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(54) Title: PTHR1 RECEPTOR COMPOUNDS

(57) Abstract: The invention relates generally to compounds which are allosteric modulators (e.g., negative and positive allosteric modulators, allosteric agonists, and ago-allosteric modulators) of the G protein coupled receptor PTHR1, also known as parathyroid hormone/parathyroid hormone related protein receptor. The PTHR1 compounds are derived from the intracellular loops and domains of the PTHR1 receptor. The invention also relates to the use of these PTHR1 receptor compounds and pharmaceutical compositions comprising the PTHR1 receptor compounds in the treatment of diseases and conditions associated with PTHR1 receptor modulation, such as osteoporosis; humoral hypercalcemia of malignancy; osteolytic and osteoblastic metastasis to bone; primary and secondary hyperparathyroidism associated increase in bone absorption; vascular calcification; psychiatric disorders and cognitive disorders associated with hyperparathyroidism; dermatological disorders; and excess hair growth.

PTHR1 RECEPTOR COMPOUNDS

RELATED APPLICATIONS

This application claims the benefit of U.S. Provisional Application No. 61/198,299, filed on November 4, 2008. The entire teachings of the above application is incorporated herein by reference.

BACKGROUND OF THE INVENTION

G protein coupled receptors (GPCRs) constitute one of the largest families of genes in the human genome. GPCRs are integral membrane signaling proteins. Hydrophobicity mapping of the amino acid sequences of G-protein coupled receptors has led to a model of the typical G-protein-coupled receptor as containing seven hydrophobic membrane-spanning regions with the amino terminal on the extracellular side of the membrane and the carboxyl terminal on the intracellular side of the membrane.

GPCRs mediate the transmission of intracellular signals ("signal transduction") by activating guanine nucleotide-binding proteins (G proteins) to which the receptor is coupled. GPCRs are activated by a wide range of endogenous stimuli, including peptides, amino acids. hormones, light, and metal ions. The following reviews are incorporated by reference: Hill, British J. Pharm 147: s27 (2006); Palczeski, Ann Rev Biochemistry 75: 743-767 (2006); Dorsham & Gutkind, Nature Reviews 7: 79-94 (2007); Kobilka & Schertler, Trends Pharmacol Sci. 2: 79-83 (2008).

GPCRs are important targets for drug discovery as they are involved in a wide range of cellular signaling pathways and are implicated in many pathological conditions (e.g., cardiovascular and mental disorders, cancer, AIDS). In fact, GPCRs are targeted by 40-50% of approved drugs, illustrating the critical importance of this class of pharmaceutical targets. Interestingly, this number represents only about 30 GPCRs, a small fraction of the total number of GPCRs thought to be relevant to human disease. Over 1000 GPCRs are known in the human genome, and GPCRs remain challenging targets from a research and development perspective in part because these amembrane bound receptors with complex pharmacology.

There remains a need for the development of new pharmaceuticals that are GPCR modulators (e.g., agonists, partial agonists, inverse agonists and antagonists and especially those that are allosteric modulators of GPCRs (e.g., negative and positive allosteric modulators, allosteric agonists, and ago-allosteric modulators).

SUMMARY OF THE INVENTION

The invention relates generally to compounds which are allosteric modulators (e.g., negative and positive allosteric modulators, allosteric agonists, and ago-allosteric modulators) of the G protein coupled receptor PTHR1, also known as parathyroid hormone/parathyroid hormone related protein receptor. The PTHR1 compounds are derived from the intracellular loops and domains of the PTHR1 receptor. The invention also relates to the use of these PTHR1 receptor compounds and pharmaceutical compositions comprising the PTHR1 receptor compounds in the treatment of diseases and conditions associated with PTHR1 receptor modulation, such as osteoporosis; humoral hypercalcemia of malignancy; osteolytic and osteoblastic metastasis to bone; primary and secondary hyperparathyroidism associated increase in bone absorption; vascular calcification; psychiatric disorders and cognitive disorders associated with hyperparathyroidism; dermatological disorders ; and excess hair growth.

More specifically, the invention relates to compounds represented by Formula I:

TLP,

or pharmaceutically acceptable salts thereof, wherein:

P is a peptide comprising at least three contiguous amino-acid residues of an intracellular i1, i2, i3 loop or an intracellular i4 domain of the PTHR1 receptor;

L is a linking moiety represented by C(O) and bonded to P at an N terminal nitrogen of an N-terminal amino-acid residue;

and T is a lipophilic tether moiety bonded to L.

The invention also relates to pharmaceutical compositions comprising one or more compounds of the invention and a carrier, and the use of the disclosed compounds and compositions in methods of treating diseases and conditions responsive to modulation (inhibition or activation) of the PTHR1 receptor.

The invention also relates to pharmaceutical compositions comprising one or more compounds of the invention and a carrier, and the use of the disclosed compounds and

compositions in methods of treating diseases and conditions responsive to modulation of the PTHR1 receptor.

DETAILED DESCRIPTION OF THE INVENTION

A description of example embodiments of the invention follows.

G PROTEIN COUPLED RECEPTORS (GPCRs)

G protein coupled receptors (GPCRs) constitute one of the largest superfamilies of genes in the human genome; these transmembrane proteins enable the cell the respond to its environment by sensing extracellular stimuli and initiating intracellular signal transduction cascades. GPCRs mediate signal transduction through the binding and activation of guanine nucleotide-binding proteins (G proteins) to which the receptor is coupled. Wide arrays of ligands bind to these receptors, which in turn orchestrate signaling networks integral to many cellular functions. Diverse GPCR ligands include small proteins, peptides, amino acids, biogenic amines, lipids, ions, odorants and even photons of light. The following reviews are incorporated by reference: Hill, British J. Pharm 147: s27 (2006); Dorsham & Gutkind, Nature Reviews 7: 79-94 (2007).

