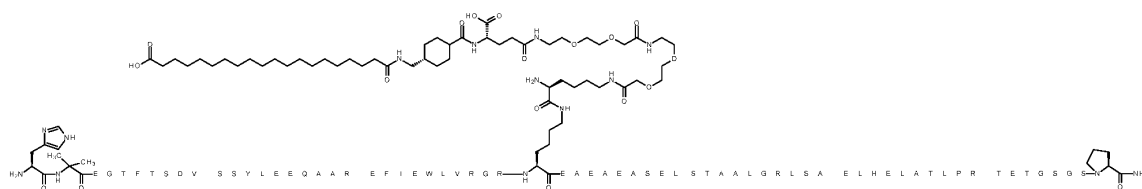
25 ETGSGSP has SEQ ID NO: 170

Compound 0180

H-Aib-EGTFTSDVSSYLEEQAAREFIEWLVRGR-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[(19-

30 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth

oxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl))-
EAEAEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₉ H₅₉₃ N₉₇ O₁₂₄

5 Molecular weight (average) calculated: 8372.2322 g/mol

mono isotopic mass: 8367.3078 g/mol

LCMS34: found (M + 5H)⁵⁺ 1674.46 (mono isotopic)

The amino acid sequence of

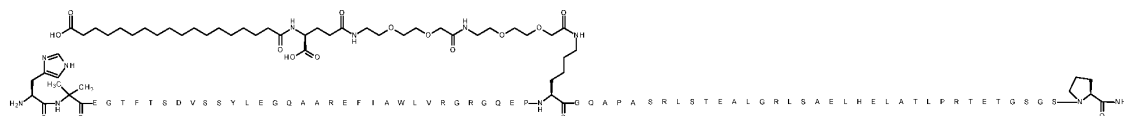
HXEGTFTSDVSSYLEEQAAREFIEWLVRGRKEAEAEASELSTAALGRLSAELHELATLPRRT

10 ETGSGSP has SEQ ID NO: 170

Compound 0191

H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRGRGQEP-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

15 carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GQAPASRLSTEALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₂ H₅₈₂ N₁₀₂ O₁₁₉

Molecular weight (average) calculated: 8267.1065 g/mol

20 mono isotopic mass: 8262.2625 g/mol

LCMS34: found (M + 5H)⁵⁺ 1653.45 (mono isotopic)

The amino acid sequence of

HXEGTFTSDVSSYLEGQAAREFIAWLVRGRGQEPKGQAPASRLSTEALGRLSAELHELAT

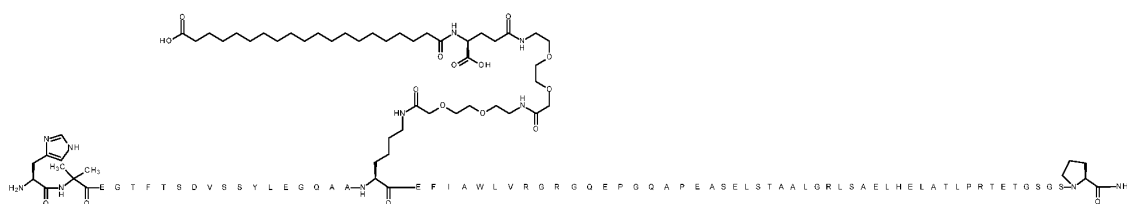
LPRRTETGSGSP has SEQ ID NO: 171

25

Compound 0202

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-

carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVRGRGQEPGQAPEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₀ H₅₇₄ N₉₆ O₁₂₁

Molecular weight (average) calculated: 8182.9802 g/mol

mono isotopic mass: 8178.1713 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1636.67 (mono isotopic)

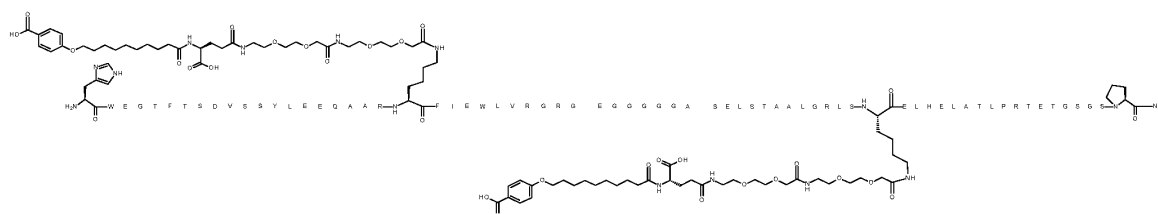
The amino acid sequence of

HXEGTFTSDVSSYLEGQAAKEFIAWLVRGRGQEPGQAPEASELSTAALGRLSAELHELAT
LPRTETGSGSP has SEQ ID NO: 121.

10 Compound 0231

HWEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIEWLVRGRGEGGGGGASELSTAALGRLS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-

- 15 carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-ELHELATLPRTETGSGSP-amide



C₃₈₇ H₆₀₂ N₁₀₀ O₁₃₁

Molecular weight (average) calculated: 8751.5122 g/mol

- 20 mono isotopic mass: 8746.3519 g/mol

LCMS34: found (M + 5H)⁵⁺ 1751.09 (most abundant)

The amino acid sequence of

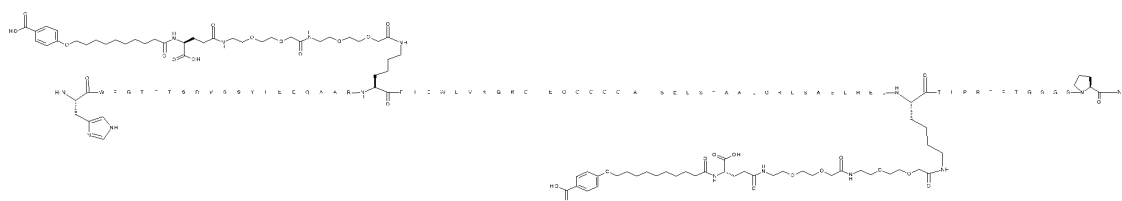
HWEGTFTSDVSSYLEEQAARKFIEWLVRGRGEGGGGGASELSTAALGRLSKELHELATLP
RTETGSGSP has SEQ ID NO: 172.

25

Compound 0232

HWEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-FIEWLVRGRGEGGGGGASELSTAALGRLSAELHEL-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-

- 5 carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-TLPRTETGSGSP-amide



C₃₈₇ H₆₀₂ N₁₀₀ O₁₃₁

Molecular weight (average) calculated: 8751.5122 g/mol

- 10 mono isotopic mass: 8746.3519 g/mol

LCMS34: found (M + 5H)⁵⁺ 1751.17 (most abundant)

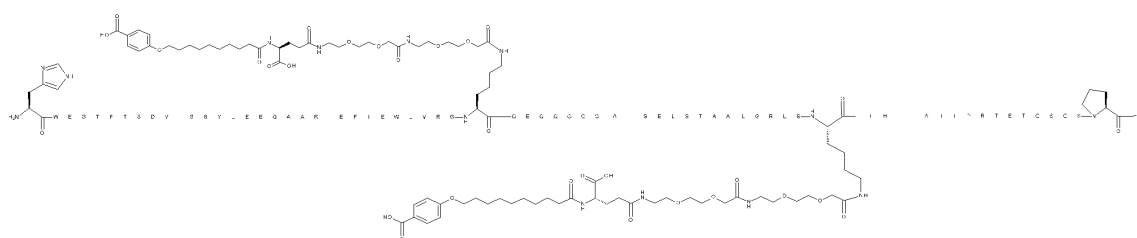
The amino acid sequence of

HWEGTFTSDVSSYLEEQAARKFIEWLVRGRGEGGGGGASELSTAALGRLSAELHELKTLP
RTETGSGSP has SEQ ID NO: 173.

- 15

Compound 0233

HWEGTFTSDVSSYLEEQAAREFIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GEGGGGGASELSTAALGRLS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-ELHELATLPRRTETGSGSP-amide



C₃₈₆ H₅₉₇ N₉₇ O₁₃₃

Molecular weight (average) calculated: 8724.4405 g/mol

- 25 mono isotopic mass: 8719.2933 g/mol

LCMS34: found (M + 5H)⁵⁺ 1745.54 (most abundant)

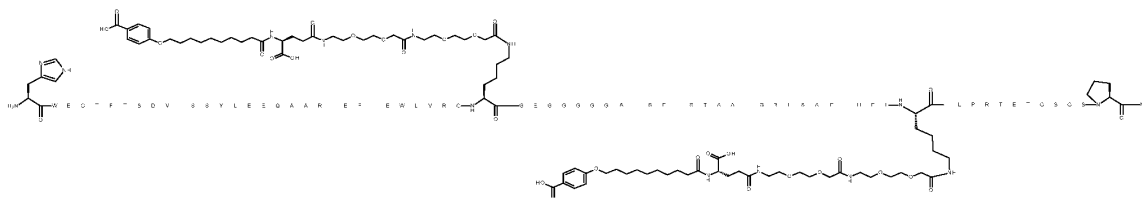
The amino acid sequence of

HWEGTFTSDVSSYLEEQAAREFIEWLVRGKGEGGGGGASELSTAALGRLSKELHELATLP
RTETGSGSP has SEQ ID NO: 174

5 Compound 0234

HWEGTFTSDVSSYLEEQAAREFIEWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GEGGGGGASELSTAALGRLSAELHEL-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(4-

10 carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-TLPRTETGSGSP-amide



C₃₈₆ H₅₉₇ N₉₇ O₁₃₃

Molecular weight (average) calculated: 8724.4405 g/mol

15 mono isotopic mass: 8719.2933 g/mol

LCMS34: found (M + 5H)⁵⁺ 1745.73 (most abundant)

The amino acid sequence of

HWEGTFTSDVSSYLEEQAAREFIEWLVRGKGEGGGGGASELSTAALGRLSAELHELKTLP
RTETGSGSP has SEQ ID NO: 175.

20

Compound 0235

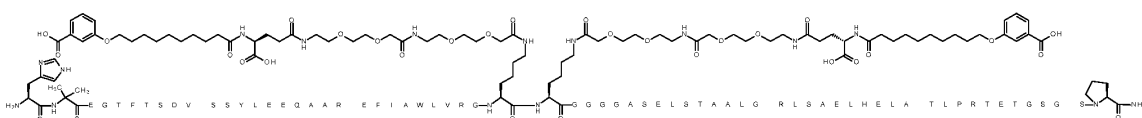
H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(3-

carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]

25 acetyl))-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[10-(3-

carboxyphenoxy)decanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]

acetyl))-GGGGASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₁ H₅₈₄ N₉₄ O₁₂₇

Molecular weight (average) calculated: 8393.1603 g/mol

mono isotopic mass: 8388.2129 g/mol

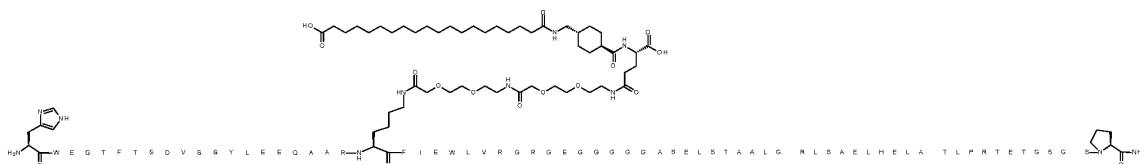
LCMS34: found (M + 5H)⁵⁺ 1678.65 (mono isotopic)

The amino acid sequence of

- 5 HXEGTFTSDVSSYLEEQAAREFIAWLVRGKKGGGGASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 176.

Compound 0254

- 10 HWEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRGRGEGGGGGASELSTAALGRLSAELHELATLPRTETGSGSP-amide

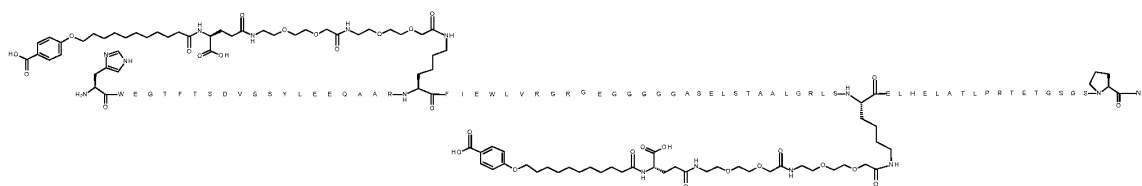


C₃₆₁ H₅₇₁ N₉₇ O₁₁₈

- 15 Molecular weight (average) calculated: 8157.9755 g/mol
mono isotopic mass: 8153.1662 g/mol
LCMS34: found (M + 5H)⁵⁺ 1632.55 (most abundant)
The amino acid sequence of
20 HWEGTFTSDVSSYLEEQAARKFIEWLVRGRGEGGGGGASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 177.

Compound 0255

- 25 HWEGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[11-(4-carboxyphenoxy)undecanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRGRGEGGGGGASELSTAALGRLS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[11-(4-carboxyphenoxy)undecanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-ELHELATLPRTETGSGSP-amide



C₃₈₉ H₆₀₆ N₁₀₀ O₁₃₁

Molecular weight (average) calculated: 8779.5653 g/mol

mono isotopic mass: 8774.3832 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1756.92 (most abundant)

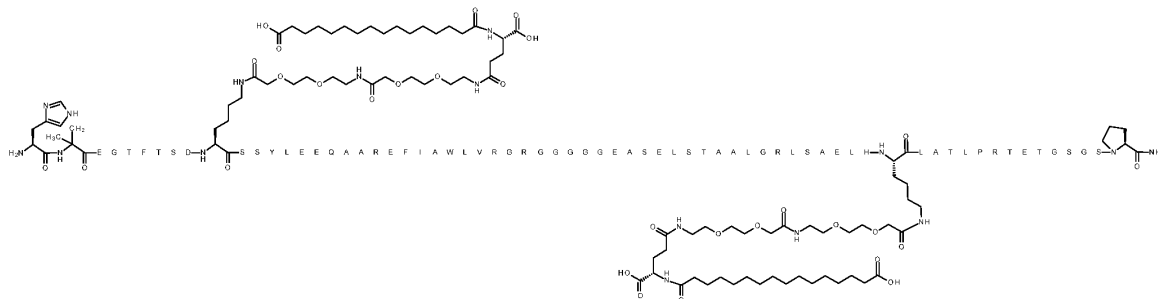
The amino acid sequence of

HWEGTFTSDVSSYLEEQAARKFIEWLVRGRGEGGGGASELSTAALGRLSKELHELATLP
RTETGSGSP has SEQ ID NO: 172.

10 Compound 0259

H-Aib-EGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-
carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl)]-SSYLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELH-K([2-[2-[2-[[2-[2-[2-
[[[(4S)-4-carboxy-4-(15-

- 15 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl)]-LATLPRTETGSGSP-amide



C₃₇₂ H₆₀₂ N₉₈ O₁₂₆

Molecular weight (average) calculated: 8463.3413 g/mol

- 20 mono isotopic mass: 8458.3711 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1693.6 (most abundant)

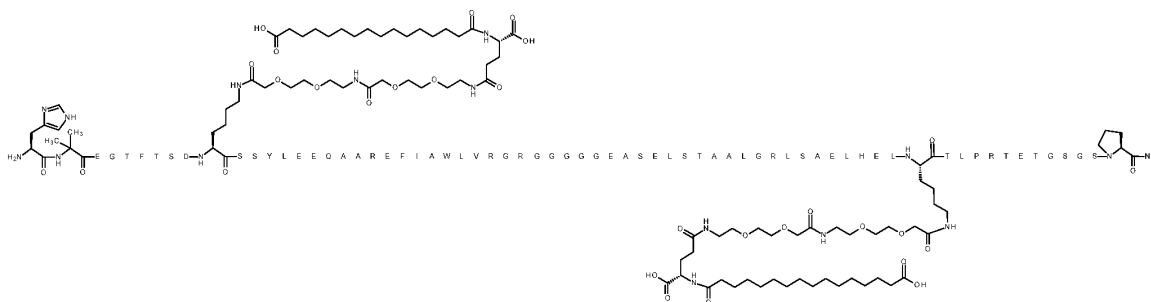
The amino acid sequence of

HXEGTFTSDKSSYLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHKLATLP
TETGSGSP has SEQ ID NO: 178.

25

Compound 0260

H-Aib-EGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-SSYLEEQAAAREFIAWLVGRGGGGGEASELSTAALGRLSAELHEL-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-TLPRTETGSGSP-amide



C₃₇₄ H₆₀₄ N₉₈ O₁₂₈

10 Molecular weight (average) calculated: 8521.3774 g/mol

mono isotopic mass: 8516.3766 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1705.3 (most abundant)

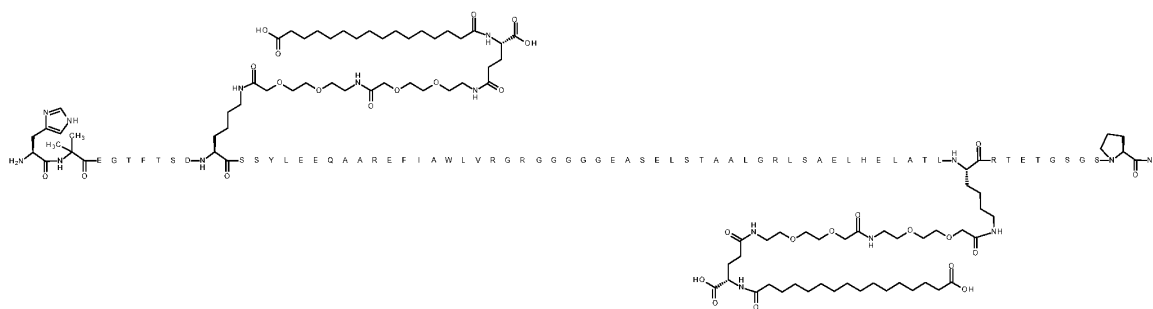
The amino acid sequence of

HXEGTFTSDKSSYLEEQAAAREFIAWLVGRGGGGGEASELSTAALGRLSAELHELKTLPR

15 TETGSGSP has SEQ ID NO: 179.

Compound 0261

H-Aib-EGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-SSYLEEQAAAREFIAWLVGRGGGGGEASELSTAALGRLSAELHELATL-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-RTETGSGSP-amide



C₃₇₂ H₆₀₂ N₉₈ O₁₂₈

Molecular weight (average) calculated: 8495.3401 g/mol

mono isotopic mass: 8490.3610 g/mol

- 5 LCMS_ZQ: found (M + 5H)⁵⁺ 1699.9 (most abundant)

The amino acid sequence of

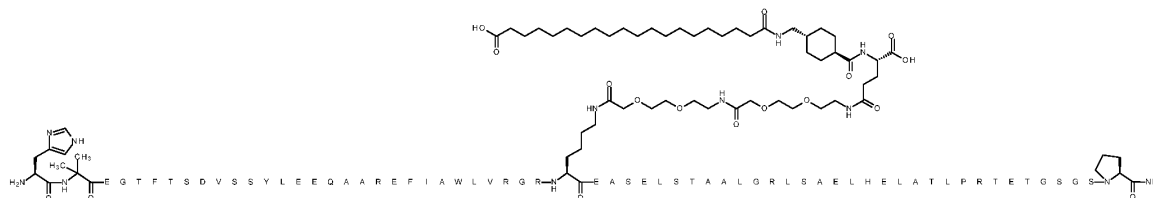
HXEGTFTSDKSSYLEEQAAAREFIAWLVGRGSGGGGGEASELSTAALGRLSAELHELATLKR

TETGSGSP has SEQ ID NO: 180.

10 Compound 0263

H-Aib-EGTFTSDVSSYLEEQAAAREFIAWLVGRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-[[4-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-EASELSTAALGRLSAELHELATLPRRTETGSGSP-

- 15 amide



C₃₄₅ H₅₅₅ N₉₁ O₁₁₃

Molecular weight (average) calculated: 7785.6401 g/mol

mono isotopic mass: 7781.0480 g/mol

- 20 LCMS_ZQ: found (M + 5H)⁵⁺ 1558.2 (most abundant)

The amino acid sequence of

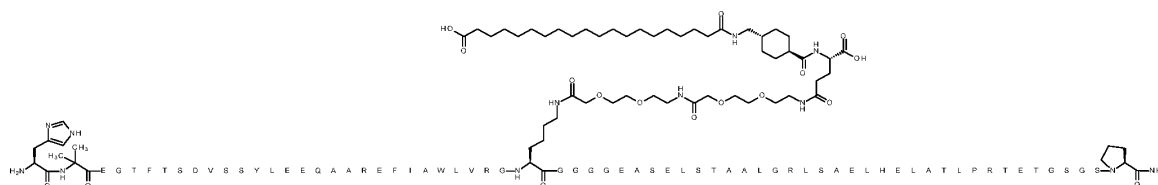
HXEGTFTSDVSSYLEEQAAAREFIAWLVGRGKEASELSTAALGRLSAELHELATLPRRTETGS

GSP has SEQ ID NO: 150.

25 Compound 0264

H-Aib-EGTFTSDVSSYLEEQAAAREFIAWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

5 GGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₇ H₅₅₅ N₉₁ O₁₁₆

Molecular weight (average) calculated: 7857.6597 g/mol

mono isotopic mass: 7853.0327 g/mol

10 LCMS_ZQ: found (M + 5H)⁵⁺ 1572.5 (most abundant)

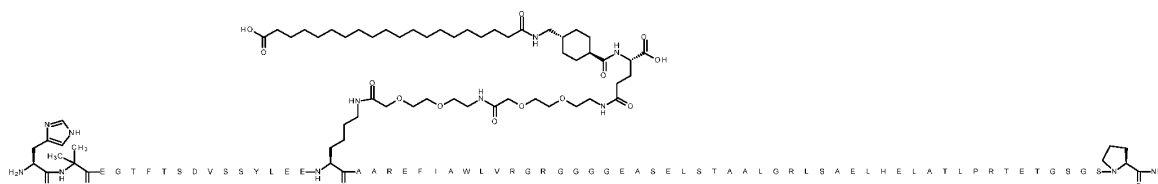
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAAREFIAWLVRGKGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 181.