In addition to modulating a diverse array of homeostatic processes, GPCR signaling pathways are integral components of many pathological conditions (e.g., cardiovascular and mental disorders, cancer, AIDS). In fact, GPCRs are targeted by 40-50% of approved drugs illustrating the critical importance of this class of pharmaceutical targets. Interestingly, this number represents only about 30 GPCRs, a small fraction of the total number of GPCRs thought to be relevant to human disease. GPCRs are membrane bound receptors that exhibit complex pharmacological properties and remain challenging targets from a research and development perspective. Given their importance in human health combined with their prevalence (over 1000 known GPCRs in the human genome) GPCRs represent an important target receptor class for drug discovery and design.

GPCRs are integral membrane proteins that mediate diverse signaling cascades through an evolutionarily conserved structural motif. All GPCRs are thought to consist of seven hydrophobic transmembrane spanning α-helices with the amino terminus on the extracellular side of the membrane and the carboxyl terminus on the intracellular side of the membrane. The transmembrane helices are linked together sequentially by extracellular (e1,

e2, e3) and intracellular (cytoplasmic) loops (i1, i2, i3). The intracellular loops or domains are intimately involved in the coupling and turnover of G proteins and include: i1, which connects TM1-TM2; i2, connecting TM3-TM4; i3, connecting TM5-TM6; and a portion of the C-terminal cytoplasmic tail (domain 4). Due in part to the topological homology of the 7TM domains and the recent high resolution crystal structures of several GPCRs (Palczewski et al., *Science* 289, 739-45 (2000), Rasmussen, S.G. et al., *Nature* 450, 383-7 (2007)) skilled modelers are now able to predict the general boundaries of GPCR loop domains through the alignment of several related receptors. These predictions are aided in part by a number of programs used by computational biologists, including EMBOSS, ClustalW2, Kalign, and MAFFT (Multiple Alignment using Fast Fourier Transform). Importantly, many of these programs are publically available (see, for example, The European Bioinformatics Institute (EMBL-EBI) web site http://www.ebi.ac.uk/Tools/) and most have web-based interfaces.

GPCR mediated signal transduction is initiated by the binding of a ligand to its cognate receptor. In many instances GPCR ligand binding is believed to take place in a hydrophilic pocket generated by a cluster of helices near the extracellular domain. However, other ligands, such as large peptides, are thought to bind to the extracellular region of protein and hydrophobic ligands are postulated to intercalate into a receptor binding pocket through the membrane between gaps in the helices. The process of ligand binding induces conformational changes within the receptor. These changes involve the outward movement of helix 6, which in turn alters the conformations of the intracellular loops and ultimately results in a receptor form that is able to bind and activate a heterotrimeric G protein (Farrens, D., et al. *Science* 274, 768-770 (1996), Gether, U. and Kobilka, B., *J. Biol. Chem.* 273, 17979-17982 (1998)). Upon binding the receptor catalyzes the exchange of GTP for GDP in the alpha subunit of the heterotrimeric G protein, which results in a separation of the G protein from the receptor as well a dissociation of the alpha and beta/gamma subunits of the G protein itself. Notably, this process is catalytic and results in signal amplification in that activation of one receptor may elicit the activation and turnover of numerous G proteins, which in turn may regulate multiple second messenger systems. Signaling diversity is further achieved through the existence of numerous G protein types as well as differing isoforms of alpha, beta and gamma subunits. Typically, GPCRs interact with G proteins to regulate the synthesis or inhibition of intracellular second messengers such as cyclic AMP, inositol

phosphates, diacylglycerol and calcium ions, thereby triggering a cascade of intracellular events that eventually leads to a biological response.

GPCR signaling may be modulated and attenuated through cellular machinery as well as pharmacological intervention. Signal transduction may be 'switched off' with relatively fast kinetics (seconds to minutes) by a process called rapid desensitization. For GPCRs, this is caused by a functional uncoupling of receptors from heterotrimeric G proteins, without a detectable change in the total number of receptors present in cells or tissues. This process involves the phosphorylation of the receptor C terminus, which enables the protein arrestin to bind to the receptor and occlude further G protein coupling. Once bound by arrestin the receptor may be internalized into the cell and either recycled back to the cell surface or degraded. The alpha subunit of the G protein possesses intrisic GTPase activity, which attenuates signaling and promotes re-association with the beta/gamma subunits and a return to the basal state. GPCR signaling may also be modulated pharmacologically. Agonist drugs act directly to activate the receptors, whereas antagonist drugs act indirectly to block receptor signaling by preventing agonist activity through their associating with the receptor.

GPCR binding and signaling can also be modified through allosteric modulation, that is by ligands that bind not at the orthosteric binding site but through binding at an allosteric site elsewhere in the receptors. Allosteric modulators can include both positive and negative modulators of orthosteric ligand mediated activity, allosteric agonists (that act in the absence of the orthosteric ligand), and ago-allosteric modulators (ligands that have agonist activity on their own but that can also modulate the activity of the orthosteric ligand).

The large superfamily of GPCRs may be divided into subclasses based on structural and functional similarities. GPCR families include Class A Rhodopsin like, Class B Secretin like, Class C Metabotropic glutamate / pheromone, Class D Fungal pheromone, Class E cAMP receptors (Dictyostelium), the Frizzled/Smoothened family, and various orphan GPCRs. In addition, putative families include Ocular albinism proteins, Insect odorant receptors, Plant Mlo receptors, Nematode chemoreceptors, Vomeronasal receptors (VIR & V3R) and taste receptors.

PTHR1 is a class B GPCR, also called family B or secretin-like. In general, class B receptors are activated by peptide ligands typically 30 to 40 amino acids in length. Activation of these receptors results in activation of adenylyl cyclase and signal transduction through increase in cAMP as a primary signaling pathway. Class B receptors have a large N-

terminal extracellular domain with 4 very highly conserved cysteine residues. This domain is important for the binding of endogenous peptide ligands and resulting receptor activation. While these receptors signal primarily through Gs activation of adenylyl cyclase, they also couple to Gq, resulting in calcium release and may also couple to Gi/G0, which modulate adenylyl cyclase activity.