15 Compound 0265

H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

AAREFIAWLVRGRGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



20

C₃₄₈ H₅₅₉ N₉₃ O₁₁₅

Molecular weight (average) calculated: 7885.7162 g/mol

mono isotopic mass: 7881.0752 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1577.9 (most abundant)

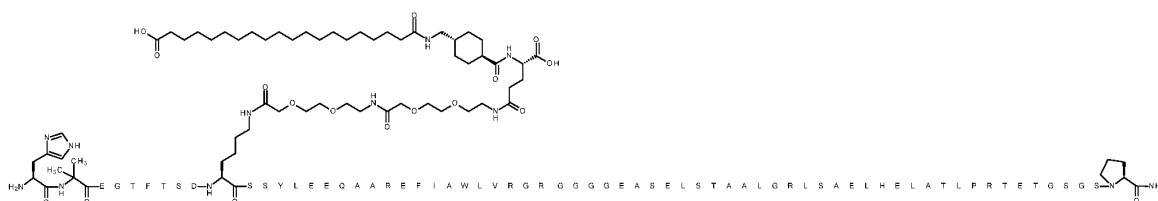
25 The amino acid sequence of

HXEGTFTSDVSSYLEEKAAREFIAWLVRGRGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 182.

Compound 0266

H-Aib-EGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

- 5 SSYLEEQAAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₈ H₅₅₈ N₉₄ O₁₁₆

Molecular weight (average) calculated: 7914.7143 g/mol

- 10 mono isotopic mass: 7910.0654 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1583.9 (most abundant)

The amino acid sequence of

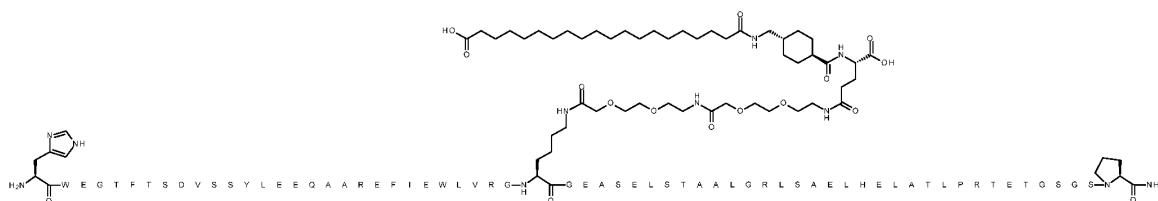
HXEGTFTSDKSSYLEEQAAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 183.

- 15

Compound 0267

HWEGTFTSDVSSYLEEQAAAREFIEWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GEASELSTAALGRLSAELHELATLPRTETGSGSP-

- 20 amide



C₃₅₀ H₅₅₁ N₈₉ O₁₁₅

Molecular weight (average) calculated: 7845.6472 g/mol

mono isotopic mass: 7841.0003 g/mol

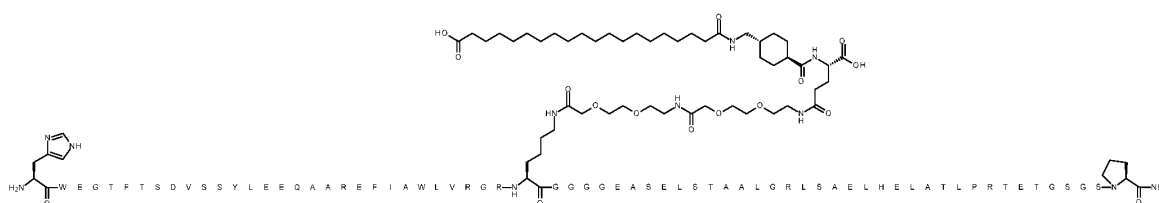
- 25 LCMS_ZQ: found (M + 5H)⁵⁺ 1570.1 (most abundant)

The amino acid sequence of
HWEGTFTSDVSSYLEEQAAREFIEWLVRGKGEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 184.

5 Compound 0268

HWEGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-
[(19-
carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth
oxy]acetyl]amino]ethoxy]ethoxy]acetyl))-

10 GGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide


$$\text{C}_{360} \text{H}_{570} \text{N}_{96} \text{O}_{117}$$

Molecular weight (average) calculated: 8114.9508 g/mol

mono isotopic mass: 8110.1604 g/mol

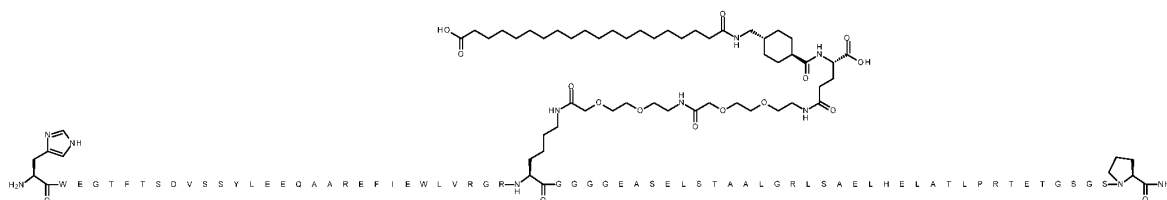
15 LCMS ZQ: found (M + 5H)⁵⁺ 1624.0 (most abundant)

The amino acid sequence of
HWEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 185.

20 Compound 0269

HWEGTFTSDVSSYLEEQAAREFIEWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-
[(19-
carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth
oxy]acetyl]amino]ethoxy]ethoxy]acetyl))-

25 GGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide


$$\text{C}_{362} \text{H}_{572} \text{N}_{96} \text{O}_{119}$$

Molecular weight (average) calculated: 8172.9869 g/mol

mono isotopic mass: 8168.1658 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1634.5 (mono isotopic)

The amino acid sequence of

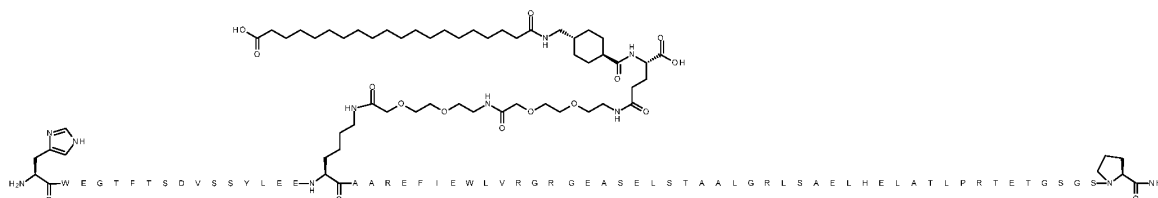
5 HWEGTFTSDVSSYLEEQAAREFIEWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 186.

Compound 0270

HWEGTFTSDVSSYLEE-K[2-2-2-2-2-2-[(4S)-4-carboxy-4-[[4-(19-

10 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-

AAREFIEWLVVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide


$$\text{C}_{351} \text{H}_{555} \text{N}_{91} \text{O}_{114}$$

15 Molecular weight (average) calculated: 7873.7037 g/mol

mono isotopic mass: 7869.0429 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1575.4 (most abundant)

The amino acid sequence of

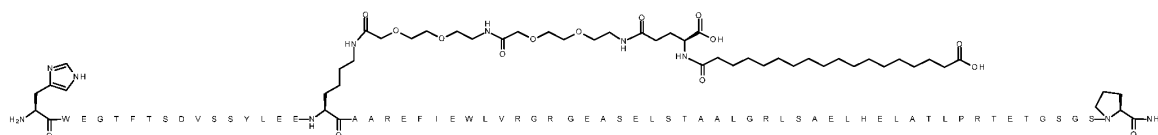
HWEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

20 GSP has SEQ ID NO: 187.

Compound 0271

HWEGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[[(4S)-4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
25 ty]) -AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide


$$\text{C}_{341} \text{H}_{538} \text{N}_{90} \text{O}_{113}$$

Molecular weight (average) calculated: 7706.4556 g/mol

mono isotopic mass: 7701.9119 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1542.27 (most abundant)

The amino acid sequence of

HWEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

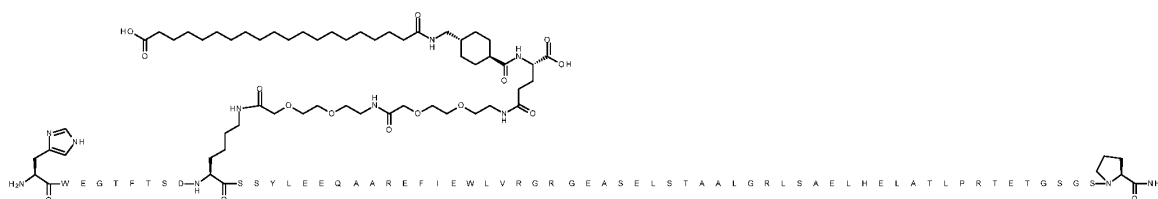
GSP has SEQ ID NO: 187.

5

Compound 0272

HWEGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

10 SSYLEEQAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₁ H₅₅₄ N₉₂ O₁₁₅

Molecular weight (average) calculated: 7902.7019 g/mol

mono isotopic mass: 7898.0330 g/mol

15 LCMS_ZQ: found (M + 5H)⁵⁺ 1581.4 (most abundant)

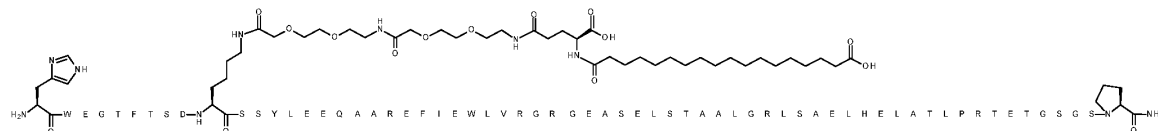
The amino acid sequence of

HWEGTFTSDKSSYLEEQAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETG

SGSP has SEQ ID NO: 188.

20 Compound 0273

HWEGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-SSYLEEQAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



25

C₃₄₁ H₅₃₇ N₉₁ O₁₁₄

Molecular weight (average) calculated: 7735.4538 g/mol

mono isotopic mass: 7730.9020 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1547.9 (most abundant)

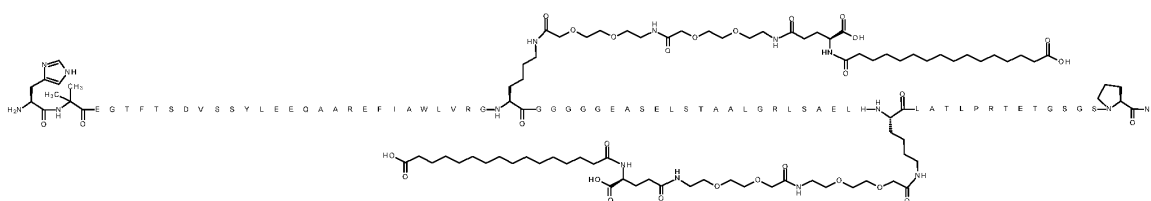
The amino acid sequence of

HWEGTFTSDKSSYLEEQAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETG
SGSP has SEQ ID NO: 188.

5 Compound 0280

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GGGGGEASELSTAALGRLSAELH-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]

10 tyI]-LATLPRTETGSGSP-amide


$$\text{C}_{371} \text{H}_{599} \text{N}_{95} \text{O}_{126}$$

Molecular weight (average) calculated: 8406.2867 g/mol

mono isotopic mass: 8401.3384 g/mol

15 LCMS34: found (M + 5H)⁵⁺ 1682.1 (most abundant)

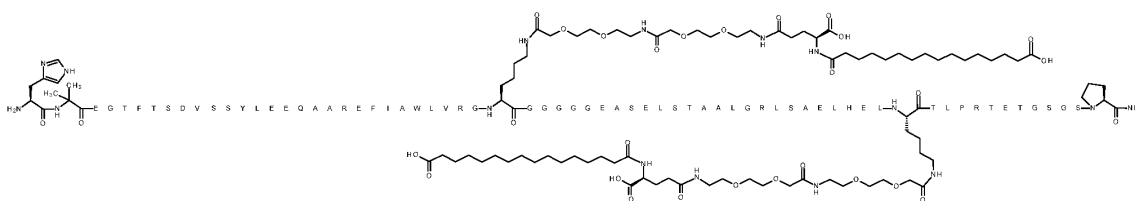
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGKGGGGGEASELSTAALGRLSAELHKLATLPR
 TETGSGSP has SEQ ID NO: 189.

20 Compound 0281

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRG-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GGGGGEASELSTAALGRLSAELHEL-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]

25 tyI)]-TLPRTETGSGSP-amide


$$\text{C}_{373} \text{H}_{601} \text{N}_{95} \text{O}_{128}$$

Molecular weight (average) calculated: 8464.3227 g/mol

mono isotopic mass: 8459.3439 g/mol

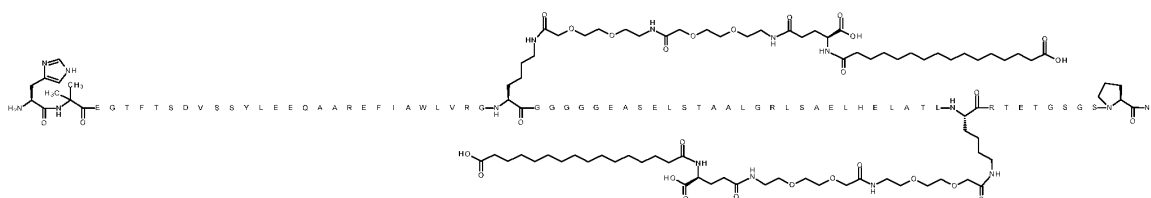
LCMS34: found (M + 5H)⁵⁺ 1693.7 (most abundant)

The amino acid sequence of

- 5 HXEGTFTSDVSSYLEEQAAREFIAWLVRGKGGGGGEASELSTAALGRLSAELHELKTLPR
TETGSGSP has SEQ ID NO: 190.

Compound 0284

- 10 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGGGGEASELSTAALGRLSAELHELATL-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-RTETGSGSP-amide



- 15 C₃₇₁ H₅₉₉ N₉₅ O₁₂₈

Molecular weight (average) calculated: 8438.2855 g/mol

mono isotopic mass: 8433.3283 g/mol

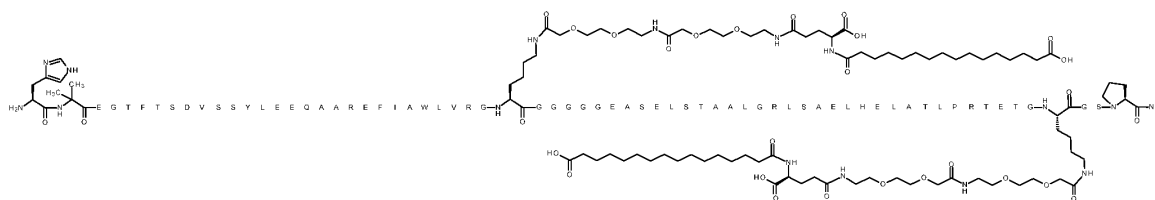
LCMS34: found (M + 5H)⁵⁺ 1688.7 (most abundant)

The amino acid sequence of

- 20 HXEGTFTSDVSSYLEEQAAREFIAWLVRGKGGGGGEASELSTAALGRLSAELHELATLKR
TETGSGSP has SEQ ID NO: 191.

Compound 0285

- 25 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGGGGEASELSTAALGRLSAELHELATLPR TETG-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GSP-amide



C₃₇₃ H₆₀₁ N₉₅ O₁₂₇

Molecular weight (average) calculated: 8448.3233 g/mol

mono isotopic mass: 8443.3490 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1690.7 (most abundant)

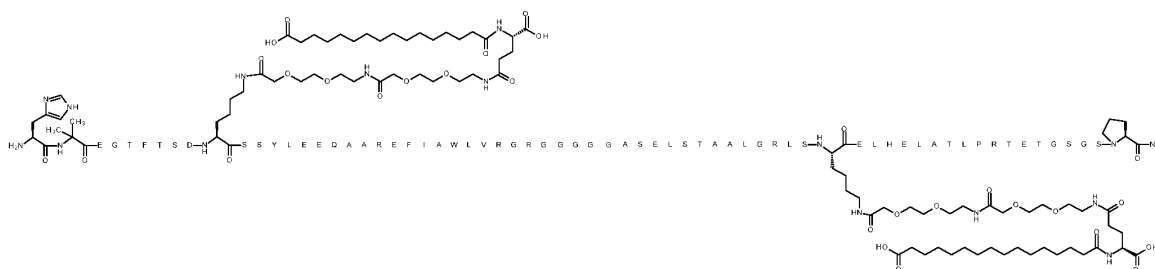
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGKGGGGGEASELSTAALGRLSAELHELATLPRTET
TETGKGSP has SEQ ID NO: 192.

10 Compound 0292

H-Aib-EGTFTSD-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-
carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl])-SSYLEEQAAREFIAWLVRGRGGGGGASELSTAALGRLS-K([2-[2-[2-[[2-[2-[2-[(4S)-4-
carboxy-4-(15-

- 15 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl])-ELHELATLPRTETGSGSP-amide



C₃₆₉ H₅₉₇ N₉₇ O₁₂₅

Molecular weight (average) calculated: 8392.2634 g/mol

- 20 mono isotopic mass: 8387.3340 g/mol

LCMS34: found (M + 5H)⁵⁺ 1679.24 (most abundant)

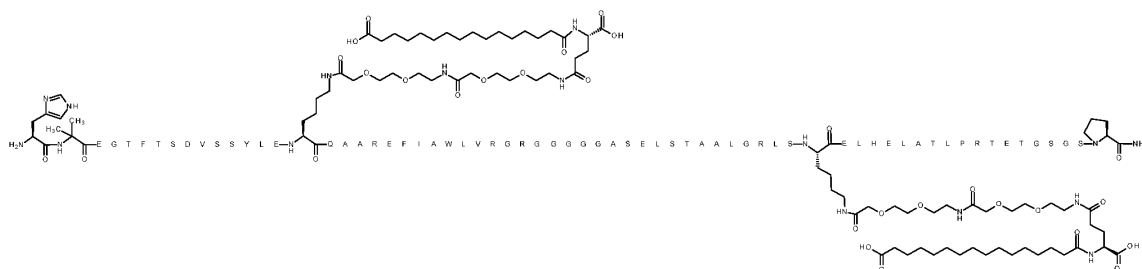
The amino acid sequence of

HXEGTFTSDKSSYLEEQAAREFIAWLVRGRGGGGGASELSTAALGRLSKELHELATLPRT
ETGSGSP has SEQ ID NO: 193.

- 25

Compound 0294

H-Aib-EGTFTSDVSSYLE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-QAAREFIAWLVRGRGGGGGASELSTAALGRLS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-ELHELATLPRRTETGSGSP-amide



C₃₆₉ H₅₉₉ N₉₇ O₁₂₃

Molecular weight (average) calculated: 8362.2805 g/mol

10 mono isotopic mass: 8357.3598 g/mol

LCMS34: found (M + 5H)⁵⁺ 1673.26 (most abundant)

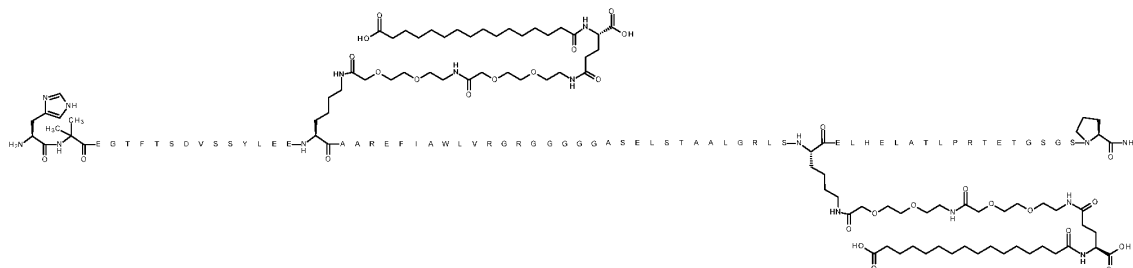
The amino acid sequence of

HXEGTFTSDVSSYLEKQAAREFIAWLVRGRGGGGGASELSTAALGRLSKELHELATLPRRTETGSGSP has SEQ ID NO: 194.

15

Compound 0295

H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-AAREFIAWLVRGRGGGGGASELSTAALGRLS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-ELHELATLPRRTETGSGSP-amide



C₃₆₉ H₅₉₈ N₉₆ O₁₂₄

Molecular weight (average) calculated: 8363.2652 g/mol

mono isotopic mass: 8358.3439 g/mol

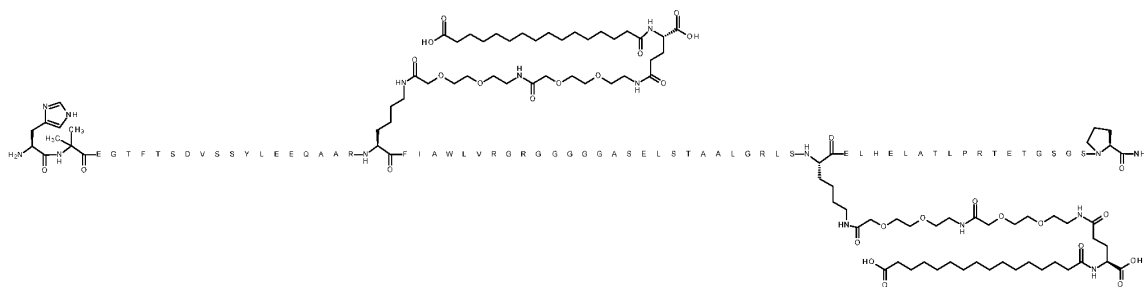
LCMS34: found (M + 5H)⁵⁺ 1673.65 (most abundant)

The amino acid sequence of

- 5 HXEGTFTSDVSSYLEEKAAREFIAWLVRGRGGGGGASELSTAALGRLSKELHELATLPRT
ETGSGSP has SEQ ID NO: 195.