PEPTIDES

As defined herein, P is a peptide comprising at least three contiguous amino-acid residues (e.g., at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15) of an intracellular i1, i2 or i3 loop or intracellular i4 domain of the PTHR1 receptor. It is understood that, the N-terminal nitrogen of the N-terminal amino acid residue of P to which the linking moiety C(O) is bonded can be one of the at least three contiguous amino acid residues or it can be an amino acid residue distinct from the at least three contiguous amino acid residues.

Intracellular i1 loop as used herein refers to the loop which connects TM1 to TM2 and the corresponding transmembrane junctional residues.

Intracellular i2 loop as used herein refers to the loop which connects TM3 to TM4 and the corresponding transmembrane junctional residues.

Intracellular i3 loop as used herein refers to the loop which connects TM5 to TM6 and the corresponding transmembrane junctional residues.

Intracellular i4 domain as used herein refers to the C-terminal cytoplasmic tail and the transmembrane junctional residue.

In a specific embodiment, P comprises at least three, at least four, at least five, at least six, at least seven, at least eight, at least nine, at least ten, at least eleven, at least twelve, at least thirteen, at least fourteen or at least fifteen contiguous amino acid residues of the intracellular i1, i2 or i3 loop or intracellular i4 domain of the PTHR1 receptor

In a more specific embodiment, the at least three contiguous amino acids of P (e.g., at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15) are derived from the intracellular i1, i2 or i3 loop or intracellular i4 domain of the PTHR1 receptor, wherein the amino acid sequence of each loop and the i4 domain is as defined in Table 1.

Table 1:

Intracellular Loop	PTHR1 Receptor

or Domain	
i1	LAYFRRLHCTRNYIHMHLFL (SEQ ID NO: 1)
i2	YWILVEGLYLHSLIFMAFFSEKKYLWGFT (SEQ ID NO: 34)
i3	INIVRVLATKLRETNAGRCDTRQQYRKLLKSTLV (SEQ ID NO: 45)
i4	AIIYCFCNGEVQAEIKKSWSRWTLALDFKRKAR SGSSSYSYGPMVSHTSVTNVGPRVGLGLPLSPRLLP TATTNGHPQLPGHAKPGTPALETLETTPPAMAAPKDD GFLNGSCSGLDEEASGPERPPALLQEEWETVM (SEQ ID NO: 100)

It is understood that in addition to the amino acids shown in the sequences in Table 1, the intracellular loop for the i1 loop, i2 loop, i3 loop and i4 domain can also include the transmembrane junctional residues. For example, the i1 loop can include SEQ ID NO: 1 where one or more residues from the transmembrane junctional residues are included on either the C-terminus, the N-terminus or both.

In another embodiment, P comprises at least three, at least four, at least five, at least six, at least seven, at least eight, at least nine, at least ten, at least eleven, at least twelve, at least thirteen, at least fourteen, or at least fifteen contiguous amino acid residues of the i1 intracellular loop of the PTHR1 receptor.

In an even more specific embodiment, P is selected from the group consisting of SEQ ID NOS: 2-33 as listed in Table 2 below:

Table 2:

PTHR1 i-Loop	Sequence	SEQ ID NO.:
i1	LAYFRRLHSTRNYIHMH	2
i1	LAAFRRLHSTRNYIH	3
i1	LAYARRLHSTRNYIH	4
i1	LAYFARLHSTRNYIH	5
i1	LAYFKRLHSTRNYIH	6
i1	LAYFRALHSTRNYIH	7
i1	LAYFRKLHSTRNYIH	8
i1	LAYFRRAHSTRNYIH	9
i1	LAYFRRLASTRNYIH	10
i1	LAYFRRLHATRNYIH	11
i1	LAYFRRLHSARNYIH	12
i1	LAYFRRLHSTANYIH	13

i1	LAYFRRLHSTKNYIH	14
i1	LAYFRRLHSTRAYIH	15
i1	LAYFRRLHSTRNYAH	16
i1	LAYFRRLHSTRNYIA	17
i1	LAYFRRLHSTRNYIH	18
i1	GGYFRRLHSTRNYIH	19
i1	GSYFRRLHSTRNYIH	20
i1	AYFRRLHSTRNYIH	21
i1	LAYFRRLHSTRNYI	22
i1	RRLHSTRNYIHMHL	23
i1	SSYFRRLHSTRNYIH	24
i1	SGRRLHSTRNYIHMH	25
i1	LAYFRRLHSTRNY	26
i1	RRLHSTRNYIHMH	27
i1	LAYFRRLHSTRN	28
i1	FRRLHSTRNYIH	29
i1	RRLHSTRNYIHM	30
i1	YFRRLHSTRNYIH	31
i1	LAYFRRLHSTR	32
i1	RRLHSTRNYIH	33

In another specific embodiment, the at least three contiguous amino acids of P (e.g., at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15) are derived from the i2 intracellular loop of the PTHR1 receptor.

In a more specific embodiment, P is selected from the group consisting of SEQ ID NOS: 35-44 as listed in Table 3 below:

PTHR1 i-Loop	Sequence	SEQ ID NO.:
i2	LYLHSLIFMSFFSEKK	35
i2	LYLHSLIFMAFFSEKKYLWGFT	34
i2	LYLHSLIFMAFFSEKKYLWG	35
i2	LYLHSLIFMAFFSEKKYL	36
i2	LYLHSLIFMAFFSEKK	37
i2	YLHSLIFMAFFSEKKYLWGFT	38
i2	LHSLIFMAFFSEKKYLWGFT	39
i2	HSLIFMAFFSEKKYLWGFT	40
i2	HSLIFMAFFSEKKYL	41

i2	GSEKKYLWGFTVF	42
i2	GSEKKYLWGFT	43
i2	GSEKKYLWG	44

In yet another specific embodiment, P comprises at least three contiguous amino (e.g., at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15) of the i3 intracellular loop of thePTHR1 receptor.

In a more specific embodiment, P is selected from the group consisting of SEQ ID NOS: 46-99 as listed in Table 4 below:

Table 4:

PTHR1 i-Loop	Sequence	SEQ ID NO.:
i3	NIVRVLATKLRETNAGRSD	46
i3	NIVRVLATKLRETNAGR	47
i3	NIVRVLATKLRE	48
i3	SGRVLATKLRETNAGR	49
i3	SGRVLATKLRETNA	50
i3	SGRVLATKLRET	51
i3	SGRVLATKLR	52
i3	VRVLATKLRETNAGRSDTR	53
i3	RVLATKLRETNAGR	54
i3	VLATKLRETNAGRSDTRQQ	55
i3	KLRETNAGRSDTRQQYRKLL	56
i3	KLRETNAGRSDTRQQY	57
i3	KLRETNAGRSDTRQQRKLL	58
i3	KRETNAGRSDTRQQYRKLL	59
i3	RETNAGRSDTRQQYRKLLKS	60
i3	RETNAGRSDTRQQYRKLLFS	61
i3	RETNAGRSDTRQQYRKLL	62
i3	RETNAGRSDTRQQYRK	63
i3	RETNAGRSDTRQQYRF	64
i3	RETNAGRSDTRQQY	65
i3	RETNAGRSDTRQQRKLLKS	66
i3	RETNAGRSDTRQ	67
i3	TNAGRSDTRQQYRKLLKSTL	68
i3	TNAGRSDTRQQYRKLLKS	69
i3	TNAGRSDTRQQYRKLLK	70
i3	TNAGRSDTRQQYRKLLFS	71
i3	TNAGRSDTRQQYRKLLFA	72
i3	TNAGRSDTRQQYRKLLF	73

i3	TNAGRSDTRQQYRKLLA	74
i3	TNAGRSDTRQQYRKLL	75
i3	TNAGRSDTRQQYRKLA	76
i3	TNAGRSDTRQQYRKAL	77
i3	TNAGRSDTRQQYRK	78
i3	TNAGRSDTRQQYRF	79
i3	TNAGRSDTRQQYRALL	80
i3	TNAGRSDTRQQYAKLL	81
i3	TNAGRSDTRQQTRF	82
i3	TNAGRSDTRQQRKLLKSTL	83
i3	TNAGRSDTRQQARKLL	84
i3	TNAGRSDTRQAYRKLL	85
i3	TNAGRSDTRAQYRKLL	86
i3	TNAGRSDTAQQYRKLL	87
i3	TNAGRSDARQQYRKLL	88
i3	TNAGRSATRQQYRKLL	89
i3	TNAGRADTRQQYRKLL	90
i3	TNAGASDTRQQYRKLL	91
i3	TNAARSDTRQQYRKLL	92
i3	AGRSDTRQQYRKLLKS	93
i3	AGRSDTRQQYRKLLFS	94
i3	AGRSDTRQQYRKLLFA	95
i3	RSDTRQQYRKLLKS	96
i3	DTRQQYRKLLKSTL	97
i3	DTRQQYRKLLKS	98
i3	RQQYRKLLKSTL	99

In further specific embodiment, P comprises at least three contiguous amino (e.g., at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15) of the i4 intracellular domain of the PTHR1 receptor.

In a more specific embodiment, P is selected from the group consisting of SEQ ID NOS: 101-110 as listed in Table 5 below:

Table 5:

PTHR1 i-Loop	Sequence	SEQ ID NO.:
i4	EIKKSWSRWTLALDFKRKAR	101
i4	KKSWSRWTLALDFKRKAR	102
i4	NGEVQAEIKKSW	103
i4	NGEVQAEIKKSWSR	104
i4	NGEVQAEIKKSWSRWT	105

i4	NGEVQAEIKKSWSRWTLA	106
i4	NGEVQAEIKKSWSRWTLALD	107
i4	SRWTLALDFKRKAR	108
i4	SWSRWTLALDFKRKAR	109
i4	WTLALDFKRKAR	110

It is understood that the sequences presented in Tables 2-5 can be optionally functionalized at the C-terminus. Functionalized at the C-terminus means that the acid moiety present at the C-terminus is replaced by some other functional group. Suitable functional groups include $-C(O)N(R_2)_2$, $-C(O)OR_3$, or $C(O)NHC(O)OR_2$, where R_2 is hydrogen or a (C_1-C_{10}) alkyl group and R_3 is a (C_1-C_{10}) alkyl group.

It is understood that as long as P comprises the indicated number of contiguous amino acids residues from the PTHR1 intracellular loop (i1, i2 or i3) or domain (i4) from which it is derived, the remainder of the peptide, if present, can be selected from:

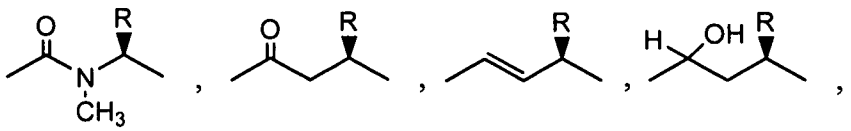
(a) any natural amino acid residue, unnatural amino acid residue or a combination thereof;

(b) a peptide sequence comprising natural amino acid residues, non-natural amino acid residues and combinations thereof;

(c) a peptide sequence according to (b) comprising one or more peptide backbone modifications;

(d) a peptide sequence according to (c) comprising one or more retro-inverso peptide linkages;

(e) a peptide sequence according to (c) wherein one or more peptide bonds are



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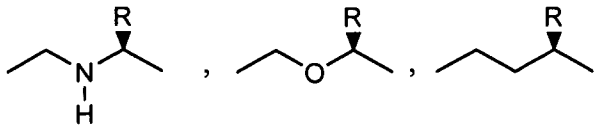


or a combination thereof;

(f) a peptide sequence according to (c) comprising one or more depsipeptide linkages, wherein the amide linkage is replaced with an ester linkage; and

(g) a peptide sequence according to (c) comprising one or more conformational restrictions; and

(h) a peptide sequence according to (c) comprising one or more of (d)-(g).