Compound 0296

- 10 H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-FIAWLVRGRGGGGGASELSTAALGRLS-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-ELHELATLPRTETGSGSP-amide



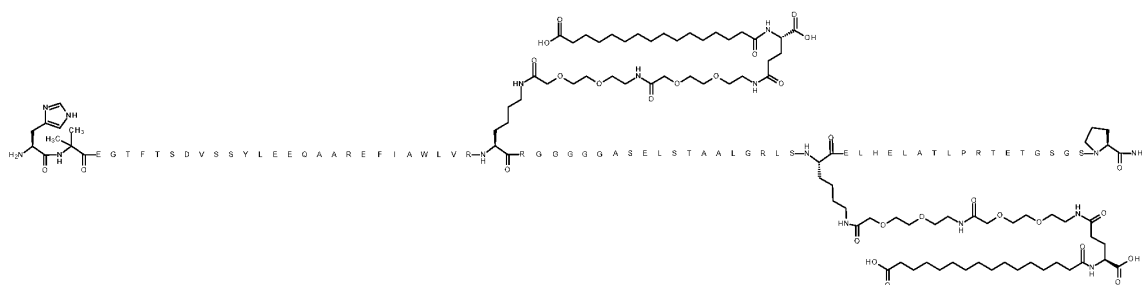
- 15 C₃₆₉ H₅₉₉ N₉₇ O₁₂₃
Molecular weight (average) calculated: 8362.2805 g/mol
mono isotopic mass: 8357.3598 g/mol
LCMS34: found (M + 5H)⁵⁺ 1673.46 (most abundant)

The amino acid sequence of

- 20 HXEGTFTSDVSSYLEEQAARKFIAWLVRGRGGGGGASELSTAALGRLSKELHELATLPRT
ETGSGSP has SEQ ID NO: 196.

Compound 0297

- 25 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-RGGGGGASELSTAALGRLS-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-ELHELATLPRTETGSGSP-amide



C₃₇₂ H₆₀₃ N₉₇ O₁₂₅

Molecular weight (average) calculated: 8434.3431 g/mol

mono isotopic mass: 8429.3810 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1687.87 (most abundant)

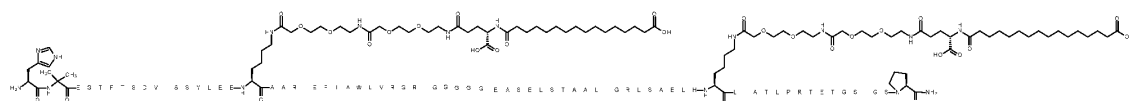
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAAREFIAWLVRKRGGGGGASELSTAALGRLSKELHELATLPRT
ETGSGSP has SEQ ID NO: 197.

10 Compound 0299

H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-
carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl])-AAREFIAWLVRGRGGGGGEASELSTAALGRLSAELH-K([2-[2-[2-[2-[2-[2-[(4S)-4-
carboxy-4-(15-

- 15 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl])-LATLPRTETGSGSP-amide



C₃₇₂ H₆₀₃ N₉₇ O₁₂₅

Molecular weight (average) calculated: 8434.3431 g/mol

- 20 mono isotopic mass: 8429.3810 g/mol

LCMS34: found (M + 5H)⁵⁺ 1688 (most abundant)

The amino acid sequence of

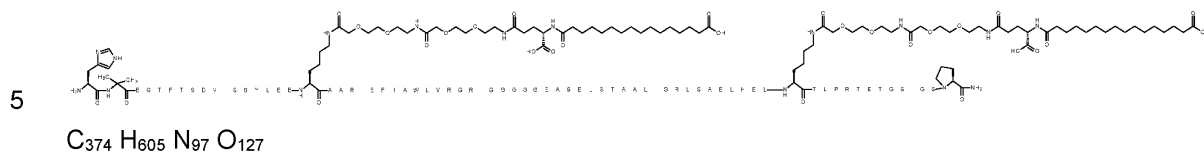
HXEGTFTSDVSSYLEEKAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHKLATLPRT
TETGSGSP has SEQ ID NO: 198.

25

Compound 0396

H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-
carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace

tyl))-AAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHEL-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-TLPRTETGSGSP-amide



Molecular weight (average) calculated: 8492.3792 g/mol

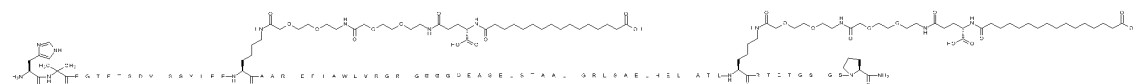
mono isotopic mass: 8487.3865 g/mol

LCMS34: found (M + 5H)⁵⁺ 1699 (most abundant)

- 10 The amino acid sequence of
HXEGTFTSDVSSYLEEKAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELKTLPR
TETGSGSP has SEQ ID NO: 199.

Compound 0397

- 15 H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-AAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATL-K([2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-RTETGSGSP-amide
- 20



C₃₇₂ H₆₀₃ N₉₇ O₁₂₇

Molecular weight (average) calculated: 8466.3419 g/mol

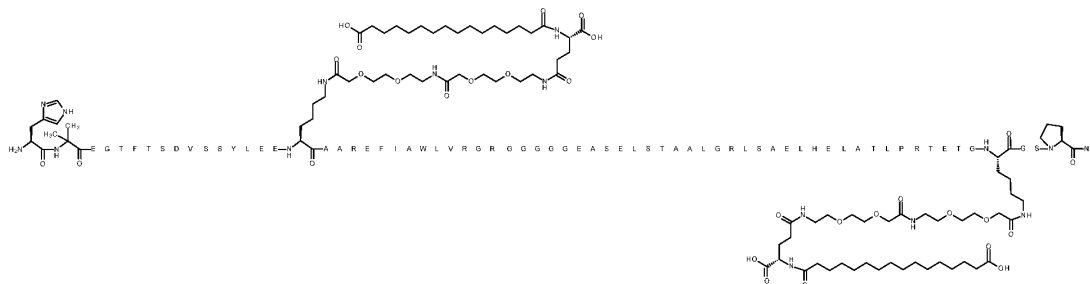
mono isotopic mass: 8461.3708 g/mol

- 25 LCMS34: found (M + 5H)⁵⁺ 1694 (most abundant)
The amino acid sequence of
HXEGTFTSDVSSYLEEKAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLKR
TETGSGSP has SEQ ID NO: 200.

Compound 0411

H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-TLPRTETGSGSP-amide

tyl))-AAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETG-K([2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-GSP-amide



5

C₃₇₄ H₆₀₅ N₉₇ O₁₂₆

Molecular weight (average) calculated: 8476.3798 g/mol

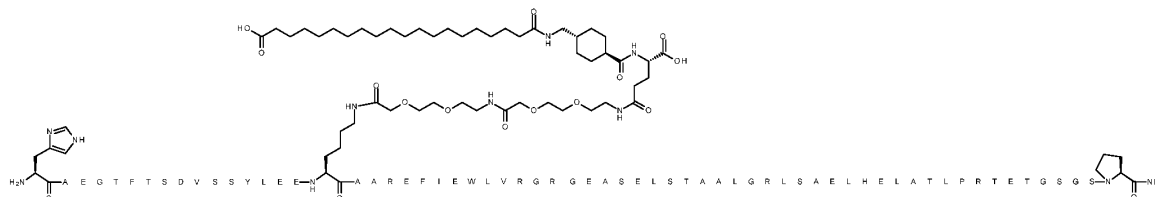
mono isotopic mass: 8471.3915 g/mol

LCMS34: found (M + 5H)⁵⁺ 1696 (most abundant)

- 10 The amino acid sequence of
HXEGTFTSDVSSYLEEKAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGKGSP has SEQ ID NO: 201.

Compound 0414

- 15 HAEGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



20

C₃₄₃ H₅₅₀ N₉₀ O₁₁₄

Molecular weight (average) calculated: 7758.5717 g/mol

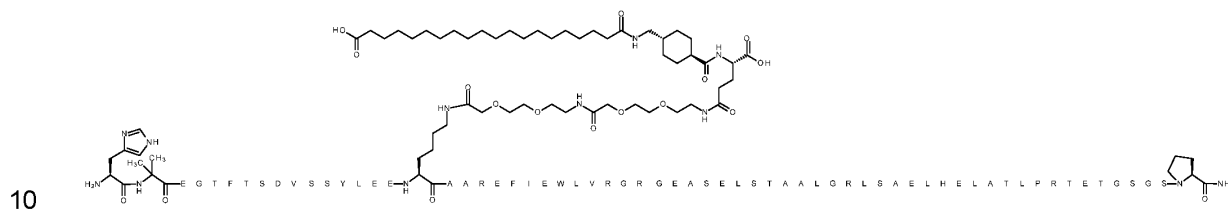
mono isotopic mass: 7754.0007 g/mol

LCMS34: found (M + 5H)⁵⁺ 1551.88 (mono isotopic)

The amino acid sequence of
HAEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 202.

5 Compound 0415

H-Aib-EGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-
AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₄ H₅₅₂ N₉₀ O₁₁₄

Molecular weight (average) calculated: 7772.5983 g/mol

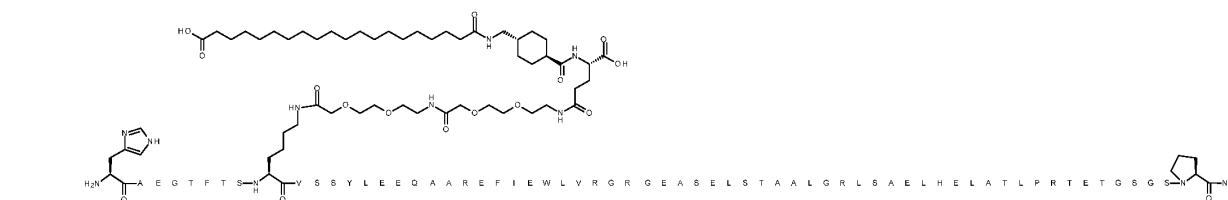
mono isotopic mass: 7768.0163 g/mol

LCMS34: found (M + 5H)⁵⁺ 1554.67 (mono isotopic)

15 The amino acid sequence of
HXEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 203.

Compound 0416

20 HAEGTFTS-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-
VSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₄ H₅₅₃ N₉₁ O₁₁₃

Molecular weight (average) calculated: 7771.6135 g/mol

mono isotopic mass: 7767.0323 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1555.3 (most abundant)

The amino acid sequence of

HAEGTFTSKVSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

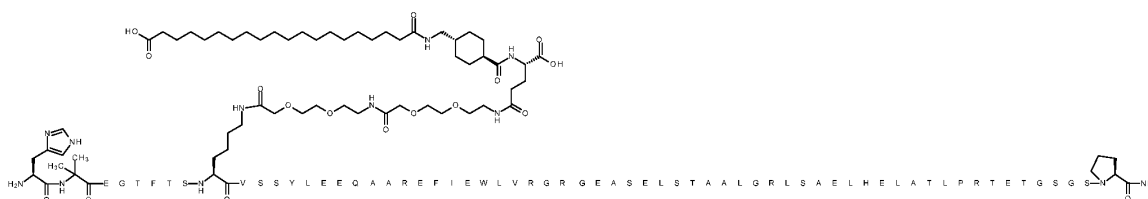
GSP has SEQ ID NO: 204.

5

Compound 0417

H-Aib-EGTFTS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

10 VSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₅ H₅₅₅ N₉₁ O₁₁₃

Molecular weight (average) calculated: 7785.6401 g/mol

mono isotopic mass: 7781.0480 g/mol

15 LCMS_ZQ: found (M + 5H)⁵⁺ 1558.1 (most abundant)

The amino acid sequence of

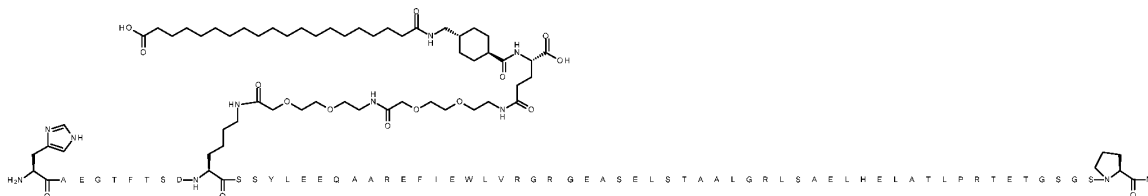
HXEGTFTSKVSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 205.

20 Compound 0431

HAEGTFTSD-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

SSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



25

C₃₄₃ H₅₄₉ N₉₁ O₁₁₅

Molecular weight (average) calculated: 7787.5699 g/mol

mono isotopic mass: 7782.9908 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1558.67 (most abundant)

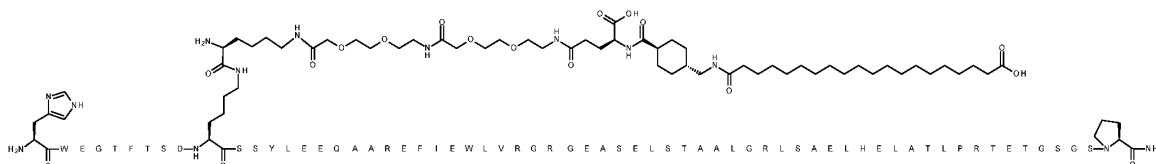
The amino acid sequence of

HAEGTFTSDKSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGS

5 GSP has SEQ ID NO: 206.

Compound 0433

HWEGTFTSD-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-SSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₇ H₅₆₆ N₉₄ O₁₁₆

Molecular weight (average) calculated: 8030.8741 g/mol

15 mono isotopic mass: 8026.1280 g/mol

LCMS34: found (M + 5H)⁵⁺ 1607.09 (most abundant)

The amino acid sequence of

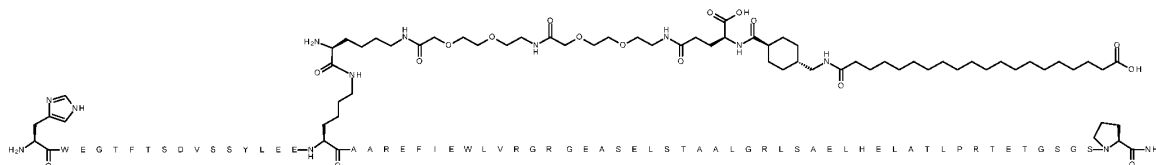
HWEGTFTSDKSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETG

SGSP has SEQ ID NO: 188.

20

Compound 0434

HWEGTFTSDVSSYLEE-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₇ H₅₆₇ N₉₃ O₁₁₅

Molecular weight (average) calculated: 8001.8760 g/mol

mono isotopic mass: 7997.1378 g/mol

LCMS34: found (M + 5H)⁵⁺ 1601.19 (most abundant)

The amino acid sequence of

HWEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 187.

5

Compound 0435

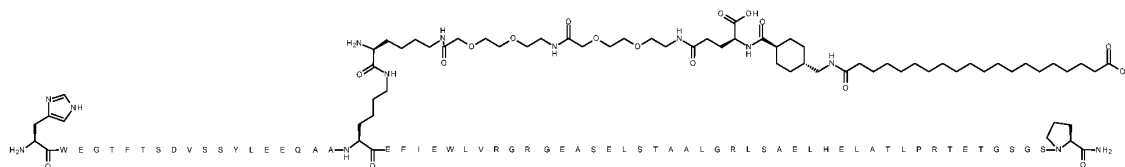
HWEGTFTSDVSSYLEEQAA-K([[(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-

[(19-

carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth

10 oxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl])-

EFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₆ H₅₆₃ N₉₁ O₁₁₆

Molecular weight (average) calculated: 7973.8195 g/mol

15 mono isotopic mass: 7969.0953 g/mol

LCMS34: found (M + 5H)⁵⁺ 1595.67 (most abundant)

The amino acid sequence of

HWEGTFTSDVSSYLEEQAAKEFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 207.

20

Compound 0436

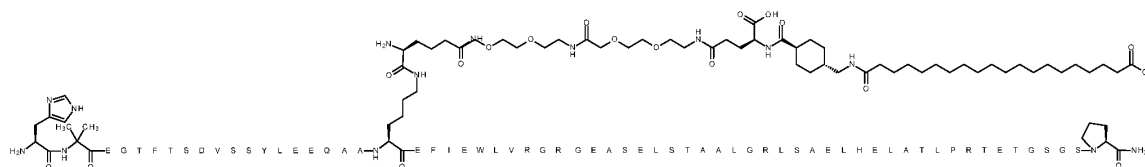
H-Aib-EGTFTSDVSSYLEEQAA-K([[(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-

[(19-

carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth

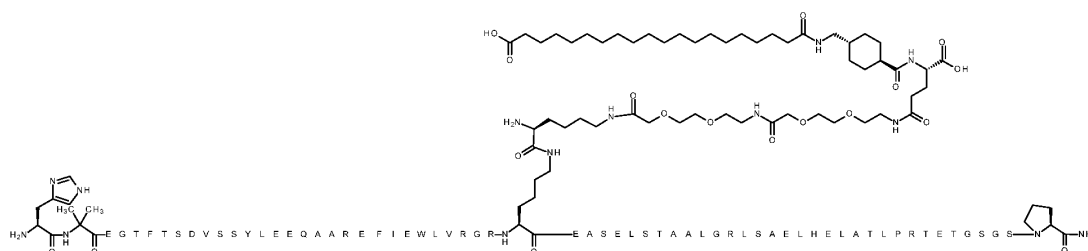
25 oxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl])-

EFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₉ H₅₆₀ N₉₀ O₁₁₆

Molecular weight (average) calculated: 7872.7141 g/mol



C₃₅₃ H₅₆₉ N₉₃ O₁₁₆

Molecular weight (average) calculated: 7971.8485 g/mol

mono isotopic mass: 7967.1484 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1595.27 (most abundant)

The amino acid sequence of

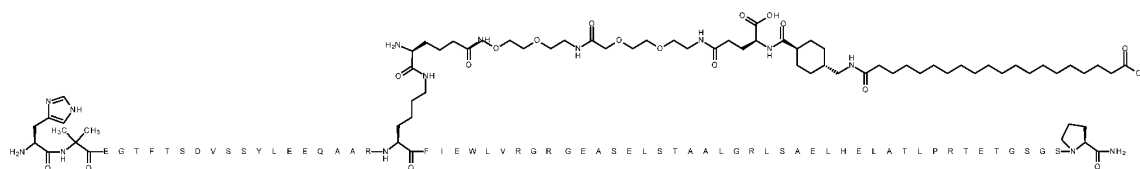
HXEGTFTSDVSSYLEEQAAREFIEWLVRGRKEASELSTAALGRLSAELHELATLPRRTETGS

GSP has SEQ ID NO: 210

10 Compound 0439

H-Aib-EGTFTSDVSSYLEEQAAR-K([[(2S)-2-amino-6-[[2-[2-[2-[2-[2-[[4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-FIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide

- 15 FIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₀ H₅₆₅ N₉₃ O₁₁₄

Molecular weight (average) calculated: 7899.7858 g/mol

mono isotopic mass: 7895.1273 g/mol

- 20 LCMS34: found (M + 5H)⁵⁺ 1580.88 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGS

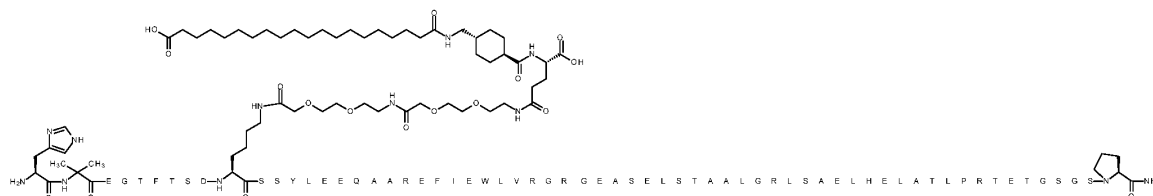
GSP has SEQ ID NO: 211.

25 Compound 0440

H-Aib-EGTFTSD-K([2-[2-[2-[[2-[2-[2-[[4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-FIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide

oxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

SSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₄ H₅₅₁ N₉₁ O₁₁₅

5 Molecular weight (average) calculated: 7801.5964 g/mol

mono isotopic mass: 7797.0065 g/mol

LCMS34: found (M + 5H)⁵⁺ 1560.48 (mono isotopic)

The amino acid sequence of

HXEGTFTSDKSSYLEEQAAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

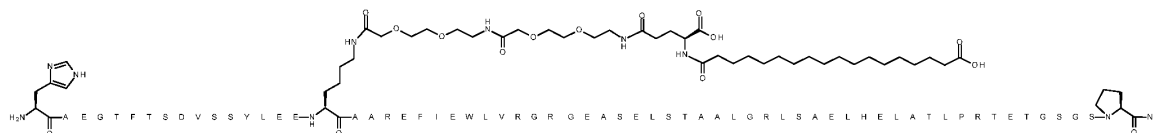
10 GSP has SEQ ID NO: 212.

Compound 0472

HAEGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace

15 tyl])-AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₃₃ H₅₃₃ N₈₉ O₁₁₃

Molecular weight (average) calculated: 7591.3236 g/mol

mono isotopic mass: 7586.8697 g/mol

20 LCMS34: found (M + 5H)⁵⁺ 1519.18 (most abundant)

The amino acid sequence of

HAEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

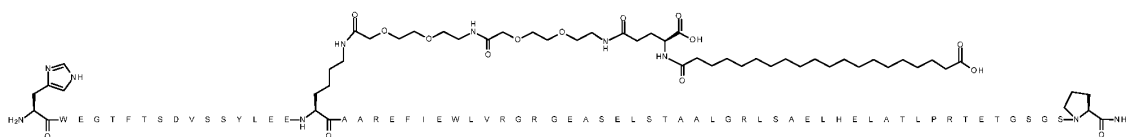
GSP has SEQ ID NO: 202.

Compound 0473

HWEGTFTSDVSSYLEE-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-

carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acet

yl])-AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide


$$\text{C}_{343} \text{H}_{542} \text{N}_{90} \text{O}_{113}$$

Molecular weight (average) calculated: 7734.5088 g/mol

mono isotopic mass: 7729.9432 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1547.82 (most abundant)

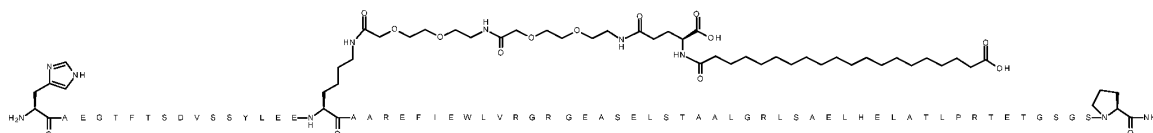
The amino acid sequence of

HWEGTFTSDVSSYLEEKAAREFIEWLVGRGRGEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 187.

- 10 Compound 0474

HAEGTFTSDVSSYLEE-K((2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



- 15 C₃₃₅ H₅₃₇ N₈₉ O₁₁₃

Molecular weight (average) calculated: 7619.3768 g/mol

mono isotopic mass: 7614.9010 g/mol

LCMS34: found (M + 5H)⁵⁺ 1524.8 (most abundant)

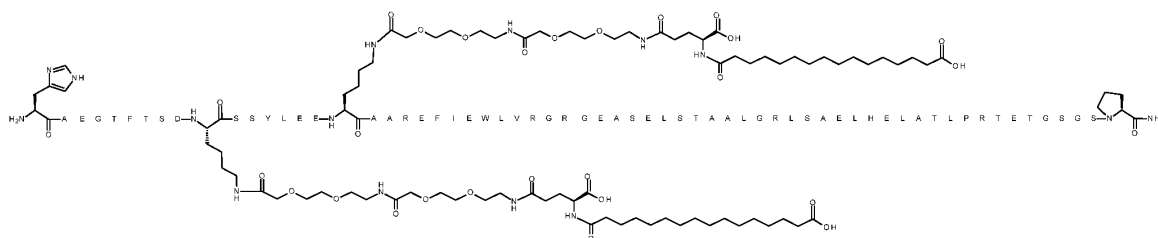
The amino acid sequence of

- 20 HAEGTFTSDVSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 202.

Compound 0475

HAEGTFTSD-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-

- 25 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl)]-SSYLEE-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-
carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl)]-AAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₅ H₅₈₉ N₉₃ O₁₂₅

Molecular weight (average) calculated: 8280.1303 g/mol

mono isotopic mass: 8275.2591 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1656.86 (most abundant)

The amino acid sequence of

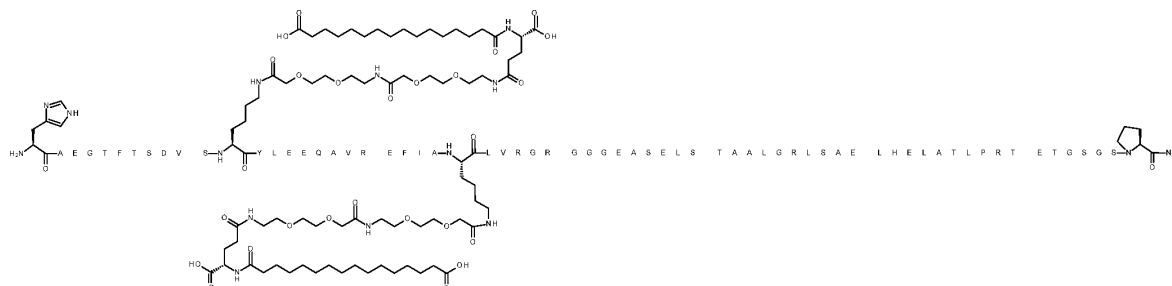
HAEGTFTSDKSSYLEEKAAREFIEWLVRGRGEASELSTAALGRLSAELHELATLPRTETGS

GSP has SEQ ID NO: 213.

- 10 Compound 0482

HAEGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)-YLEEQAVREFIA-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)-LVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide

- 15



C₃₆₅ H₅₉₉ N₉₅ O₁₂₅

Molecular weight (average) calculated: 8318.2231 g/mol

mono isotopic mass: 8313.3435 g/mol

- 20 LCMS_ZQ: found (M + 5H)⁵⁺ 1664.64 (most abundant)

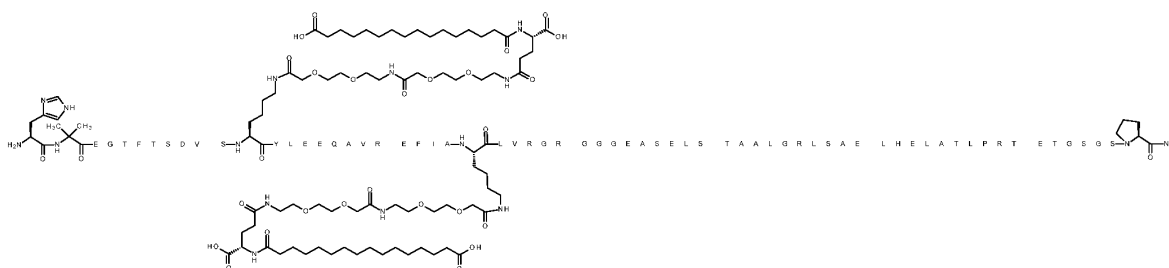
The amino acid sequence of

HAEGTFTSDVSKYLEEQAVREFIAKLVRGRGGGEASELSTAALGRLSAELHELATLPRTET

GSGSP has SEQ ID NO: 214.

Compound 0483

H-Aib-EGTFTSDVS-K([2-[2-[2-[[2-[2-[2-[[[4S]-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAVREFIA-K([2-[2-[2-[[2-[2-[2-[[[4S]-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-LVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₆ H₆₀₁ N₉₅ O₁₂₅

Molecular weight (average) calculated: 8332.2496 g/mol

10 mono isotopic mass: 8327.3592 g/mol

LCMS34: found (M + 5H)⁵⁺ 1667.47 (most abundant)

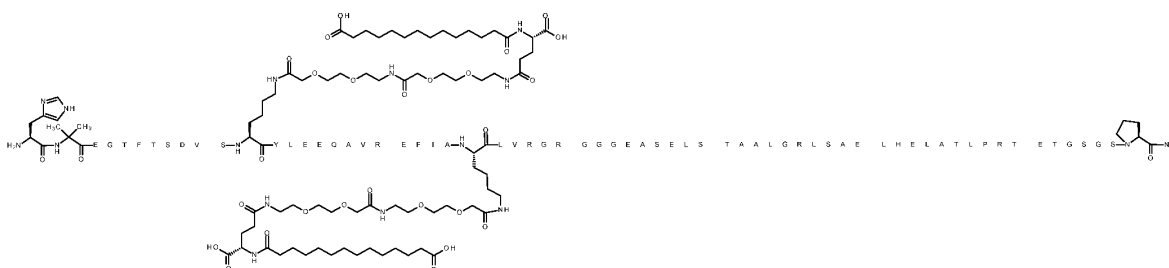
The amino acid sequence of

HXEGTFTSDVSKYLEEQAVREFIAKLVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 215.

15

Compound 0484

H-Aib-EGTFTSDVS-K([2-[2-[2-[[2-[2-[2-[[[4S]-4-carboxy-4-(13-carboxytridecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAVREFIA-K([2-[2-[2-[[2-[2-[2-[[[4S]-4-carboxy-4-(13-carboxytridecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-LVRGRGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₂ H₅₉₃ N₉₅ O₁₂₅

Molecular weight (average) calculated: 8276.1433 g/mol

mono isotopic mass: 8067.1393 g/mol

LCMS34: found (M + 5H)⁵⁺ 1614.52 (mono isotopic)

The amino acid sequence of

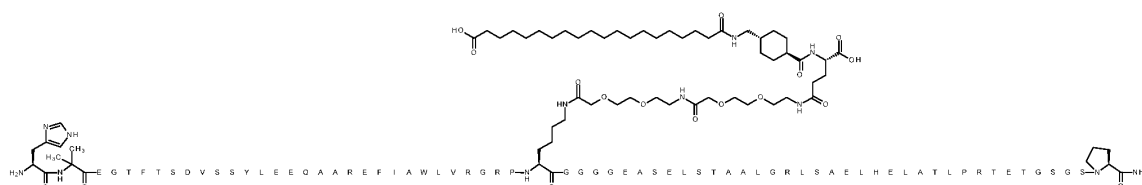
HXEGTFTSDVSSYLEEQAAREFIEWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR

5 TETGSGSP has SEQ ID NO: 216.

Compound 0504

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGRP-K([2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-

10 [[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)]-GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₈ H₅₇₄ N₉₆ O₁₁₈

15 Molecular weight (average) calculated: 8110.9606 g/mol

mono isotopic mass: 8106.1866 g/mol

LCMS34: found (M + 5H)⁵⁺ 1622.34 (most abundant)

The amino acid sequence of

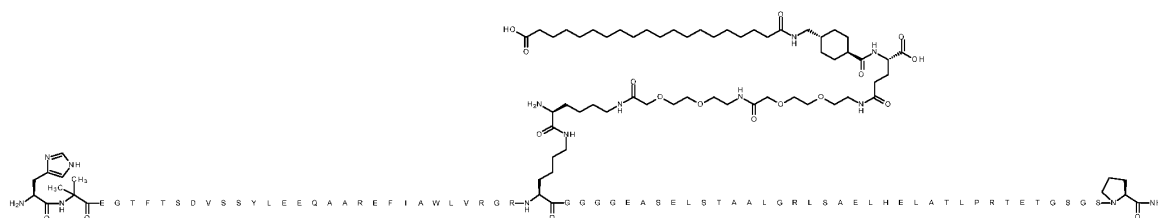
HXEGTFTSDVSSYLEEQAAREFIAWLVRGRP-KGGGGEASELSTAALGRLSAELHELATLP

20 RTETGSGSP has SEQ ID NO: 217.

Compound 0506

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGRP-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-[[4-[(19-

25 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₉ H₅₇₉ N₉₇ O₁₁₈

Molecular weight (average) calculated: 8142.0177 g/mol

mono isotopic mass: 8137.2288 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1629.33 (most abundant)

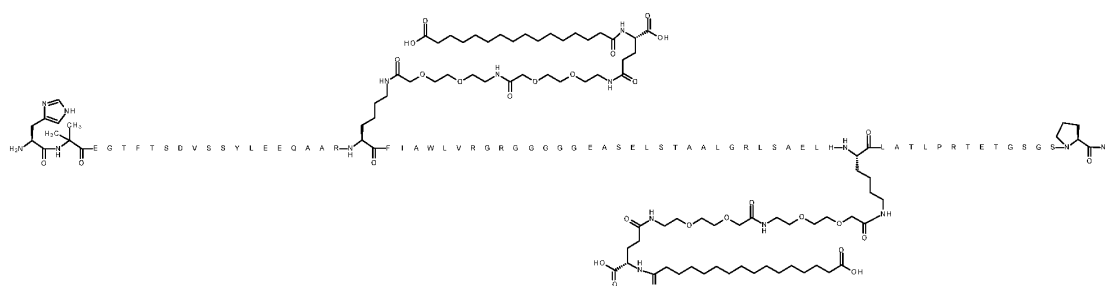
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

- 10 Compound 0509

H-Aib-EGTFTSDVSSYLEEQAAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGRGGGGGEASELSTAALGRLSAELH-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-

- 15 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-LATLPR



C₃₇₂ H₆₀₄ N₉₈ O₁₂₄

Molecular weight (average) calculated: 8433.3584 g/mol

- 20 mono isotopic mass: 8428.3970 g/mol

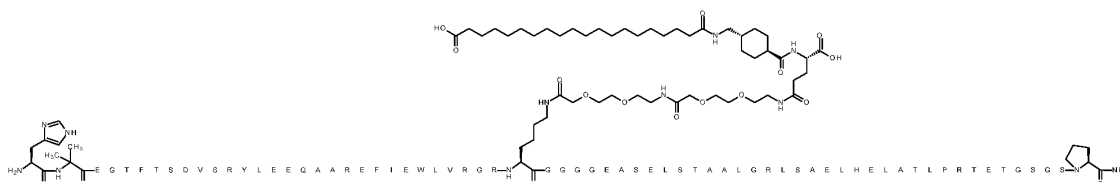
LCMS34: found (M + 5H)⁵⁺ 1687.55 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAARKFIAWLVRGRGGGGGEASELSTAALGRLSAELHKLATLPR
TETGSGSP has SEQ ID NO: 218.

Compound 0511

H-Aib-EGTFTSDVSRYLEEQAAREFIEWLVRGR-K([2-[2-[2-[2-[2-[[4S)-4-carboxy-4-[[4-
 [(19-
 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth
 5 oxy]acetyl]amino]ethoxy]ethoxy]acetyl])-
 GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₈ H₅₇₆ N₉₈ O₁₁₈

Molecular weight (average) calculated: 8140.9898 g/mol

10 mono isotopic mass: 8136.2084 g/mol

LCMS34: found (M + 5H)⁵⁺ 1629.34 (most abundant)

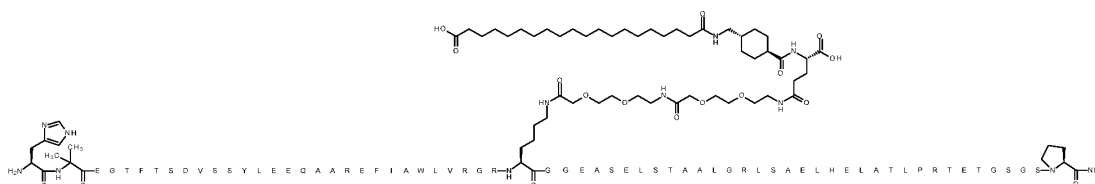
The amino acid sequence of

HXEGTFTSDVSRYLEEQAAREFIEWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
 TETGSGSP has SEQ ID NO: 219.

15

Compound 0512

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[[4S)-4-carboxy-4-[[4-
 [(19-
 carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]eth
 20 oxy]acetyl]amino]ethoxy]ethoxy]acetyl])-
 GGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₄₉ H₅₆₁ N₉₃ O₁₁₅

Molecular weight (average) calculated: 7899.7427 g/mol

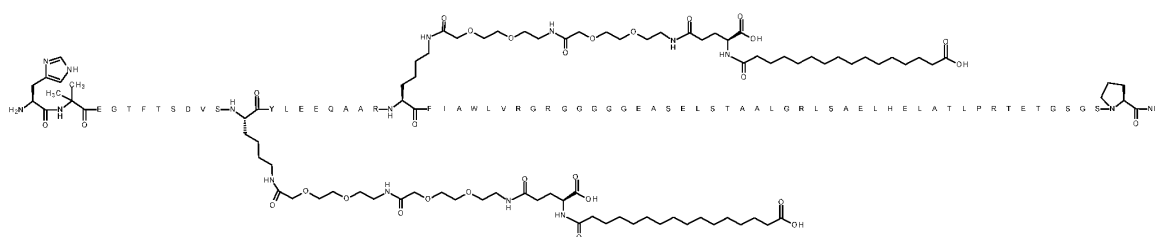
25 mono isotopic mass: 7895.0909 g/mol

LCMS34: found (M + 5H)⁵⁺ 1580.31 (most abundant)

The amino acid sequence of
HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 154.

5 Compound 0516

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₄ H₆₀₆ N₉₈ O₁₂₅

Molecular weight (average) calculated: 8475.3950 g/mol

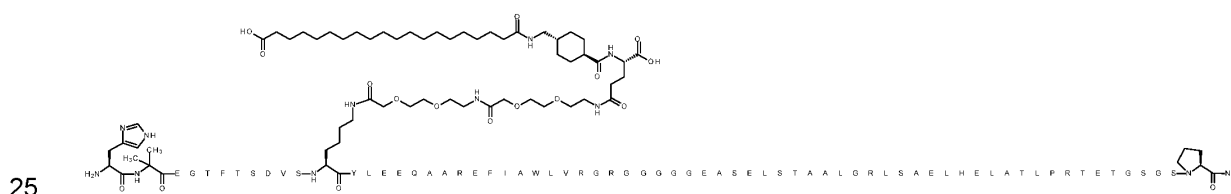
mono isotopic mass: 8470.4075 g/mol

15 LCMS34: found (M + 5H)⁵⁺ 1694.99 (mono isotopic)

The amino acid sequence of
HXEGTFTSDVSKYLEEQAARKFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 220.

20 Compound 0518

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[(4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₂ H₅₆₅ N₉₅ O₁₁₆

Molecular weight (average) calculated: 7983.8194 g/mol

mono isotopic mass: 7979.1232 g/mol

LCMS34: found (M + 5H)⁵⁺ 1597.7 (most abundant)

The amino acid sequence of

HXEGTFTSDVSKYLEEQAAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR

5 TETGSGSP has SEQ ID NO: 221.

Compound 0528

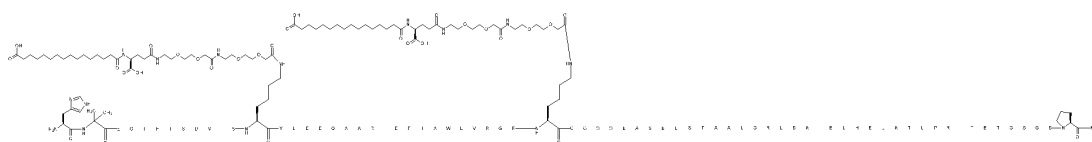
H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-

carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace

10 tyl)-YLEEQAAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-

carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace

tyl)-GGGGEASELSTAALGRLSAELHELATLPR-TETGSGSP-amide



C₃₇₇ H₆₁₀ N₉₈ O₁₂₇

15 Molecular weight (average) calculated: 8547.4577 g/mol

mono isotopic mass: 8542.4287 g/mol

LCMS34: found (M + 5H)⁵⁺ 1710.41 (most abundant)

The amino acid sequence of

HXEGTFTSDVSKYLEEQAAAREFIAWLVRGRKGGGGGEASELSTAALGRLSAELHELATLPR

20 TETGSGSP has SEQ ID NO: 222.

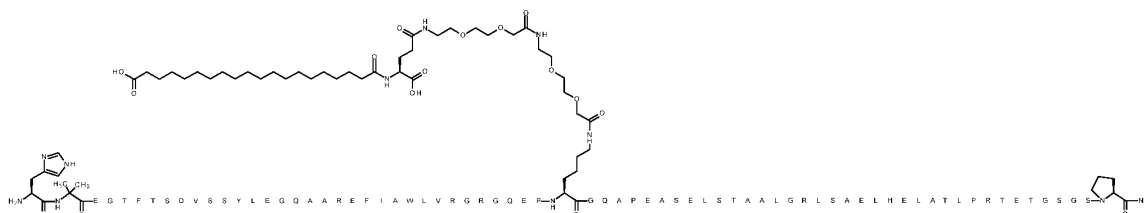
Compound 0529

H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRGRGQEP-K([2-[2-[2-[2-[2-[2-[(4S)-4-

carboxy-4-(19-

25 carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acet

yl)-GQAPEASELSTAALGRLSAELHELATLPR-TETGSGSP-amide



C₃₆₆ H₅₈₆ N₁₀₀ O₁₂₂

Molecular weight (average) calculated: 8339.1658 g/mol

mono isotopic mass: 8334.2724 g/mol

LCMS34: found (M + 5H)⁵⁺ 1668.7 (most abundant)

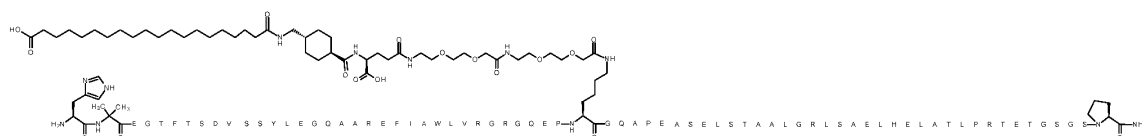
The amino acid sequence of

- 5 HXEGTFTSDVSSYLEGQAAREFIAWLVRGRGQEPKGQAPEASELSTAALGRLSAELHELA
TLPRTETGSGSP has SEQ ID NO: 137.

Compound 0539

H-Aib-EGTFTSDVSSYLEGQAAREFIAWLVRGRGQEP-K([2-[2-[2-[2-[2-[2-[(4S)-4-

- 10 carboxy-4-[[4-[(19-
carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]-
GQAPEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



- 15 C₃₇₄ H₅₉₉ N₁₀₁ O₁₂₃

Molecular weight (average) calculated: 8478.3608 g/mol

mono isotopic mass: 8473.3721 g/mol

LCMS34: found (M + 5H)⁵⁺ 1696.5011 (most abundant)

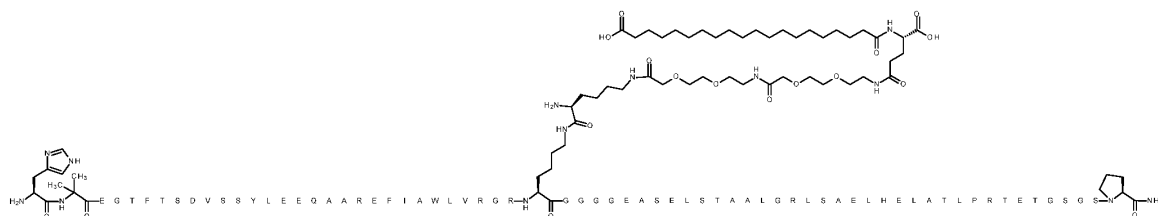
The amino acid sequence of

- 20 HXEGTFTSDVSSYLEGQAAREFIAWLVRGRGQEPKGQAPEASELSTAALGRLSAELHELA
TLPRTETGSGSP has SEQ ID NO: 137.

Compound 0552

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[2-[(4S)-

- 25 4-carboxy-4-(19-
carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acet
yl]amino]hexanoyl])-GGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₁ H₅₆₆ N₉₆ O₁₁₇

Molecular weight (average) calculated: 8002.8227 g/mol

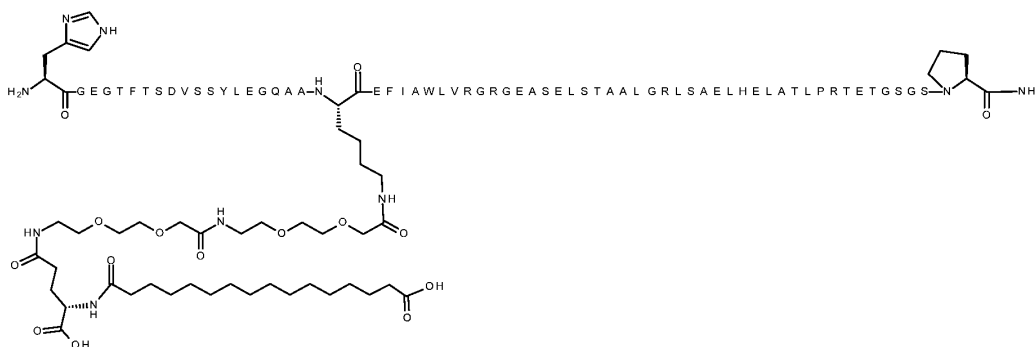
mono isotopic mass: 7998.1291 g/mol

LCMS34: found (M + 5H)⁵⁺ 1600.55 (mono isotopic)

- 5 The amino acid sequence of
HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

Compound 0560

- 10 HEGTFTSDVSSYLEGQAA-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVRGRGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide

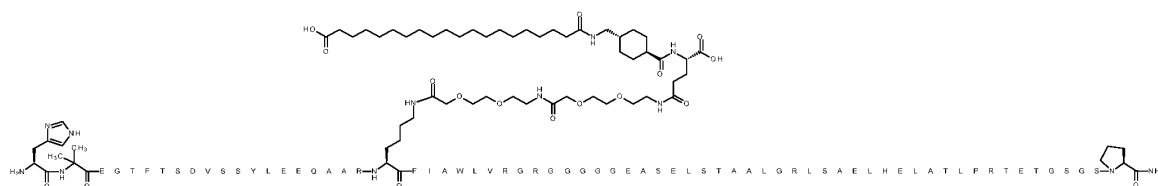


C₃₂₄ H₅₁₇ N₈₇ O₁₁₀

- 15 Molecular weight (average) calculated: 7391.0887 g/mol
mono isotopic mass: 7386.7536 g/mol
LCMS34: found (M + 5H)⁵⁺ 1478.31 (mono isotopic)
The amino acid sequence of
HGETFTSDVSSYLEGQAAKEFIAWLVRGRGEASELSTAALGRLSAELHELATLPR TETGS
20 GSP has SEQ ID NO: 223.