Furthermore, it is understood that even within the indicated number of contiguous amino acid residues derived from the GPCR intracellular loop (i1, i2 or i3) or domain (i4), there can be: peptide backbone modifications such as, but not limited to, those described in (e) above; retro-inverso peptide linkages; despsipeptide linkages; conformational restrictions; or a combination thereof.

It is noted that P of Formula I can be optionally functionalized at the C-terminus. Functionalized at the C-terminus means that the acid moiety present at the C-terminus is replaced by some other functional group. Suitable functional groups include $-C(O)N(R_2)_2$, $-C(O)OR_3$, or $C(O)NHC(O)OR_2$, where R_2 is hydrogen or a (C_1-C_{10}) alkyl group and R_3 is a (C_1-C_{10}) alkyl group . Functionalization of the C-terminus can result from the methods used to prepare.

Peptidomimetic as used herein refers to a compound comprising non-peptidic structural elements in place of a peptide sequence.

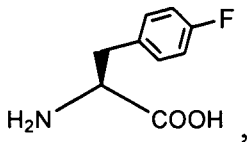
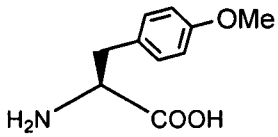
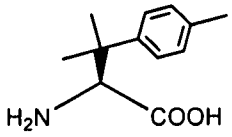
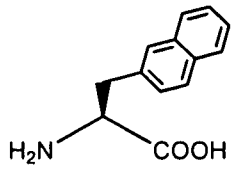
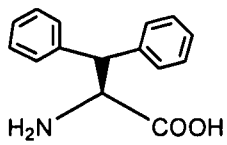
As used herein, the term "amino acid" includes both a naturally occurring amino acid and a non-natural amino acid.

As used herein, the term "naturally occurring amino acid" means a compound represented by the formula NH_2-CHR-COOH, wherein R is the side chain of a naturally occurring amino acids such as lysine, arginine, serine, tyrosine etc. as shown in the Table below.

Table of Common Naturally Occurring Amino Acids

	Amino acid	Three letter code	One letter code
Non-polar; neutral at pH 7.4	alanine	Ala	A
	isoleucine	Ile	I
	leucine	Leu	L
	methionine	Met	M
	phenylalanine	Phe	F
	proline	Pro	P
	tryptophan	Trp	W
	valine	Val	V

Polar, uncharged at pH 7.0	asparagine	Asn	N
	cysteine	Cys	C
	glycine	Gly	G
	glutamine	Gln	Q
	serine	Ser	S
	threonine	Thr	T
	tyrosine	Tyr	Y
Polar; charged at pH 7	glutamic acid	Glu	E
	arginine	Arg	R
	aspartic acid	Asp	D
	histidine	His	H
	lysine	Lys	K

"Non-natural amino acid" means an amino acid for which there is no nucleic acid codon. Examples of non-natural amino acids include, for example, the D-isomers of the natural α-amino acids such as D-proline (D-P, D-Pro) as indicated above; natural α-amino acids with non-natural side chains (e.g.,  ,  ,  ,  , and  related to phenylalanine); Aib (aminobutyric acid), bAib (3-aminoisobutyric acid), Nva (norvaline), β-Ala, Aad (2-aminoadipic acid), bAad (3-aminoadipic acid), Abu (2-aminobutyric acid), Gaba (γ-aminobutyric acid), Acp (6-aminocaproic acid), Dbu (2,4-diaminobutryic acid), α-aminopimelic acid, TMSA (trimethylsilyl-Ala), aIle (allo-isoleucine), Nle (norleucine), tert-Leu, Cit (citrulline), Orn (ornithine, O), Dpm (2,2'-diaminopimelic acid), Dpr (2,3-diaminopropionic acid), α or .β-Nal, Cha (cyclohexyl-Ala), hydroxyproline, Sar (sarcosine), and the like.

Unnatural amino acids also include cyclic amino acids; and amino acid analogs, for example, N^{α}-alkylated amino acids such as MeGly (N^{α}-methylglycine), EtGly (N^{α}-ethylglycine) and EtAsn (N^{α}-ethylasparagine); and amino acids in which the α-carbon bears two side-chain substituents. As with the natural amino acids, the residues of the unnatural

amino acids are what are left behind when the unnatural amino acid becomes part of a peptide sequence as described herein.

Amino acid residues are amino acid structures as described above that lack a hydrogen atom of the amino group or the hydroxyl moiety of the carboxyl group or both resulting in the units of a peptide chain being amino-acid residues.

TETHERS (T)

T of Formula I is a lipohilic tether moiety which imparts lipophilicity to the PTHR1 receptor compounds of the invention. The lipophilicity which T imparts, can promote penetration of the PTHR1 receptor compounds into the cell membrane and tethering of the PTHR1 receptor compounds to the cell membrane. As such, the lipophilicity imparted by T can facilitate interaction between the PTHR1 receptor compounds of the invention and the cognate receptor.

The relative lipophilicity of compounds suitable for use as the lipophilic tether moiety of Formula I can be quantified by measuring the amount of the compound that partitions into an organic solvent layer (membrane-like) vs. an aqueous solvent layer (analogous to the extracellular or cytoplasmic environment). The partition coefficient in a mixed solvent composition, such as octanol/water or octanol/PBS, is the ratio of compound found at equilibrium in the octanol vs. the aqueous solvent (Partition coeff P = $[compound]_{octanol}/[compound]_{aqueous}$). Frequently, the partition coefficient is expressed in logarithmic form, as the log P. Compounds with greater lipophilicity have a more positive log P than more hydrophilic compounds and tend to interact more strongly with membrane bilayers.

Computational programs are also available for calculating the partition coefficient for compounds suitable for use as the lipophilic tether moiety (T). In situations where the chemical structure is being varied in a systematic manner, for example by adding additional methylene units ($-CH_2-$) onto to an existing alkyl group, the trend in log P can be calculated using, for example, ChemDraw (CambridgeSoft, Inc).

In one embodiment, T is an optionally substituted (C_6-C_{30})alkyl, (C_6-C_{30})alkenyl, (C_6-C_{30})alkynyl wherein 0-3 carbon atoms are replaced with oxygen, sulfur, nitrogen or a combination thereof.

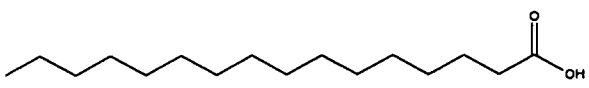
In a specific embodiment, the $(C_6\text{-}C_{30})$alkyl, $(C_6\text{-}C_{30})$alkenyl, $(C_6\text{-}C_{30})$alkynyl are substituted at one or more substitutable carbon atoms with halogen, -CN, -OH, $-NH_2$, NO_2, $-NH(C_1\text{-}C_6)$alkyl, $-N((C_1\text{-}C_6)alkyl)_2$, $(C_1\text{-}C_6)$alkyl, $(C_1\text{-}C_6)$haloalkyl, $(C_1\text{-}C_6)$alkoxy, $(C_1\text{-}C_6)$haloalkoxy, aryloxy, $(C_1\text{-}C_6)$alkoxycarbonyl, $-CONH_2$, $-OCONH_2$, $-NHCONH_2$, $-N(C_1\text{-}C_6)alkylCONH_2$, $-N(C_1\text{-}C_6)alkylCONH(C_1\text{-}C_6)$alkyl, $-NHCONH(C_1\text{-}C_6)$alkyl, $-NHCON((C_1\text{-}C_6)alkyl)_2$, $-N(C_1\text{-}C_6)alkylCON((C_1\text{-}C_6)alkyl)_2$, $-NHC(S)NH_2$, $-N(C_1\text{-}C_6)alkylC(S)NH_2$, $-N(C_1\text{-}C_6)alkylC(S)NH(C_1\text{-}C_6)$alkyl, $-NHC(S)NH(C_1\text{-}C_6)$alkyl, $-NHC(S)N((C_1\text{-}C_6)alkyl)_2$, $-N(C_1\text{-}C_6)alkylC(S)N((C_1\text{-}C_6)alkyl)_2$, $-CONH(C_1\text{-}C_6)$alkyl, $-OCONH(C_1\text{-}C_6)$alkyl $-CON((C_1\text{-}C_6)alkyl)_2$, $-C(S)(C_1\text{-}C_6)$alkyl, $-S(O)_p(C_1\text{-}C_6)$alkyl, $-S(O)_pNH_2$, $-S(O)_pNH(C_1\text{-}C_6)$alkyl, $-S(O)_pN((C_1\text{-}C_6)alkyl)_2$, $-CO(C_1\text{-}C_6)$alkyl, $-OCO(C_1\text{-}C_6)$alkyl, $-C(O)O(C_1\text{-}C_6)$alkyl, $-OC(O)O(C_1\text{-}C_6)$alkyl, -C(O)H or $-CO_2H$; and p is 1 or 2.

In a specific embodiment, T is selected from the group consisting of: $CH_3(CH_2)_9OPh$-, $CH_3(CH_2)_6C{=}C(CH_2)_6$, $CH_3(CH_2)_{11}O(CH_2)_3$, $CH_3(CH_2)_9O(CH_2)_2$ and $CH_3(CH_2)_{13}$.

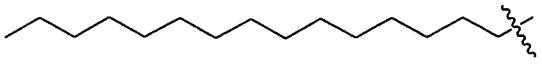
In a specific embodiment, T is selected from the group consisting of: $CH_3(CH_2)_{16}$, $CH_3(CH_2)_{15}$, $CH_3(CH_2)_{14}$, $CH_3(CH_2)_{13}$,$CH_3(CH_2)_{12}$, $CH_3(CH_2)_{11}$, $CH_3(CH_2)_{10}$, $CH_3(CH_2)_9$, $CH_3(CH_2)_8$, $CH_3(CH_2)_9OPh$-, $CH_3(CH_2)_6C{=}C(CH_2)_6$, $CH_3(CH_2)_{11}O(CH_2)_3$, and $CH_3(CH_2)_9O(CH_2)_2$ and $CH_3(CH_2)_{13}$.

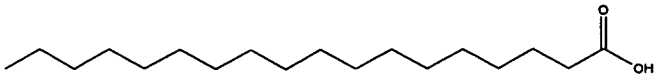
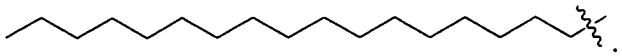
It is understood that the lipophilic moiety (T) of Formula I can be derived from precursor liphophilic compounds (e.g., fatty acids and bile acids). As used herein, "derived from" with regard to T, means that T is derived from a precursor lipophilic compound and that reaction of the precursor lipophilic compound in preparing the APJ receptor compounds of Formula I, results in a lipophilic tether moiety represented by T in Formula I that is structurally modified in comparison to the precursor lipophilic compound.