Compound 0561

- 25 H-Aib-EGTFTSDVSSYLEEQAAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-
FIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide



C₃₅₀ H₅₆₃ N₉₅ O₁₁₅

Molecular weight (average) calculated: 7941.7827 g/mol

mono isotopic mass: 7937.1127 g/mol

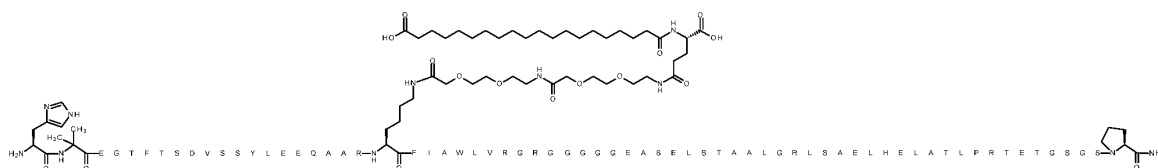
- 5 LCMS01: found (M + 5H)⁵⁺ 1589.3 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 224.

10 Compound 0562

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])]-FIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR)TETGSGSP-amide



- 15 C₃₄₂ H₅₅₀ N₉₄ O₁₁₄

Molecular weight (average) calculated: 7802.5878 g/mol

mono isotopic mass: 7798.0130 g/mol

LCMS01: found (M + 5H)⁵⁺ 1561.5 (most abundant)

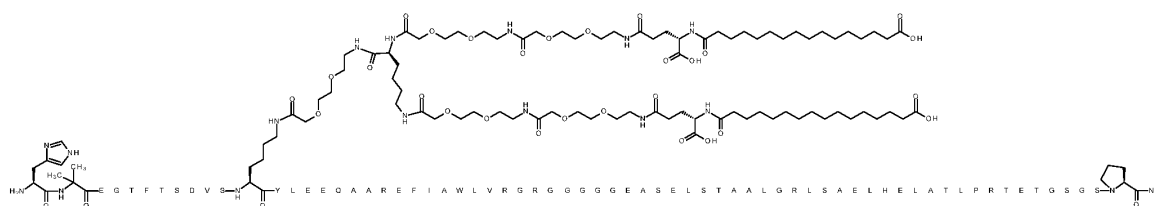
The amino acid sequence of

- 20 HXEGTFTSDVSSYLEEQAARKFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 224.

Compound 0564

H-Aib-EGTFTSDVS-K([2-[2-[2-[(2S)-2,6-bis[[2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(15-

- 25 carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl]amino]ethoxy]ethoxy]acetyl])-
YLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR)TETGSGSP-amide



C₃₈₅ H₆₂₄ N₁₀₀ O₁₃₁

Molecular weight (average) calculated: 8749.6655 g/mol

mono isotopic mass: 8744.5240 g/mol

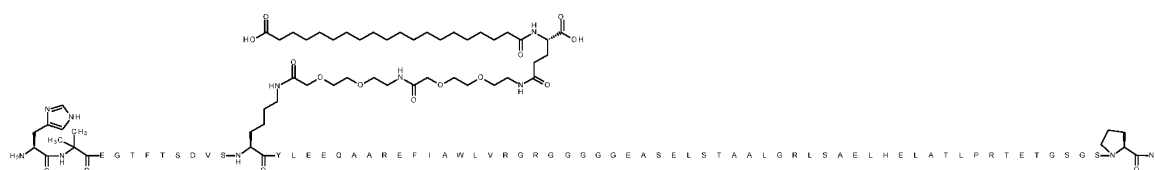
- 5 LCMS34: found (M + 5H)⁵⁺ 1749.8 (mono isotopic)

The amino acid sequence of

HXEGTFTSDVSKYLEEQAAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 221.

10 Compound 0565

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)-YLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



15

C₃₄₄ H₅₅₂ N₉₄ O₁₁₅

Molecular weight (average) calculated: 7844.6245 g/mol

mono isotopic mass: 7840.0235 g/mol

LCMS34: found (M + 5H)⁵⁺ 1568.89 (mono isotopic)

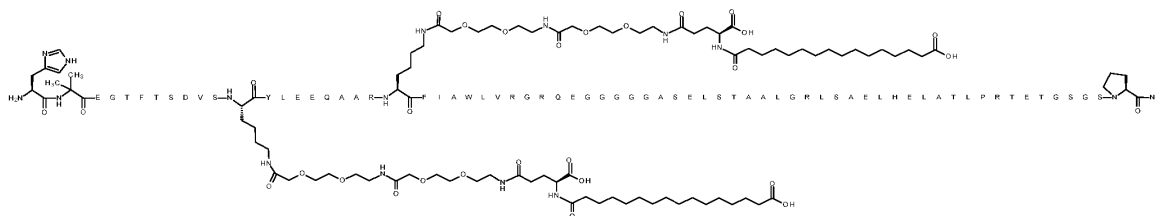
- 20 The amino acid sequence of

HXEGTFTSDVSKYLEEQAAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP has SEQ ID NO: 221.

Compound 0575

- 25 H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl)-YLEEQAAR-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-

carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-FIAWLVRGRQEGGGGGASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₉ H₆₁₄ N₁₀₀ O₁₂₇

5 Molecular weight (average) calculated: 8603.5243 g/mol

mono isotopic mass: 8598.4661 g/mol

LCMS34: found (M + 5H)⁵⁺ 1721.5 (most abundant)

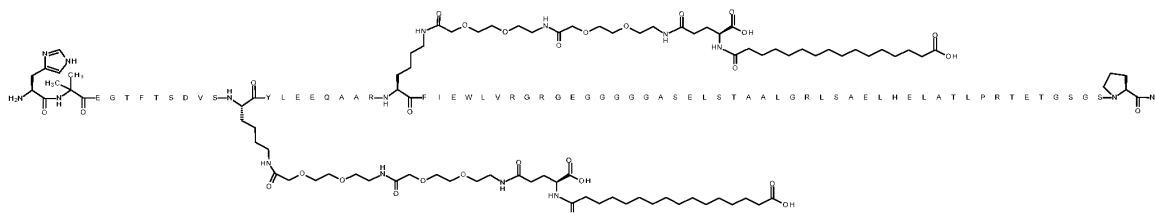
The amino acid sequence of

HXEGTFTSDVSKYLEEQAARKFIAWLVRGRQEGGGGGASELSTAALGRLSAELHELATLP

10 RTETGSGSP has SEQ ID NO: 225.

Compound 0576

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-YLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl))-FIEWLVRGRGEGGGGGASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₈ H₆₁₁ N₉₉ O₁₂₈

20 Molecular weight (average) calculated: 8590.4824 g/mol

mono isotopic mass: 8585.4345 g/mol

LCMS34: found (M + 5H)⁵⁺ 1719.1 (most abundant)

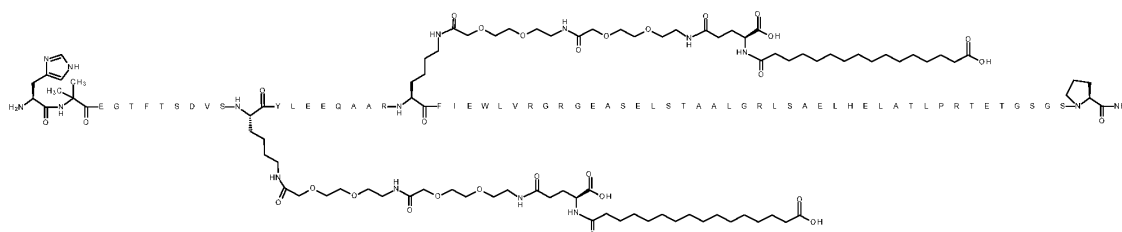
The amino acid sequence of

HXEGTFTSDVSKYLEEQAARKFIEWLVRGRGEGGGGGASELSTAALGRLSAELHELATLP

25 RTETGSGSP has SEQ ID NO: 226.

Compound 0577

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₈ H₅₉₆ N₉₄ O₁₂₃

Molecular weight (average) calculated: 8305.2258 g/mol

mono isotopic mass: 8300.3272 g/mol

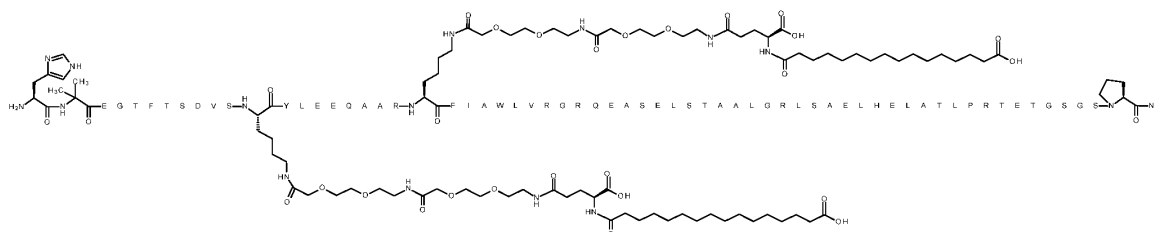
10 LCMS34: found (M + 5H)⁵⁺ 1661.9 (most abundant)

The amino acid sequence of

HXEGTFTSDVSKYLEEQAARKFIEWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP has SEQ ID NO: 227.

15 Compound 0578

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGRQEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₆₉ H₅₉₉ N₉₅ O₁₂₂

Molecular weight (average) calculated: 8318.2677 g/mol

mono isotopic mass: 8313.3588 g/mol

25 LCMS34: found (M + 5H)⁵⁺ 1664.5 (most abundant)

The amino acid sequence of
HXEGTFTSDVSKYLEEQAARKFIAWLVRGRQEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 228.

5 Compound 0580

H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGGASELSTAALGRLSAELHELATLPRTETGSGSP-amide



10 C₃₂₃ H₅₁₉ N₈₅ O₁₀₆

Molecular weight (average) calculated: 7289.0829 g/mol

mono isotopic mass: 7284.7834 g/mol

LCMS01: found (M + 5H)⁵⁺ 1458.76 (most abundant)

The amino acid sequence of H-Aib-

15 EGTFTSDVSSYLEEQAARKFIAWLVRGGASELSTAALGRLSAELHELATLPRTETGSGSP
has SEQ ID NO: 229.

Compound 0581

20 H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRGASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₂₃ H₅₁₈ N₈₄ O₁₀₇

Molecular weight (average) calculated: 7290.0676 g/mol

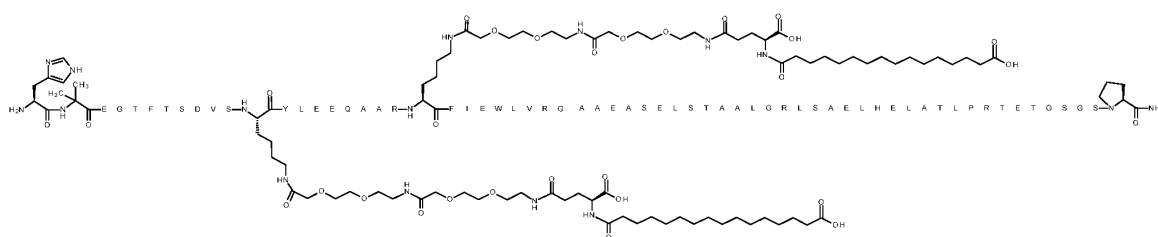
25 mono isotopic mass: 7285.7674 g/mol

LCMS01: found (M + 5H)⁵⁺ 1458.96 (most abundant)

The amino acid sequence of
HXEGTFTSDVSSYLEEQAARKFIEWLVRGASELSTAALGRLSAELHELATLPRTETGSGSP
has SEQ ID NO: 230.

5 Compound 0629

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-YLEEQAAR-K([2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIEWLVRGAAEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₆ H₅₉₁ N₉₁ O₁₂₃

Molecular weight (average) calculated: 8234.1446 g/mol

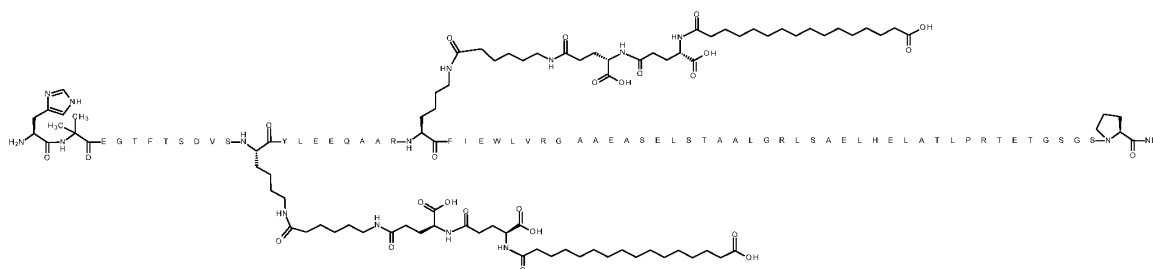
mono isotopic mass: 8229.2788 g/mol

15 LCMS01: found (M + 5H)⁵⁺ 1647.55 (most abundant)

The amino acid sequence of
HXEGTFTSDVSKYLEEQAARKFIEWLVRGAAEASELSTAALGRLSAELHELATLPRTETGS
GSP has SEQ ID NO: 231.

20 Compound 0630

H-Aib-EGTFTSDVS-K(6-[[[(4S)-4-carboxy-4-[[[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]butanoyl]amino]hexanoyl])-YLEEQAAR-K(6-[[[(4S)-4-carboxy-4-[[[(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino]butanoyl]amino]hexanoyl])-FIEWLVRGAAEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₆₄ H₅₈₃ N₉₁ O₁₁₉

Molecular weight (average) calculated: 8138.0621 g/mol

mono isotopic mass: 8133.2365 g/mol

- 5 LCMS01: found (M + 5H)⁵⁺ 1628.55 (most abundant)

The amino acid sequence of

HXEGTFTSDVSKYLEEQAARKFIEWLVRGAAEASELSTAALGRLSAELHELATLPRTETGSG

GSP has SEQ ID NO: 231.

- 10 Compound 0631

H-Aib-EGTFTSDVS-K([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-

carboxy-4-(15-

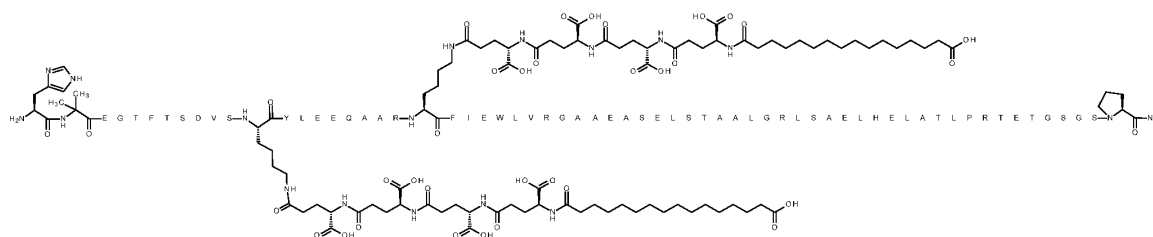
carboxypentadecanoylamino)butanoyl]amino)butanoyl]amino)butanoyl]amino)butanoyl])-

YLEEQAAR-K([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-

- 15 (15-

carboxypentadecanoylamino)butanoyl]amino)butanoyl]amino)butanoyl]amino)butanoyl])-

FIEWLVRGAAEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₇₂ H₅₈₉ N₉₃ O₁₂₉

- 20 Molecular weight (average) calculated: 8428.2028 g/mol

mono isotopic mass: 8423.2388 g/mol

LCMS34: found (M + 5H)⁵⁺ 1686.7 (most abundant)

The amino acid sequence of

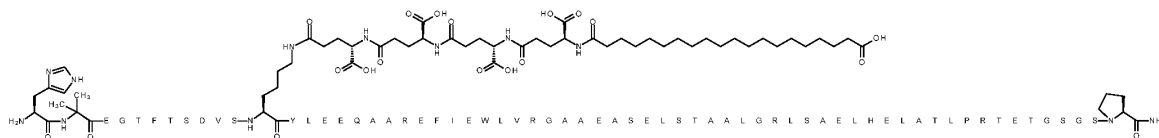
HXEGTFTSDVSKYLEEQAARKFIEWLVRGAAEASELSTAALGRLSAELHELATLPRTETGSG

- 25 GSP has SEQ ID NO: 231.

Compound 0632

H-Aib-EGTFTSDVS-K([[(4S)-4-carboxy-4-[[[(4S)-4-carboxy-4-[[[(4S)-4-carboxy-4-[[[(4S)-4-

- 5 carboxynonadecanoylamino)butanoyl]amino]butanoyl]amino]butanoyl]amino]butanoyl))-
YLEEQAAAREFIEWLVRGAAEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₃₉ H₅₃₆ N₈₈ O₁₁₆

Molecular weight (average) calculated: 7700.4031 g/mol

- 10 mono isotopic mass: 7695.8748 g/mol

LCMS34: found (M + 5H)⁵⁺ 1541.1 (most abundant)

The amino acid sequence of

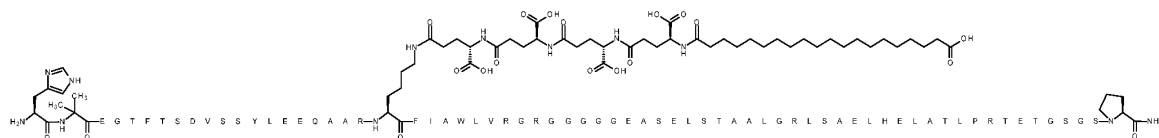
HXEGTFTSDVSKYLEEQAAAREFIEWLVRGAAEASELSTAALGRLSAELHELATLPRRTETGS
GSP has SEQ ID NO: 232.

15

Compound 0633

H-Aib-EGTFTSDVSSYLEEQAAAR-K([[(4S)-4-carboxy-4-[[[(4S)-4-carboxy-4-[[[(4S)-4-carboxy-

- 4-[[[(4S)-4-carboxy-4-(19-
carboxynonadecanoylamino)butanoyl]amino]butanoyl]amino]butanoyl]amino]butanoyl))-
20 FIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₄₅ H₅₄₉ N₉₅ O₁₁₇

Molecular weight (average) calculated: 7899.6169 g/mol

mono isotopic mass: 7894.9930 g/mol

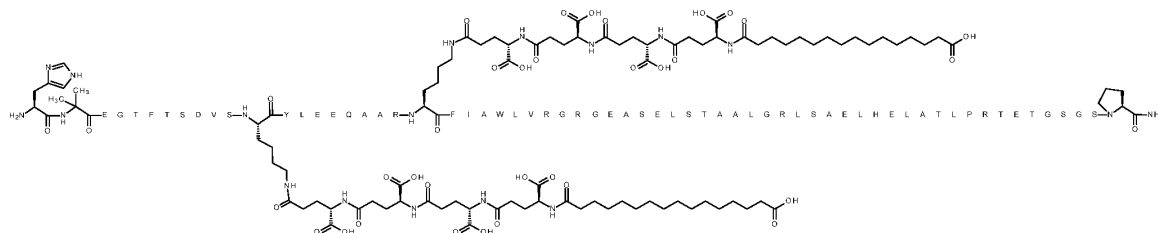
- 25 LCMS34: found (M + 5H)⁵⁺ 1580.9 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAAARKFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 224.

- 30 Compound 0634

H-Aib-EGTFTSDVS-K([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino)butanoyl]amino)butanoyl]amino)butanoyl]amino)butanoyl])-YLEEQAAR-K([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino)butanoyl]amino)butanoyl]amino)butanoyl]amino)butanoyl])-FIAWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₇₂ H₅₉₂ N₉₆ O₁₂₇

10 Molecular weight (average) calculated: 8441.2479 g/mol

mono isotopic mass: 8436.2817 g/mol

LCMS34: found (M + 5H)⁵⁺ 1689.1 (most abundant)

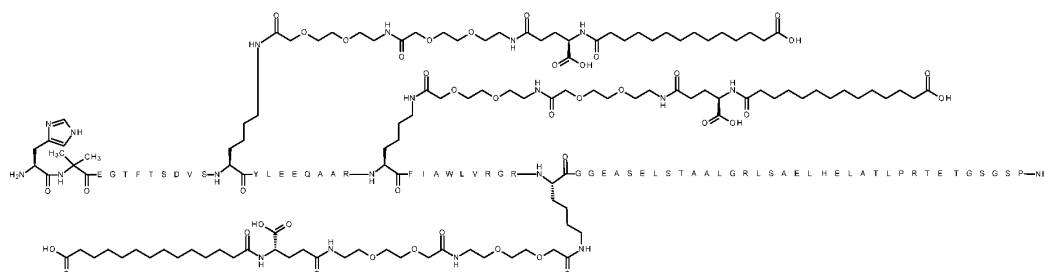
The amino acid sequence of

HXEGTFTSDVSKYLEEQAARKFIAWLVRGRGEASELSTAALGRLSAELHELATLPRRTETGS

15 GSP has SEQ ID NO: 233.

Compound 0635

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[2-([(4S)-4-carboxy-4-(13-carboxytridecanoylamino)butanoyl]amino)ethoxy]ethoxy]acetyl]amino)ethoxy]ethoxy]acetyl])-YLEEQAAR-K([2-[2-[2-[2-[2-[2-([(4R)-4-carboxy-4-(13-carboxytridecanoylamino)butanoyl]amino)ethoxy]ethoxy]acetyl]amino)ethoxy]ethoxy]acetyl])-FIAWLVRGR-K([2-[2-[2-[2-[2-[2-([(4S)-4-carboxy-4-(13-carboxytridecanoylamino)butanoyl]amino)ethoxy]ethoxy]acetyl]amino)ethoxy]ethoxy]acetyl])-GGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



25

C₄₀₁ H₆₅₄ N₁₀₀ O₁₃₅

Molecular weight (average) calculated: 9036.0725 g/mol

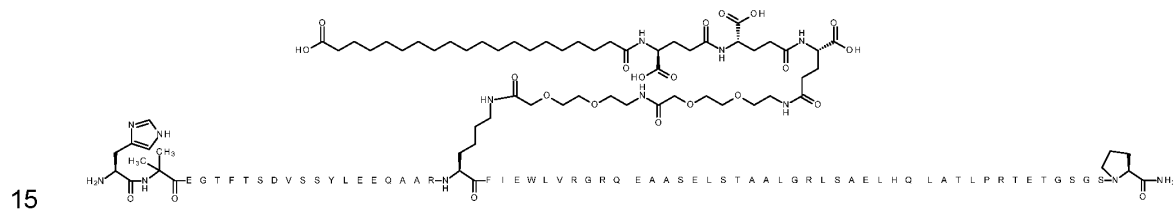
mono isotopic mass: 9030.7384 g/mol

LCMS34: found (M + 5H)⁵⁺ 1807.05 (mono isotopic)

- 5 The amino acid sequence of
HXEGTFTSDVSKYLEEQAARKFIAWLVRGRKGGEASELSTAALGRLSAELHELATLPRTET
GSGSP has SEQ ID NO: 234.

Compound 0636

- 10 H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-[(4S)-4-carboxy-4-
[[4S)-4-carboxy-4-(19-
carboxynonadecanoylamino)butanoyl]amino]butanoyl]amino]butanoyl]amino]ethoxy]ethoxy]
acetyl]amino]ethoxy]ethoxy]acetyl))-
FIEWLVRGRQEAASELSTAALGRLSAELHQLATLPRTETGSGSP-amide



C₃₅₂ H₅₆₅ N₉₅ O₁₁₉

Molecular weight (average) calculated: 8031.8176 g/mol

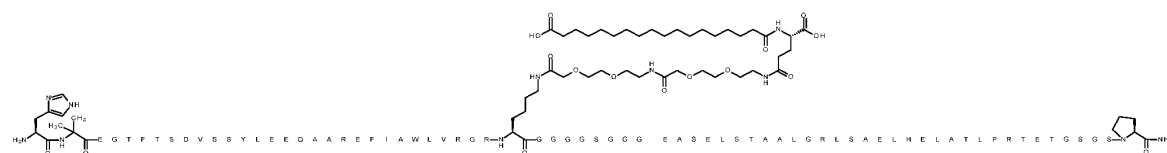
mono isotopic mass: 8027.1080 g/mol

LCMS34: found (M + 5H)⁵⁺ 1606.3 (mono isotopic)

- 20 The amino acid sequence of
HXEGTFTSDVSSYLEEQAARKFIEWLVRGRQEAASELSTAALGRLSAELHQLATLPRTETG
SGSP has SEQ ID NO: 235.

Compound 0637

- 25 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-
carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
tyl))-GGGGSGGGGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₅₂ H₅₆₄ N₉₈ O₁₂₁

Molecular weight (average) calculated: 8104.8286 g/mol

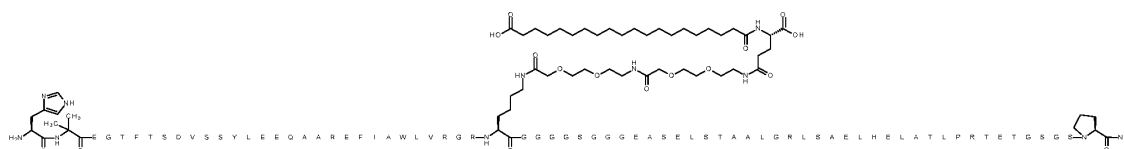
mono isotopic mass: 8100.0992 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1621.8 (most abundant)

- 5 The amino acid sequence of
HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGSGGGGEASELSTAALGRLSAELHELA
TLPRTEGSGSP has SEQ ID NO: 236.

Compound 0638

- 10 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGGGSGGGGEASELSTAALGRLSAELHELATLPRTEGSGSP-amide

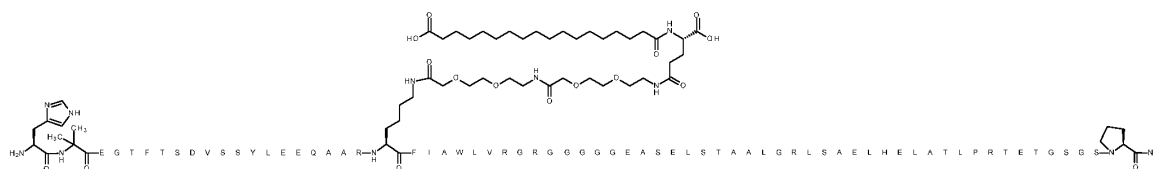


C₃₅₄ H₅₆₈ N₉₈ O₁₂₁

- 15 Molecular weight (average) calculated: 8132.8817 g/mol
mono isotopic mass: 8128.1305 g/mol
LCMS_ZQ: found (M + 5H)⁵⁺ 1627.5 (most abundant)
The amino acid sequence of
HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGSGGGGEASELSTAALGRLSAELHELA
20 TLPRTEGSGSP has SEQ ID NO: 236.

Compound 0639

- H-Aib-EGTFTSDVSSYLEEQAAR-K([2-[2-[2-[2-[2-[2-[[[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-FIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPRTEGSGSP-amide



C₃₄₀ H₅₄₆ N₉₄ O₁₁₄

Molecular weight (average) calculated: 7774.5346 g/mol

mono isotopic mass: 7769.9817 g/mol

LCMS_ZQ: found (M + 5H)⁵⁺ 1556.0 (most abundant)

The amino acid sequence of

HXEGTFTSDVSSYLEEQAARKFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR

TETGSGSP has SEQ ID NO: 224.

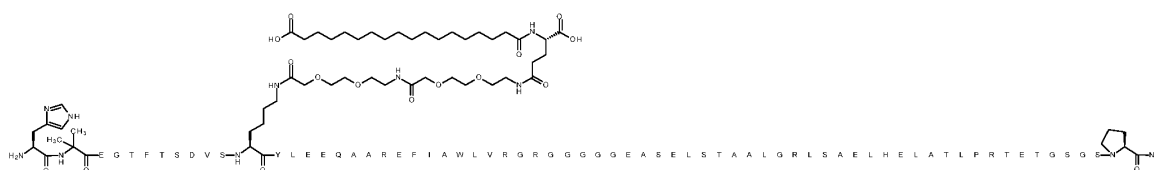
5

Compound 0640

H-Aib-EGTFTSDVS-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]-YLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR

10 amide



C₃₄₂ H₅₄₈ N₉₄ O₁₁₅

Molecular weight (average) calculated: 7816.5713 g/mol

mono isotopic mass: 7811.9922 g/mol

15 LCMS34: found (M + 5H)⁵⁺ 1564.3 (most abundant)

The amino acid sequence of

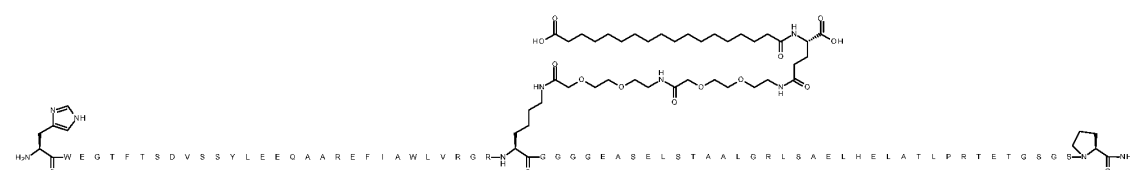
HXEGTFTSDVSKYLEEQAAREFIAWLVRGRGGGGGEASELSTAALGRLSAELHELATLPR

TETGSGSP has SEQ ID NO: 221.

20 Compound 0648

HWEGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-

carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]-GGGGGEASELSTAALGRLSAELHELATLPR



25 C₃₅₀ H₅₅₃ N₉₅ O₁₁₆

Molecular weight (average) calculated: 7947.7027 g/mol

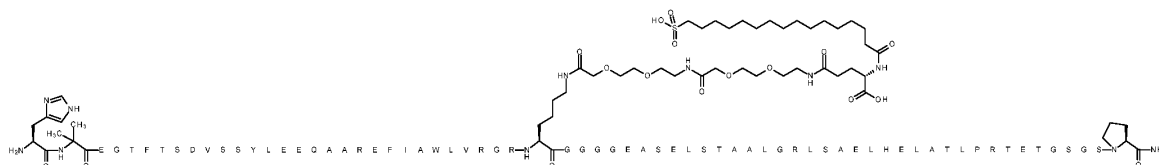
mono isotopic mass: 7943.0293 g/mol

LCMS34: found (M + 5H)⁵⁺ 1590.5 (most abundant)

The amino acid sequence of
 HWEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGGEASELSTAALGRLSAELHELATLPR
 TETGSGSP has SEQ ID NO: 185.

5 Compound 0654

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[[2-[2-[[[(4S)-4-carboxy-4-(16-sulfohexadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])
 -GGGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide



10 C₃₄₁ H₅₄₈ N₉₄ O₁₁₇ S

Molecular weight (average) calculated: 7868.6244 g/mol

mono isotopic mass: 7863.9541 g/mol

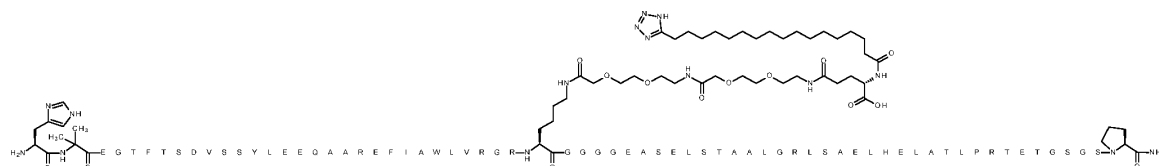
LCMS34: found (M + 5H)⁵⁺ 1573.79 (mono isotopic)

The amino acid sequence of

15 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGGEASELSTAALGRLSAELHELATLPR
 TETGSGSP has SEQ ID NO: 153.

Compound 0655

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[[2-[2-[[[(4S)-4-carboxy-4-[17-(1H-tetrazol-5-yl)heptadecanoylamino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])
 -GGGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide



C₃₄₃ H₅₅₀ N₉₈ O₁₁₄

25 Molecular weight (average) calculated: 7870.6253 g/mol

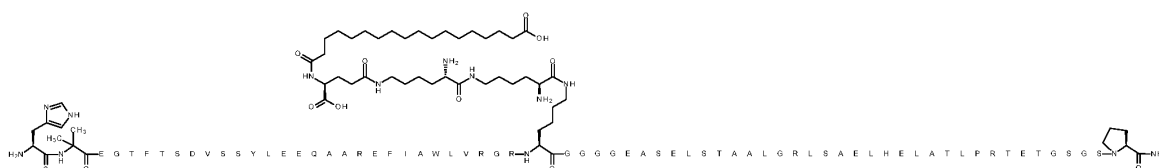
mono isotopic mass: 7866.0253 g/mol

LCMS34: found (M + 5H)⁵⁺ 1574.2 (mono isotopic)

The amino acid sequence of
HXEGTFTSDVSSYLEEQAAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

5 Compound 0656

H-Aib-EGTFTSDVSSYLEEQAAAREFIAWLVRGR-K([(2S)-2-amino-6-([(2S)-2-amino-6-([(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]hexanoyl]amino]hexanoyl)]-GGGGEASELSTAALGRLSAELHELATLPR)TETGSGSP-amide



10 C₃₄₃ H₅₅₂ N₉₆ O₁₁₂

Molecular weight (average) calculated: 7812.6290 g/mol

mono isotopic mass: 7808.0449 g/mol

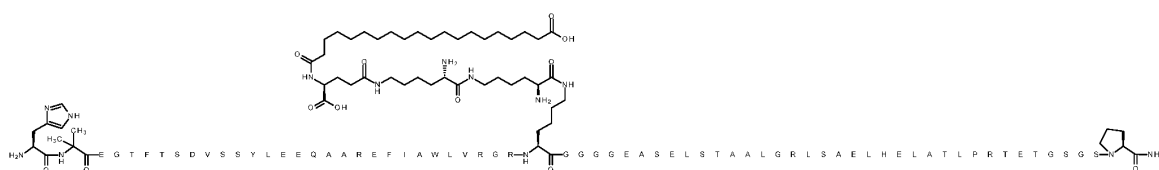
LCMS34: found (M + 5H)⁵⁺ 1562.61 (mono isotopic)

The amino acid sequence of

15 HXEGTFTSDVSSYLEEQAAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

Compound 0657

H-Aib-EGTFTSDVSSYLEEQAAAREFIAWLVRGR-K([(2S)-2-amino-6-([(2S)-2-amino-6-([(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]hexanoyl]amino]hexanoyl)]-GGGGEASELSTAALGRLSAELHELATLPR)TETGSGSP-amide



C₃₄₅ H₅₅₆ N₉₆ O₁₁₂

Molecular weight (average) calculated: 7840.6821 g/mol

25 mono isotopic mass: 7836.0762 g/mol

LCMS34: found (M + 5H)⁵⁺ 1568.21 (mono isotopic)

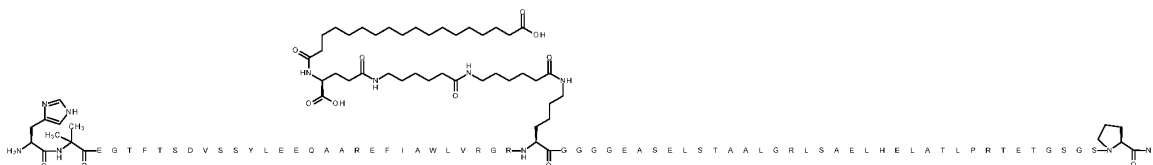
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

Compound 0658

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K(6-[6-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]hexanoylamino]hexanoyl)-

5 GGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide



C₃₄₃ H₅₅₀ N₉₄ O₁₁₂

Molecular weight (average) calculated: 7782.5997 g/mol

mono isotopic mass: 7778.0231 g/mol

10 LCMS34: found (M + 5H)⁵⁺ 1556.6 (mono isotopic)

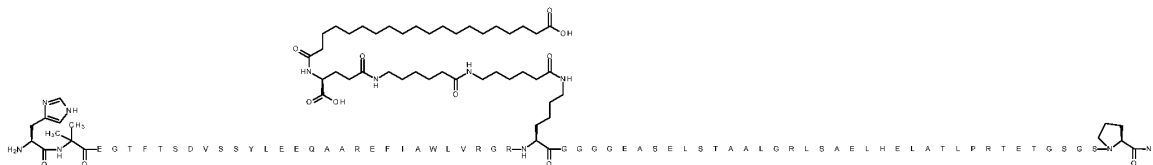
The amino acid sequence of

HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

15 Compound 0659

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K(6-[6-[(4S)-4-carboxy-4-(19-carboxynonadecanoylamino)butanoyl]amino]hexanoylamino]hexanoyl)-

GGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide

20 C₃₄₅ H₅₅₄ N₉₄ O₁₁₂

Molecular weight (average) calculated: 7810.6529 g/mol

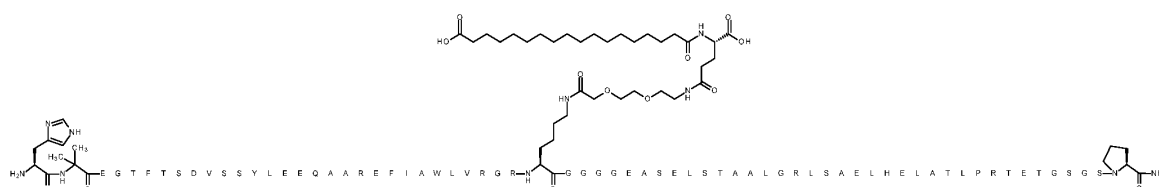
mono isotopic mass: 7806.0544 g/mol

LCMS34: found (M + 5H)⁵⁺ 1562.21 (mono isotopic)

The amino acid sequence of

25 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.Compound 0660

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl])-GGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide



5 C₃₃₇ H₅₃₉ N₉₃ O₁₁₃

Molecular weight (average) calculated: 7701.4409 g/mol

mono isotopic mass: 7696.9289 g/mol

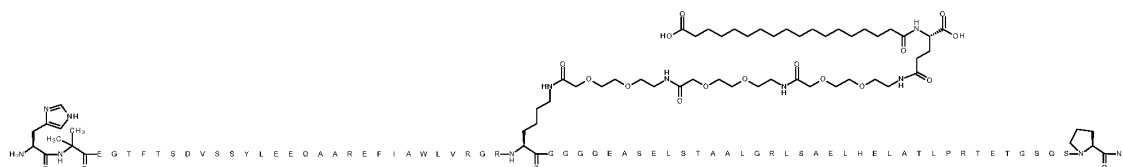
LCMS34: found (M + 5H)⁵⁺ 1540.38 (mono isotopic)

The amino acid sequence of

10 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

Compound 0661

15 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-GGGGEASELSTAALGRLSAELHELATLPR TETGSGSP-amide



20 C₃₄₉ H₅₆₁ N₉₅ O₁₁₉

Molecular weight (average) calculated: 7991.7537 g/mol

mono isotopic mass: 7987.0767 g/mol

LCMS34: found (M + 5H)⁵⁺ 1598.41 (mono isotopic)

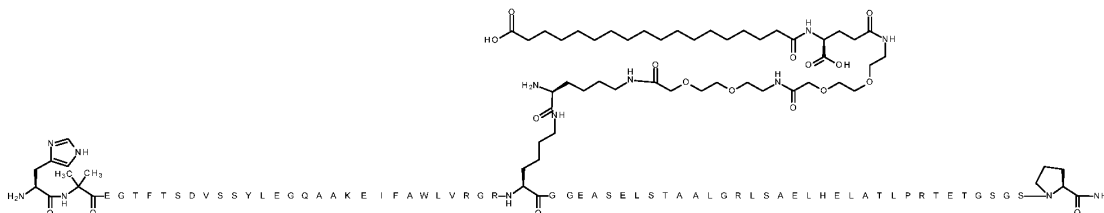
The amino acid sequence of

25 HXEGTFTSDVSSYLEEQAAREFIAWLVRGRKGGGGEASELSTAALGRLSAELHELATLPR
TETGSGSP has SEQ ID NO: 153.

Compound 0662

Example 1b: Comparator compoundsComparator compound 0164

H-Aib-EGTFTSDVSSYLEGQAAKEIFAWLVRGR-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-GGEASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₃₄₂ H₅₅₂ N₉₂ O₁₁₃

10 Molecular weight (average) calculated: 7760.5909 g/mol

mono isotopic mass: 7756.0276 g/mol

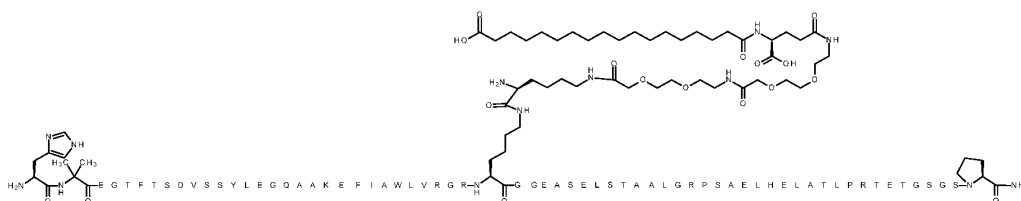
LCMS34: found (M + 5H)⁵⁺ 1553.02 (most abundant).

The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 241.

15

Comparator compound 0185

H-Aib-EGTFTSDVSSYLEGQAAKEIFAWLVRGR-K([(2S)-2-amino-6-[[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl]amino]hexanoyl)]-GGEASELSTAALGRPSAELHELATLPRTETGSGSP-amide



C₃₄₁ H₅₄₈ N₉₂ O₁₁₃

Molecular weight (average) calculated: 7744.5484 g/mol

mono isotopic mass: 7739.9963 g/mol

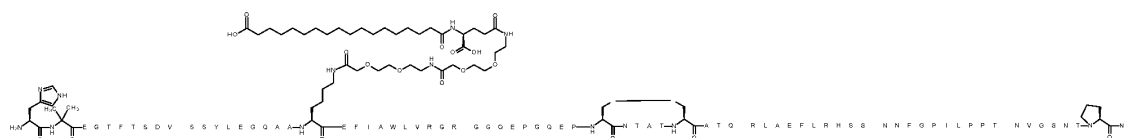
25 LCMS34: found (M + 5H)⁵⁺ 1549.81 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 242.

Comparator compound 0015

H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVGRGGQEPGQEP-NTATCATQRLAEFLRHSSNNFGPILPPTNVGSNTP-amide

5



C₃₈₄ H₅₉₆ N₁₀₆ O₁₂₄ S₂

Molecular weight (average) calculated: 8745.6068 g/mol

mono isotopic mass: 8740.3031 g/mol

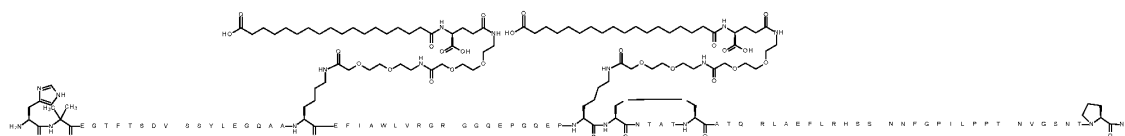
10 LCMS_ZQ: found (M + 5H)⁵⁺ 1749.9 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 243.