For example, the lipophilic tether moiety, T of Formula I, can be derived from a fatty acid or a bile acid. It is understood that in accordance with Formula I, when T is derived from a fatty acid (i.e., a fatty acid derivative) it is attached to L-P at the carbon atom alpha to the carbonyl carbon of the acid functional group in the fatty acid from which it is derived. For example, when T is derived from palmitic acid,

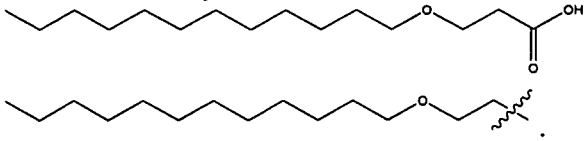


, T of Formula I has the following structure:

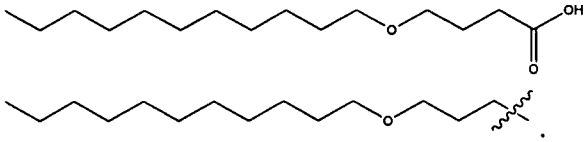
 . Similarly, when T is derived from stearic acid,

 , T of Formula I has the following structure:  .

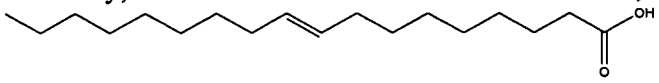
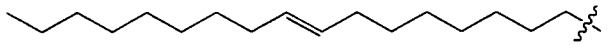
Similarly, when T is derived from 3-(dodecyloxy)propanoic acid,

 , T of Formula I has the following structure:

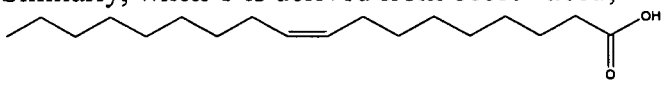
Similarly, when T is derived from 4-(undecyloxy)butanoic acid,

 , T of Formula I has the following structure:

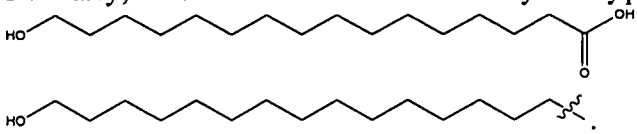
Similarly, when T is derived from elaidic acid,

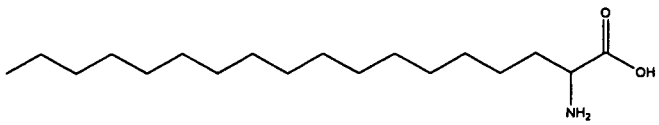
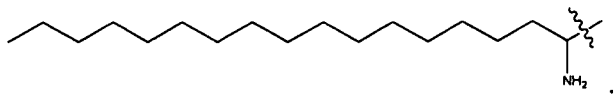
 , T of Formula I has the following structure:  .

Similarly, when T is derived from oleic acid,

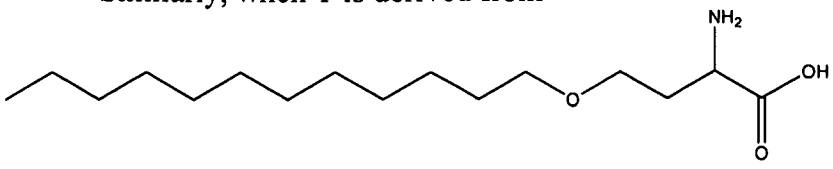
 , T of Formula I has the following structure:  .

Similarly, when T is derived from 16-hydroxypalmitic acid,

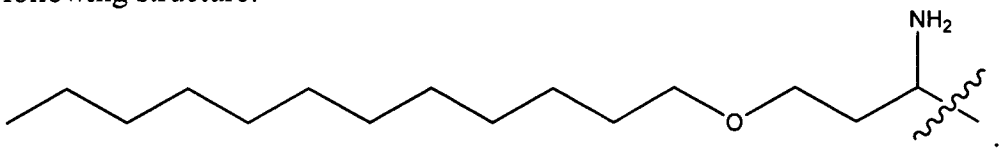
 , T of Formula I has the following structure:

Similarly, when T is derived from 2-aminooctadecanoic acid  , T of Formula I has the following structure:  .

Similarly, when T is derived from 2-amino-4-(dodecyloxy)butanoic acid,



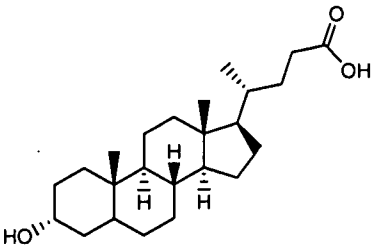
, T of Formula I has the following structure:



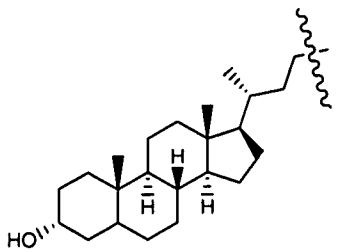
In a further embodiment, T is derived from a fatty acid. In a specific embodiment, T is derived from a fatty acid selected from the group consisting of: butyric acid, caproic acid, caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, arachidic acid, behenic acid, and lignoceric acid.

In another specific embodiment, T is derived from a fatty acid selected from the group consisting of: myristoleic acid, palmitoleic acid, oleic acid, linoleic acid, α-linolenic acid, arachidonic acid, eicosapentaenoic acid, erucic acid, docosahexaenoic acid

In another embodiment, T of Formula I can be derived from a bile acid. Similar to the embodiment where T is a fatty acid derivative, it is understood that in accordance with Formula I, when T is derived from a bile acid (i.e., a bile acid derivative) it is attached to L-P at the carbon atom alpha to the carbonyl carbon of the acid functional group in the bile acid from which it is derived. For example, when T is derived from



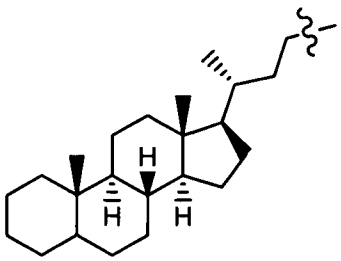
lithocholic acid, , T of Formula I has the following structure:



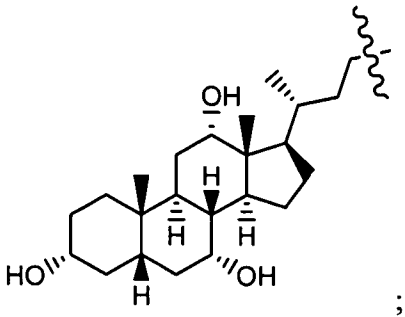
In a further embodiment, T is derived from a bile acid. In a specific embodiment, T is derived from a bile acid selected from the group consisting of: lithocholic acid,

chenodeoxycholic acid, deoxycholic acid, cholanic acid, cholic acid, ursocholic acid, ursodeoxycholic acid, isoursodeoxycholic acid, lagodeoxycholic acid, dehydrocholic acid, hyocholic acid, hyodeoxycholic acid and the like.