Comparator compound 0016

15 H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVGRGGQEPGQEP-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-CNTATCATQRLAEFLRHSSNNFGPILPPTNVGSNTP-amide

20



C₄₂₅ H₆₆₉ N₁₁₁ O₁₃₇ S₂

Molecular weight (average) calculated: 9589.6509 g/mol

mono isotopic mass: 9583.8236 g/mol

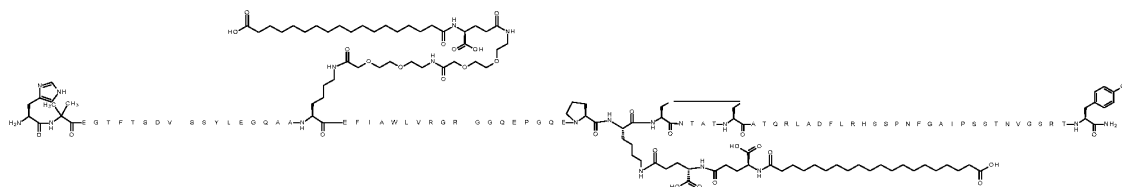
LCMS_ZQ: found (M + 5H)⁵⁺ 1918.9 (most abundant)

25 The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 244.

Comparator compound 0668

30 H-Aib-EGTFTSDVSSYLEGQAA-K([2-[2-[2-[[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-EFIAWLVGRGGQEPGQEP-NTATCATQRLAEFLRHSSNNFGPILPPTNVGSNTP-amide

tyl))-EFIAWLVRGRGGQEPGQEP-K([(4S)-4-carboxy-4-([(4S)-4-carboxy-4-(15-carboxypentadecanoylamino)butanoyl]amino)butanoyl])-CNTATCATQRLADFLRHSSPNFGAIPSSSTNVGSRTY-amide



5 C₄₁₉ H₆₅₅ N₁₁₁ O₁₃₅ S₂

Molecular weight (average) calculated: 9471.4767 g/mol

mono isotopic mass: 9465.7242 g/mol

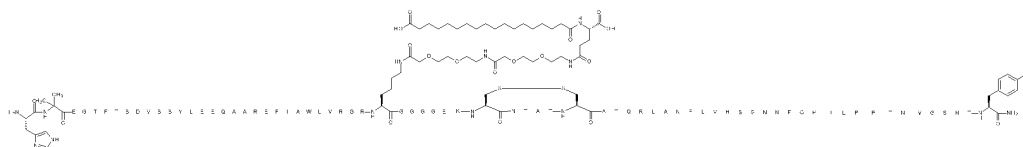
LCMS_ZQ: found (M + 5H)⁵⁺ 1895.2 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ

10 ID NO: 245.

Comparator compound 0671

H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-([(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino)ethoxy]ethoxy]acetyl]amino)ethoxy]ethoxy]acetyl))-GGGGEKCNATCATQRLANFLVHSSNNFGPILPPTNVGSNTY-amide



C₃₇₈ H₅₈₈ N₁₀₄ O₁₂₀ S₂

Molecular weight (average) calculated: 8573.4681 g/mol

mono isotopic mass: 8568.2547 g/mol

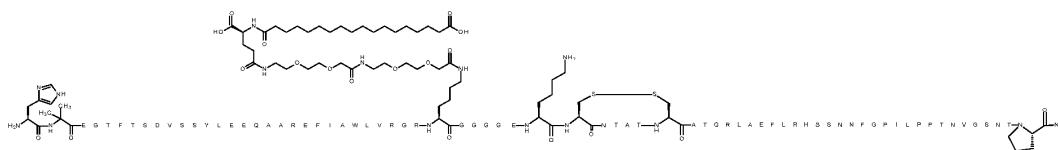
20 LCMS_ZQ: found (M + 5H)⁵⁺ 1715.4 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ

ID NO: 251.

Comparator compound 0672

25 H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-([(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino)ethoxy]ethoxy]acetyl]amino)ethoxy]ethoxy]acetyl))-GGGGEKCNATCATQRLAEFLRHSSNNFGPILPPTNVGSNTP-amide



C₃₇₆ H₅₉₀ N₁₀₆ O₁₂₀ S₂

Molecular weight (average) calculated: 8579.4760 g/mol

5 mono isotopic mass: 8574.2765 g/mol

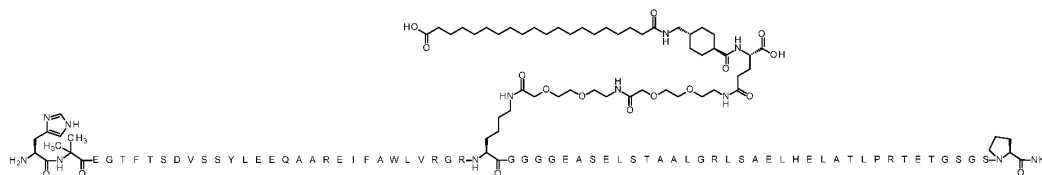
LCMS_ZQ: found (M + 5H)⁵⁺ 1716.8 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 252.

10 Comparator compound 0167

H-Aib-EGTFTSDVSSYLEEQAAREIFAWLVRGR-K([2-[2-[2-[[2-[2-[[[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

15 GGGGEASELSTAALGRLSAELHELATLPRRTETGSGSP-amide



C₃₅₃ H₅₆₇ N₉₅ O₁₁₇

Molecular weight (average) calculated: 8013.8454 g/mol

20 mono isotopic mass: 8009.1338 g/mol

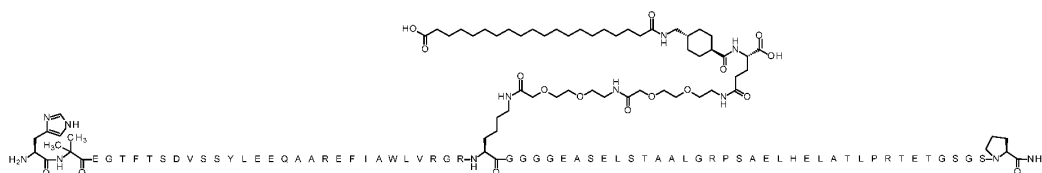
LCMS34: found (M + 5H)⁵⁺ 1603.63 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 253.

25 Comparator compound 0192

H-Aib-EGTFTSDVSSYLEEQAAREIFAWLVRGR-K([2-[2-[2-[[2-[2-[[[(4S)-4-carboxy-4-[[4-[(19-carboxynonadecanoylamino)methyl]cyclohexanecarbonyl]amino]butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]acetyl])-

30 GGGGEASELSTAALGRPSAELHELATLPRRTETGSGSP-amide



C₃₅₂ H₅₆₃ N₉₅ O₁₁₇

Molecular weight (average) calculated: 7997.8029 g/mol

mono isotopic mass: 7993.1025 g/mol

- 5 LCMS34: found (M + 5H)⁵⁺ 1600.63 (most abundant)

The amino acid sequence of the peptide backbone in this comparator compound has SEQ ID NO: 254.

Example 1c: Amylin receptor agonist

10

Compound 1806

EASELSTAALGRLSAELHELATLPRTETGSGSP-amide



C₁₄₁ H₂₃₆ N₄₂ O₅₂ Molecular weight (average) calculated: 3351.6327 g/mol

- 15 mono isotopic mass: 3349.7114 g/mol

LCMS_ZQ: found (M + 3H)³⁺ 1118.4 (most abundant).

The amino acid sequence of the peptide backbone in compound 1806 has SEQ ID NO: 250.

20 **Example 2: Potencies of compounds on the GLP-1 and amylin receptors**

The potencies of compounds were tested as described in Assays 1 and 2.

Assay 1: Human GLP-1 receptor in vitro potency assay

- 25 To determine the ability of compounds to activate or agonise the GLP-1 receptor (GLP-1R), *in vitro* potency assays in cells expressing the human GLP-1 receptor were performed as described below.

Assay principle

- 30 Activation of GLP-1 receptors leads to increased cellular concentrations of cyclic AMP (cAMP). Consequently, transcription is activated by promoters containing multiple

copies of the cAMP response element (CRE). It is thus possible to measure GLP-1 receptor activity using a CRE-luciferase reporter gene introduced into Baby Hamster Kidney (BHK) cells co-expressing the GLP-1 receptor.

5 Cells and assay reagents

Cell stocks were prepared by culturing a stably transfected cell line expressing the human GLP-1 receptor and the CRE responsive luciferase (CRE-Luc) reporter gene (BHK 467-12A KZ-10, prepared according to methods known to the person skilled in the art) in growth medium consisting of DMEM (Gibco, 61965-026) supplemented with 10% FBS (Gibco, 16140-071), 1% Pen/Strep (Gibco, 15140-122), 1 mM Na-pyruvate (Gibco, 11360-039), 1 mg/mL G418 (Gibco, 10131-027) and 240 nM MTX (Pfizer, 15936). Cells at approximately 80-90% confluence were washed once in PBS and loosened from the cell flasks with Versene (Gibco, 15040-033). After centrifugation, the cell pellet was dissolved and diluted to 1.5×10^6 cells/mL in medium consisting of DMEM (Gibco, 61965-026) supplemented with 20% FBS (Gibco, 16140-071), 1% Pen/Strep (Gibco, 15140-122), 1mM Na-Pyruvate (Gibco, 11360-039), 1 mg/mL G418 (Gibco, 10131-027), 240 nM MTX (Pfizer, 15936) and 10% DMSO (Sigma, D2650). Cells were aliquoted and stored at -180°C until use.

The assay buffer consisted of DMEM without phenol red (Gibco, 11880-028) supplemented with 1X GlutaMAX (Gibco, 35050-038), 10 mM HEPES (Gibco, 15630-056), 1% (w/v) ovalbumin (Sigma, A5503) and 0.1% (v/v) Pluronic F-68 (Gibco, 24040-032).

Procedure

To perform the assay, serial dilutions (10-fold dilutions, 8 concentrations per compound) of comparator compounds and GLP-1 receptor-amylin receptor co-agonists were performed in assay buffer without HAS, often starting from approximately 100-200 nM in a 96-well plate. Frozen stocks of human GLP-1R/CRE-Luc cells were thawed in a 37°C water bath, washed once in PBS and diluted to 100.000 cells/mL in assay buffer. For each dilution, 50 μL aliquots of comparator compounds or GLP-1 receptor-amylin receptor co-agonists were transferred to 96-well assay plates (ThermoFisher, 237105) to which 50 μL of the cell suspension was added (5.000 cells/well). The assay plates were incubated for 3 hours at 37°C in 5% CO_2 , left at room temperature for 5 minutes after which 100 μL SteadyLite Plus (PerkinElmer, 6066759) was added to each well. Plates were sealed and incubated at room temperature with gentle shaking for 30 minutes while protected from light. Luminescence was detected on a luminescence plate reader e.g. a Synergy 2

(BioTek). The EC₅₀-values [pM] were calculated by non-linear curve fitting applying a four-parameter logistic model (Hill slope = 1) using GraphPad Prism or by means of TIBCO Enterprise Runtime for R (TIBCO Software, Palo Alto, CA, USA).

5 **Assay 2: Human amylin receptor *in vitro* potency assay**

To determine the ability of compounds to activate or agonise the amylin receptor, *in vitro* potency assays on cells expressing the human amylin receptor (hAmyR3) can be performed as described below.

10 Assay principle

Activation of hAmyR3 leads to increased cellular concentrations of cAMP. Consequently, transcription is activated by promoters containing multiple copies of the cAMP response element (CRE). It is thus possible to measure hAmyR3 activity using a CRE-luciferase reporter gene introduced into Baby Hamster Kidney (BHK) cells co-expressing the hAmyR3.

Cells and assay reagents

A BHK cell line was stably transfected with the human calcitonin receptor (a) and a CRE-responsive luciferase (CRE-Luc) reporter gene according to methods known to the person skilled in the art (Holle-1 cell line, obtained from Zymogenetics described in US patent 5622839). The cell line was further transfected with human receptor modifying protein 3 (RAMP3) using standard methods. This turns the human calcitonin receptor into a human amylin-3(a) receptor.

Cells stocks were prepared by culturing of the stably transfected BHK hAmyR3/CRE-Luc cell line in growth medium consisting of DMEM (Gibco, 31966-021) supplemented with 10% FBS (Gibco, 1640-071), 1% Pen/Strep (Gibco, 15140-122), 0.5 mg/mL Geneticin (Gibco, 10131-027), 0.4 mg/mL Hygromycin (Invitrogen, 1068701) and 250 nM Methotrexate (Sigma, A6770). Cells at approximately 80-90% confluence were washed once with PBS and loosened from the cell flasks with Versene (Gibco, 15040-033). After centrifugation, the cell pellet was dissolved and diluted to 2.5x10⁶ cells/mL in Recovery™ Cell Culture Freezing Medium (Gibco, 12648-010). Cells were aliquoted and stored at -180°C until use.

The assay buffer consisted of DMEM without phenol red (Gibco, 11880-028) supplemented with 1X GlutaMAX (Gibco, 35050-038), 10 mM HEPES (Gibco, 15630-056) and 0.1% (w/v) ovalbumin (Sigma, A5503).

Procedure

To perform the assay, BHK hAmyR3/CRE-Luc cells were thawed, washed once in PBS and seeded in 40 μ L growth medium in a white 384-well culture plate (PerkinElmer, 6007688) at a cell density of 4.000 cells/well on the day before the experiment. The plate was incubated over night at 37°C in 5% CO₂. On the day of the assay, cells were washed three times in assay buffer. Serial dilutions (7-fold dilutions, 7 concentrations pr. compound and one well containing only assay buffer) of comparator compounds and GLP-1 receptor-amylin receptor co-agonists were performed in assay buffer often starting from approximately 10-100 nM in 96-well plates and 30 μ L of each concentration added to the 384-well assay plate with cells. The assay plate was incubated for 3 hours at 37°C in 5% CO₂ after which 30 μ L SteadyLite Plus (PerkinElmer, 6066759) was added to each well. The assay plate was sealed, incubated at room temperature with gentle shaking for 5 minutes followed by 30 minutes incubation without shaking while protected from light. Luminescence was detected on a luminescence plate reader e.g. a Synergy 2 (BioTek). The EC₅₀-values [pM] were calculated by non-linear curve fitting applying a four-parameter logistic model (Hill slope = 1.5, shared bottom response within each plate) using GraphPad Prism or by means of TIBCO Enterprise Runtime for R (TIBCO Software, Palo Alto, CA, USA).

Example 2a: Potencies of GLP-1 receptor-amylin receptor co-agonists on the GLP-1 and amylin receptors

The potencies of compounds according to the invention were tested as described in Assays 1 and 2. The results are provided in Table 4a. Details regarding the compounds, such as IUPAC nomenclature, may be found in Example 1 and the Sequence Listing.

Table 4a: *In vitro* human AmyR3 and GLP-1R potencies of GLP-1 receptor-amylin receptor co-agonists.

Compound no.	Human GLP-1R potency [EC50 (pM)]	Human AmyR3 potency [EC50 (pM)]
0007	13.3	9.4
0009	7.9	49.5
0010	5.4	37.0
0019	13.7	26.5

0026	8.8	14.0
0035	16.7	16.0
0039	34.8	59.5
0040	20.7	7.7
0042	16.2	10.8
0044	16.0	9.1
0045	19.4	25.9
0051	14.3	19.5
0052	17.0	19.4
0056	41.4	120.0
0057	59.0	164.8
0071	69.4	158.9
0072	55.9	95.1
0073	1.6	34.7
0074	0.8	60.9
0075	84.5	43.9
0076	1.7	46.4
0077	2.1	25.7
0083	5.4	46.2
0084	2.5	14.0
0085	2.2	55.8
0086	3.9	78.3
0087	23.8	97.9
0089	5.7	65.5
0090	32.6	70.9
0092	18.5	10.9
0093	7.5	8.6
0094	7.5	11.3
0095	7.4	6.6
0097	10.0	93.1
0098	7.9	15.6
0099	9.6	44.9
0100	6.8	30.4
0101	7.1	10.2

0102	14.7	9.8
0103	28.1	11.6
0105	1.1	30.8
0106	5.5	28.4
0109	3.2	19.1
0110	2.7	40.7
0111	3.0	11.5
0114	1.0	17.5
0115	2.2	22.8
0116	1.5	21.7
0120	9.1	10.6
0124	27.9	24.4
0125	8.0	12.2
0127	7.1	63.9
0128	9.7	110.3
0129	1.2	23.5
0131	16.1	14.5
0132	3.0	34.9
0141	43.3	13.2
0142	88.2	15.4
0144	7.1	16.8
0145	11.9	16.5
0146	25.7	14.0
0147	36.8	33.7
0151	10.4	9.5
0156	22.6	22.6
0157	28.7	18.4
0159	30.3	19.3
0160	11.9	27.3
0179	8.8	35.9
0180	6.4	37.0
0191	5.9	54.8
0202	10.5	28.4
0231	5.6	68.0

0232	6.4	74.2
0233	46.4	182.5
0234	38.9	38.4
0235	10.4	24.0
0254	8.9	7.7
0255	2.5	28.6
0259	15.8	12.0
0260	11.9	41.8
0261	17.4	23.9
0263	13.8	25.2
0264	6.5	11.7
0265	6.0	8.1
0266	6.3	8.5
0267	28.5	45.3
0268	26.4	14.4
0269	7.7	7.5
0270	6.3	8.6
0271	4.6	15.2
0272	9.2	11.3
0273	24.9	18.0
0280	34.3	18.0
0281	35.2	114.5
0284	41.7	32.8
0285	19.0	78.9
0292	12.0	33.8
0294	13.7	34.3
0295	4.8	32.7
0296	4.0	29.9
0297	30.2	46.0
0299	6.0	11.5
0396	4.8	32.8
0397	3.8	18.5
0411	3.5	45.9
0414	11.2	17.7

0415	10.3	16.1
0416	104.6	18.3
0417	97.8	21.0
0431	10.9	11.9
0433	30.7	12.9
0434	11.8	13.9
0435	92.4	22.3
0436	15.2	12.3
0437	12.0	11.8
0438	11.8	27.5
0439	8.3	10.7
0440	5.7	10.4
0472	2.8	24.0
0473	3.1	16.2
0474	2.9	21.8
0475	70.4	66.8
0482	36.5	31.8
0483	33.7	36.8
0484	72.8	32.5
0502	8.4	9.0
0503	6.5	7.7
0504	8.3	6.0
0506	5.8	9.8
0509	7.1	29.9
0511	8.8	8.4
0512	7.6	10.8
0516	3.3	8.9
0518	3.4	4.4
0528	3.8	22.0
0529	6.4	14.1
0539	5.0	12.1
0552	4.7	6.9
0561	17.5	10.7
0562	19.3	4.1

0564	24.5	14.5
0565	6.4	6.2
0575	5.6	8.3
0576	2.9	10.1
0577	5.6	19.1
0578	2.5	26.9
0580	7.4	9.3
0581	17.0	26.4
0629	7.9	97.5
0630	7.1	109.7
0632	18.8	148.4
0633	28.2	5.3
0634	14.7	59.4
0635	7.3	78.9
0636	10.8	8.5
0637	1.9	14.1
0638	4.6	6.9
0639	1.5	4.6
0640	2.3	6.0
0648	17.0	9.2
0654	2.9	11.3
0655	3.0	11.4
0656	4.1	13.3
0657	9.9	10.4
0658	3.1	11.0
0659	4.7	11.5
0660	3.9	12.7
0661	2.8	14.4
0662	3.0	14.6
0663	3.2	12.6

Example 2b: Potencies of compounds and comparator compounds on the GLP-1 and amylin receptors

The GLP-1 receptor (GLP-1R) and amylin receptor (AmyR3) potencies of one compound according to the invention (compound 0111) were compared to the potencies of the GLP-1 and amylin receptor agonists from which it was derived, as well as the potencies of comparator compounds. The results are shown in Table 4b. Details regarding the compounds, such as IUPAC nomenclature, may be found in Example 1 and the Sequence Listing.

Table 4b: In vitro human AmyR3 and GLP-1R potencies of reference and comparator compounds.

Compound no. or INN name	Description	Human GLP-1R potency [EC50 (pM)]	Human AmyR3 potency [EC50 (pM)]
Semaglutide	Active pharmaceutical ingredient (GLP-1 receptor agonist) in Ozempic®, Rybelsus® and Wegovy®.	5.5	-
Cagrilintide	Amylin receptor agonist (RA) in development.	-	11
Pramlintide	Active pharmaceutical ingredient (amylin RA) in Symlin®.	-	7.8
1806	Disulfide-free amylin RA. Amino acid sequence of amylin RA in compound 0111.	-	5.0
0672	Comparator compound derived from semaglutide and cagrilintide.	2.2	606
0671	Comparator compound derived from semaglutide and pramlintide.	4.5	230
0111	Compound derived from semaglutide and compound 1806.	3.0	11.5

Comparator compound 0672 comprises a GLP-1 RA having an amino acid sequence similar to that in semaglutide linked, via a short peptide linker, to an amylin RA having an amino acid sequence identical to that in cagrilintide. (In semaglutide, there is a lysine at position 20 (Lys20) and a glycine at position 31 (Gly31), relative to SEQ ID NO: 1 or SEQ ID NO: 238, and a protraction moiety attached to Lys20. The GLP-1 RA similar to semaglutide comprises an arginine at position 20 (Arg20) and a lysine residue at position 31 (Lys31), relative to SEQ ID NO: 1 or SEQ ID NO: 238, and a protraction moiety attached to Lys31.

Comparator compound 0671 comprises the same GLP-1 RA as that in comparator compound 0672 linked, via a short peptide linker, to an amylin RA having an amino acid sequence identical to that within pramlintide.

Compound 0111 comprises the same GLP-1 sequence as that used in compound 0672 and compound 0671 linked, via a short peptide linker, to an amylin RA having an amino acid sequence identical to that in compound 1806).

Example 2c: Potencies of compounds and comparator compounds on the GLP-1 and amylin receptors

The GLP-1 receptor (GLP-1R) and amylin receptor (AmyR3) potencies of two compounds according to the invention (compounds 0045 and compound 0120) were compared to the potencies of comparator compounds having selected mutations in either their GLP-1 or amylin portion.

Comparator compounds 0164 and 0167 are identical to compounds 0045 and compound 0120, respectively, except for the fact that their GLP-1 portion comprises two sequence mutations: Phe22Ile and Ile23Phe, relative to the numbering in SEQ ID NO: 1, SEQ ID NO: 238 or SEQ ID NO: 255.

Comparator compounds 0185 and 0192 are identical to compounds 0045 and 0120, respectively, except for the fact that their amylin portion comprises a single mutation in the peptide backbone: Leu12Pro, relative to the numbering in SEQ ID NO: 79, SEQ ID NO: 240 or SEQ ID NO: 256.

Comparator compounds 0015, 0016 and 0668 are further examples of compounds comprising a GLP-1 receptor agonist similar to semaglutide (as detailed in Example 2b) and an amylin receptor agonist having the amino acid sequence of cagrilintide (compounds 0015 and 0016) or an alternative protracted amylin receptor agonist (compound 0668).

The results are shown in Table 4c. Details regarding the compounds, such as IUPAC nomenclature, may be found in Example 1 and the Sequence Listing.

Table 4c: In vitro human AmyR3 and GLP-1R potencies of further comparator compounds.

Name or comparator compound no.	Special features	Human GLP-1R potency [EC50 (pM)]	Human AmyR3 potency [EC50 (pM)]
Semaglutide		5.5	-
Human GLP-1		10.8	-
Cagrilintide		-	11.0
Pramlintide		-	7.8
Salmon calcitonin		-	3.5
0164	Comparator to compound 0045 with potency impairing mutations in the GLP-1 receptor agonist.	852.0	29.6
0167	Comparator to compound 0120 with potency impairing mutations in the GLP-1 receptor agonist.	10000.0	10.9
0185	Comparator to compound 0045 with a potency impairing mutation in the amylin receptor agonist.	13.1	5659.0
0192	Comparator to compound 0120 with a potency impairing mutation in the amylin receptor agonist.	10.8	10000
0015	Compound comprising a GLP-1 RA similar to semaglutide, the amino acid sequence of cagrilintide and a single protraction moiety.	45.0	312.0
0016	Compound comprising a	509.5	1550.0

	GLP-1 RA similar to semaglutide, the amino acid sequence of cagrilintide and two protraction moieties.		
0668	Compound comprising a GLP-1 RA similar to semaglutide and an acylated amylin agonist.	455.0	10000.0

Results

The data in Table 4a show that all of the tested compounds agonise both the human AmyR3 and the GLP-1R, i.e., they are GLP-1 receptor-amylin receptor co-agonists. The compounds listed in Table 4a are all compounds according to the invention.

In contrast, the data in Table 4b show that linking the C-terminus of a potent GLP-1 receptor agonist (similar to semaglutide), via a peptide linker, to the N-terminus of a potent amylin receptor agonist (cagrilintide or pramlintide) does not result in a compound that is equally potent on these two receptors and that can necessarily function as a GLP-1 receptor-amylin receptor co-agonist (i.e., a compound according to the invention). A comparison of comparator compounds 0672 and 0671 with compound 0111 illustrates this point: compounds 0672 and 0671 both retained their potencies on the GLP-1 receptor but have impaired potency on the amylin receptor. Only compound 0111 retained full potency on both receptors and was similarly potent on both the GLP-1 and amylin receptors, relative to semaglutide and cagrilintide, respectively. Compound 0111 is considered a “balanced” compound.

The data in Table 4c show the comparator compounds’ impaired ability to agonise (activate) the GLP-1 or the amylin receptor. The Phe22Ile and Ile23Phe mutations in comparator compounds 0164 and 0167 impaired their ability to agonise (activate) the GLP-1 receptor. The Leu12Pro mutation in comparator compounds 0185 and 0192 impaired their ability to agonise (activate) the amylin receptor. This impaired ability is further illustrated by the data provided in Table 6b. Further data in Table 4c highlight the difficulty of linking a GLP-1 receptor agonist to an amylin receptor agonist. Comparator compound 0015 was found to be a poor agonist of the amylin receptor. Comparator compounds 0016 and 0668 do not activate the amylin receptor.

Example 3: Preparation of tablets for pharmacokinetic studies in Beagles

To be able to evaluate oral exposure following tablet dosing, tablet compositions comprising the test substance and SNAC (sodium N-(8-(2-hydroxybenzoyl)amino)caprylate) were prepared by mixing test substance with roller compacted SNAC and magnesium stearate as, e.g., described in WO 2019/149880. The amount of SNAC in the tablet composition was 100-300 mg, the amount of magnesium stearate in the tablet composition was 7.7 mg, and the target amount of each test substance in the tablet composition was 3-4 mg.

Example 4: Pharmacokinetic studies in Beagle dogs

Pharmacokinetic (PK) studies in Beagle dogs were conducted to determine the exposure of the GLP-1 receptor-amylin receptor co-agonist after peroral administration.

For the pharmacokinetic studies male Beagle dogs were used, 2-7, or 1 to 5, years of age and weighing approximately 10-15, such as 10-12, kg at the start of the studies. The dogs were group housed in pens (12 hours light: 12 hours dark), and fed individually and restrictedly once daily with Royal Canin Medium Adult dog (Royal Canin Products, China Branch, or Brogaarden A/S, Denmark). Exercise and group social were permitted daily, whenever possible. The dogs were used for repeated pharmacokinetic studies with a suitable wash-out period between successive dosing. An appropriate acclimatisation period was given prior to initiation of the first pharmacokinetic study. All handling, dosing and blood sampling of the animals were performed by trained and skilled staff. Before the studies the dogs were fasted overnight and from 0 to 4 h after dosing. Besides, the dogs were restricted to water 1 hour before dosing until 4 hours after dosing, but otherwise have ad libitum access to water during the whole period.

The tablets used for the p.o. studies described herein were immediate release SNAC-based tablets dosed orally.

The tablets were administered in the following manner: 10 min prior to tablet administration the dogs may be dosed subcutaneously with approximately 3 nmol/kg of SEQ ID NO: 237, tablets were placed in the back of the mouth of the dog to prevent chewing. The mouth was then closed, and 10 mL of tap water was given by a syringe or gavage to facilitate swallowing of the tablet.

Blood sampling, analysis and pharmacokinetic calculation

Blood was sampled at predefined time points for up till 240 hours, such as up till 10 hr, post dosing to adequately cover the full plasma concentration-time absorption profile of the GLP-1 agonist.

For each blood sampling time point approximately 0.8 mL of whole blood was collected in a 1.5 mL EDTA coated tube, and the tube was gently turned to allowing mixing of the sample with the EDTA. Blood samples (for example 0.8 mL) were collected in EDTA buffer (8mM) and then centrifuged at 4°C and 2000G for 10 minutes. Plasma was pipetted into Micronic tubes on dry ice, and kept at -20°C until analysis.

Blood samples were taken as appropriate, for example from a venflon in the cephalic vein in the front leg for the first 2 hours and then with syringe from the jugular vein for the rest of the time points (the first few drops were allowed to drain from the venflon to avoid heparin saline from the venflon in the sample).

Plasma concentration of the GLP-1 receptor-amylin receptor co-agonists were determined using LCMS. Individual plasma concentration-time profiles were analysed by a non-compartmental model in WinNonlin v. 5.0 or Phoenix v. 6.2 or 6.3 (Pharsight Inc., Mountain View, CA, USA), or other relevant software for PK analysis.

The individual plasma concentration-time profiles were analysed by non-compartmental analysis (NCA). The following PK parameters were calculated and reported: t_{max} , $C_{max}/Dose$ and $t_{1/2}$, as shown in Table 5.

Table 5: Pharmacokinetic parameters in dogs after oral administration

Details regarding the compounds, such as IUPAC nomenclature, may be found in Example 1 and the Sequence Listing.

Compound no.	T_{max} (hr)	C_{max}/D (kg/L)	AUC/D (h*kg/L)	$T_{1/2}$ (hr)
0045	1.3	0.27	10.90	41
0084	1.5	0.36	15.16	47
0111	1.5	0.46	15.00	44
	2.0	0.42	17.76	42
0120	2.0	0.24	7.3	24
	2.0	0.44	14.45	22
0231	1.0	0.27	0.83	6
0232	0.8	0.51	2.00	4
0233	1.5	0.3	8.43	43

0234	1.0	0.35	8.00	31
0235	1.0	0.22	3.3	21
0254	1.8	0.36	19.54	40
0259	1.5	0.48	31.73	97
0265	2.0	0.16	5.68	16
0292	2.0	0.72	65.29	110
0431	1.5	0.29	9.75	34
0436	1.5	0.3	15.00	35
0439	2.0	0.21	5.36	24
0440	1.8	0.29	5.78	32
0502	2.0	0.14	9.8	66
	2.0	0.24	13.92	61
	4.0	0.33	22.46	74
0503	4.0	0.14	8.72	41
0504	3.0	0.12	3.25	25
0506	2.0	0.14	6.53	23
0511	2.0	0.14	3.80	25
0512	2.0	0.13	3.21	21
0516	2.0	0.67	43.92	99
	2.0	0.39	34.57	103
0518	2.0	0.45	11.10	27
0528	1.5	0.5	37.01	107
0529	4.0	0.3	19.71	68
0539	3.0	0.22	8.71	30
0552	1.0	0.22	8.58	52
0575	1.5	0.34	27.60	91
0576	1.3	0.34	28.20	94
0577	1.3	0.45	37.60	93
0578	1.3	0.39	30.60	90
0580	2.0	0.38	12.77	39
0630	1.5	0.53	33.33	73
0640	1.5	0.37	7.57	29

All of the compounds tested demonstrated oral bioavailability in this model, as concentrations of the compound in plasma were detected ($C_{max}/D > 0$ and $AUC/D > 0$) following oral administration. Furthermore, the tested compounds had long half-lives (4-110 hrs), as compared to the half-lives of human GLP-1 and human amylin, measured in humans to be approximately 2–4 min and 15-20 min, respectively (Meier et al., Diabetes, 2004, 53(3): 654-662).

Example 5 - Experimental protocol for efficacy testing on appetite using an ad libitum fed rat model

10 Sprague Dawley (SD) rats from Taconic Europe, Denmark were used for the acute food intake experiments, wherein the principles of laboratory animal care were followed.

The rats had a body weight of 200-250 g at the start of the experiment. The rats arrived at least 10-14 days before the start of the experiment to allow acclimatisation to experimental settings. During this period, the animals were handled at least 2 times. Immediately upon arrival, rats were put on a reversed light cycle (dark from 11am – 11pm) and put in the HM2 system and ID chipped. Three rats were housed in each cage. During the acclimatisation period, in which the rats get used to the new light cycle and diet (Research diet, LF 10% (D12450B), the animals had free access to food and water. Since rats are normally active and eat their major part of their daily food intake during the dark period, rats were dosed in the morning right before lights were turned off. Such a set-up results in the lowest data variation and highest test sensitivity. Each dose of GLP-amylin receptor co-agonist was tested in a group of 5-8 rats. A vehicle group of 6-8 rats was included in each set of testing. In each cage there were animals from three different treatment groups (this was done if one of the cages should malfunction). The rats were dosed once according to body weight with a 0.01-3 mg/kg solution administered subcutaneously (sc). The time of dosing was recorded for each group.

After dosing, the rats were returned to their home cages, where they had access to food and water. The food consumption was recorded individually and continuously by on-line registration (HM2 system) up to 72 h. At the end of the experimental session, the animals were euthanised.

Table 6 shows acute food intake in lean rats. The results allowed assessment of *in vivo* potency and provided an indication of the compounds' duration of action. Food intake was performed in rats up to 48 h.

Table 6a: Pharmacodynamic screening model: Acute food intake in rat after a single administration of a GLP-1-amylin co-agonist (10 nmol/kg).

Details regarding the compounds, such as IUPAC nomenclature, may be found in Example 1 and the Sequence Listing.

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Compound No. (as provided in Example 1)	Food intake relative to vehicle (%) 0-24 hr, 10 nmol/kg	Food intake relative to vehicle (%) 24-48 hr, 10 nmol/kg
0007	67	105
0035	76	95
0042	52	101
0044	40	82
0045	46	80
0052	42	86
0073	47	89
0084	44	50
0095	55	95
0100	85	86
0103	80	80
0111	28	75
0116	83	82
0120	53	55
0131	70	80
0151	29	70
0231	27	76
0232	61	95
0254	71	46
0411	81	88
0434	39	105
0439	43	41
0440	81	77
0502	61	72
0504	73	53
0506	47	44

0511	61	75
0516	87	82
0518	50	45
0552	56	55
0565	39	42
0640	28	66

Table 6b: Pharmacodynamic screening model: Acute food intake in rats after a single administration of a GLP-1 receptor-amylin receptor co-agonist (compound 0120, 10 nmol/kg), compared to GLP-1-impaired compound 0167 and amylin-impaired compound 0192.

Details regarding the compounds, such as IUPAC nomenclature, may be found in Example 1 and the Sequence Listing.

Compound or comparator compound no. (as provided in Example 1)	Description	Food intake relative to vehicle (%) 0-24 hr, 10 nmol/kg	Food intake relative to vehicle (%) 24-48 hr, 10 nmol/kg
Vehicle	-	100	100
0120	GLP-1 receptor-amylin receptor co-agonist	53	55
0167	GLP-1 impaired comparator of compound 0120	85	73
0192	Amylin-impaired comparator of compound 0120	71	67

Following dosing of GLP-1 receptor-amylin receptor co-agonists to rats, it was observed that many of them induced a profound food intake inhibition, compared with vehicle treatment, as can be deduced from the data presented in Table 6a.

The data in Table 6b highlight the importance of a compound being able to activate both the GLP-1 receptor and the amylin receptor. A comparison is made with three compounds: a GLP-1 receptor-amylin receptor co-agonist (compound 0120) which is fully

potent on both receptors; comparator compound 0167, which has impaired GLP-1 activity but full amylin potency, and comparator compound 0192, which has impaired amylin activity but full GLP-1 potency (See Table 4c for the *in vitro* data). When compound 0120 is administered to rats (10 nmol/kg), it results in significant inhibition of food intake within the first 48 hours. When either compound 0167 or compound 0192 is administered to rats (10 nmol/kg), the rats' inhibition of food intake is, in both cases, significantly less than in the case of compound 0120. This indicates that both receptor systems are activated by compound 0120 and that it is a "balanced" compound.

While certain features of the invention have been illustrated and described herein, many modifications, substitutions, changes, and equivalents will now occur to those of ordinary skill in the art. It is, therefore, to be understood that the appended claims are intended to cover all such modifications and changes as fall within the true spirit of the invention.

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CLAIMS

1. A GLP-1 receptor-amylin receptor co-agonist comprising a polypeptide (R1) according to Formula I:

5 Z1-Z2-Z3,

comprising 1-3 lysine (Lys, K) residues and no disulfide bridge; wherein:

- Z1 is a GLP-1 receptor agonist peptide comprising a maximum of 9 amino acid modifications relative to SEQ ID NO: 1, with the proviso that Z1 does not comprise an isoleucine (Ile, I) at position 22;

10 • Z2 is an optional peptide linker;

- Z3 is an amylin receptor agonist peptide comprising a C-terminal amide and a maximum of 7 amino acid modifications relative to SEQ ID NO: 79, with the proviso that Z3 does not comprise a proline (Pro, P) at position 12;

15 and further comprising 1-3 protraction moieties (R2-R3), attached to the polypeptide (R1) via said 1-3 lysine (Lys, K) residues.

2. The GLP-1 receptor-amylin receptor co-agonist according to claim 1, wherein the GLP-1 receptor agonist peptide (Z1) comprises 0-3 lysine (Lys, K) residues at any one, or combination of, positions 9, 10, 12, 16, 19, 20, 21, 24, 25, 28, 29, 30 and/or 31, relative to
20 SEQ ID NO: 238 or 255.

3. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the GLP-1 receptor agonist peptide (Z1) comprises 2 lysine (Lys, K) residues at positions:

25 • 12 and either 21, 30 or 31, or

- 21 and either 30 or 31;

preferably, at positions

- 12 and either 21 or 31, or

- 21 and 31,

30 relative to SEQ ID NO: 238 or 255.

4. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the GLP-1 receptor agonist peptide (Z1) comprises 1 lysine residue (Lys, K) at any one of positions 12, 20, 21, 28, 30 or 31; preferably, 1 lysine (Lys, K)
35 residue at position 31.

5. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the peptide linker (Z2) comprises 0-3 lysine (Lys, K) residues; such as 1-2 lysine (Lys, K) residues; preferably, 1 lysine (Lys, K) residue.

5

6. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, wherein the amylin receptor agonist peptide (Z3) comprises 0-3 lysine (Lys, K) residues at any one of positions 1, 2, 3, 7, 14, 18, 20, 23 and/or 29, relative to SEQ ID NO: 240 or SEQ ID NO: 256; preferably, 1 lysine (Lys, K) residue at any one of
10 positions 14, 18, 20, 23, 29; even more preferably, 1 lysine (Lys, K) residue at position 18.

7. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, comprising a GLP-1 receptor agonist peptide (Z1) according to Formula II (SEQ ID NO: 255):

15

Xaa1-Xaa2-Glu-Gly-Thr-Phe-Thr-Ser-Xaa9-Xaa10-Ser-Xaa12-Tyr-Leu-Glu-Xaa16-
Xaa17-Ala-Xaa19-Xaa20-Xaa21-Phe-Ile-Xaa24-Xaa25-Leu-Val-Xaa28-Xaa29-
Xaa30-Xaa31;

wherein

20

Xaa1 is His (H) or Imp,
Xaa2 is Aib, Ala (A), Gly (G) or Trp (W),

Xaa9 is Asp (D) or Lys (K),

Xaa10 is Lys (K) or Val (V),

Xaa12 is Lys (K), Arg (R) or Ser (S),

25

Xaa16 is Glu (E), Gly (G) or Lys (K),

Xaa17 is Gln (Q) or Lys (K),

Xaa19 is Ala (A) or Val (V),

Xaa20 is Lys (K) or Arg (R),

Xaa21 is Glu (E) or Lys (K),

30

Xaa24 is Ala (A), Glu (E),

Xaa25 is Lys (K) or Trp (W),

Xaa28 is Lys (K) or Arg (R),

Xaa29 is Lys (K) or Gly (G),

Xaa30 is Ala (A), Gly (G), Lys (K), Arg (R) or absent,

35

Xaa31 is Ala (A), Lys (K), Gly (G), Gln (Q) or absent.

8. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, comprising an amylin receptor agonist peptide (Z3) according to Formula III (SEQ ID NO: 256):

5

Xaa1-Xaa2-Xaa3-Leu-Ser-Thr-Xaa7-Ala-Leu-Gly-Arg-Leu-Ser-Xaa14-Glu-Leu-His-
Xaa18-Leu-Xaa20-Thr-Leu-Xaa23-Arg-Thr-Glu-Thr-Gly-Xaa29-Gly-Ser-Pro,

wherein:

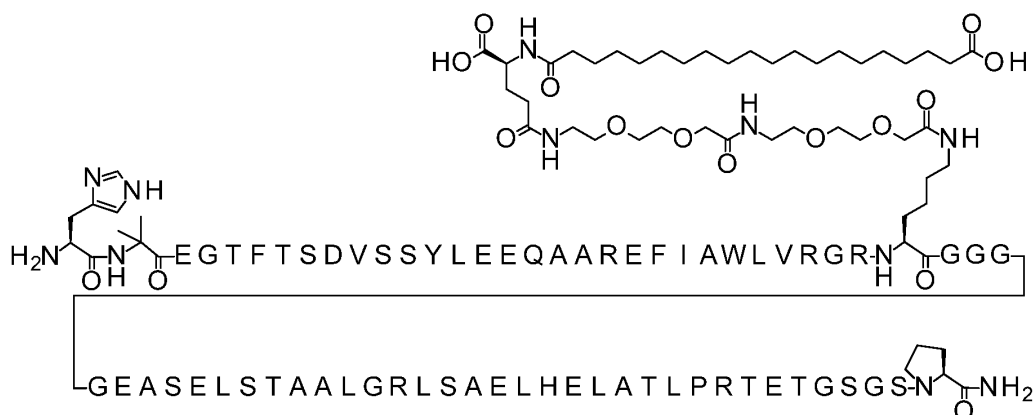
- 10 Xaa1 is Ala (A) or is absent,
Xaa2 is Lys (K) or Ser (S),
Xaa3 is Glu (E) or Arg (R),
Xaa7 is Ala (A) or Glu (E),
Xaa14 is Ala (A), or Lys (K),
Xaa18 is Glu (E), Lys (K) or Gln (Q),
15 Xaa20 is Ala (A), or Lys (K),
Xaa23 is Lys (K) or Pro (P),
Xaa29 is Ser (S) or Lys (K).

9. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, comprising:
- 20
- one to two C14-C20 diacids; more preferably, one C16-C20 diacid or two C14-C18 diacids; even more preferably, a single C18 diacid; or
 - 1-2 protractors selected from the group presented in Table 2.

- 25 10. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, comprising a polypeptide represented by any one of SEQ ID NOs 117-236; preferably, the polypeptide represented by SEQ ID NO: 153.

11. The GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims, which is any one of the compounds listed in Example 1a; preferably, any one of the compounds listed in Table 5.
- 30

12. A GLP-1 receptor-amylin receptor co-agonist, which is H-Aib-EGTFTSDVSSYLEEQAAREFIAWLVRGR-K([2-[2-[2-[2-[2-[2-[(4S)-4-carboxy-4-(17-carboxyheptadecanoylamino)butanoyl]amino]ethoxy]ethoxy]acetyl]amino]ethoxy]ethoxy]ace
- 35



13. A pharmaceutical formulation comprising a GLP-1 receptor-amylin receptor co-agonist according to any one of the preceding claims.
14. The GLP-1 receptor-amylin receptor co-agonist according to any one of claims 1-12, or the pharmaceutical formulation according to claim 13, for use as a medicament.
15. The GLP-1 receptor-amylin receptor co-agonist according to any one of claims 1-12, or the formulation according to claim 13, for use in the treatment of subjects with; an initial body mass index (BMI) of 27 or more, such as 30 or more, optionally in the presence of at least one weight-related comorbidity; diabetes, optionally in the presence of at least one weight-related comorbidity; cardiovascular disease, non-steroidal steatohepatitis and/or cognitive impairment, such as that caused by Alzheimer's disease.