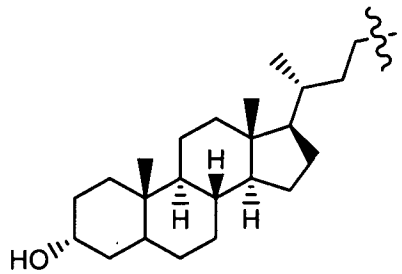
For example, T is selected from:



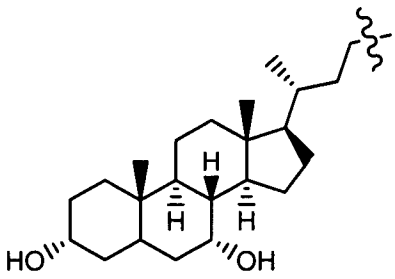
;



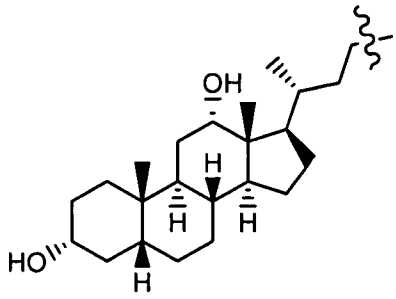
;



;

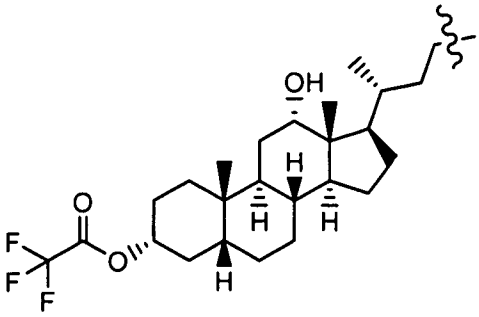


; and



.

In another further embodiment, T is derived from a bile acid described above that has been modified at other than the acid functional group. For example, T can be derived from any of the bile acids described above, where the hydroxy position has been modified to form an ester or a halo ester. For example, T can be:



.

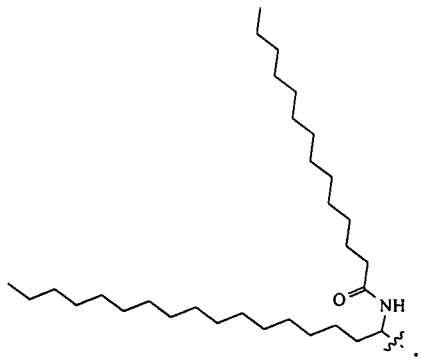
Other lipophilic moieties suitable for use as the lipophilic membrane tether, T, of Formula I, include but are not limited to steroids. Suitable steroids include, but are not limited to, sterols; progestagens; glucocorticoids; mineralcorticoids; androgens; and estrogens. Generally any steroid capable of attachment or which can be modified for incorporation into Formula I can be used. It is understood that the lipophilic membrane

tether, T, may be slightly modified from the precursor lipophilic compound as a result of incorporation into Formula I.

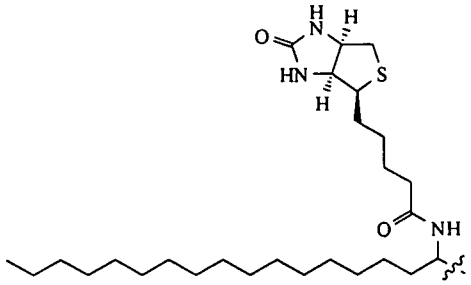
Suitable sterols for use in the invention at T, include but are not limited to: cholestanol, coprostanol, cholesterol, epicholesterol, ergosterol, ergocalciferol, and the like. Preferred sterols are those that provide a balance of lipophilicity with water solubility.

Suitable progestagens include, but are not limited to progesterone. Suitable glucocorticoids include, but are not limited to cortisol. Suitable mineralcorticoids include, but are not limited to aldosterone. Suitable androgens include, but are not limited to testosterone and androstenedione. Suitable estrogens include, but are not limited to estrone and estradiol.

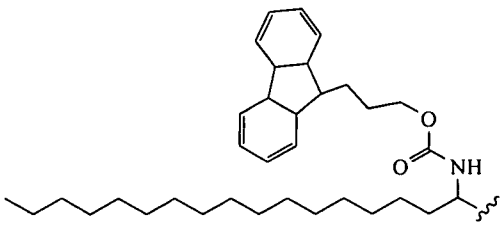
In another specific embodiment, T can be derived from 2-tetradecanamideooctadecanoid acid. Similar to the embodiment where T is a fatty acid derivative, it is understood that in accordance with Formula I, when T is derived from 2-tetradecanamideooctadecanoid acid it is attached to L-P at the carbon atom alpha to the carbonyl carbon of the acid functional group in the bile acid from which it is derived. For example, when T is derived from 2-tetradecanamideooctadecanoid acid, the tether is:



In another embodiment, T of Formula I can be derived from 2-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamido)octadecanoic acid. For example, when T is derived from 2-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamido)octadecanoic acid, the tether is:



In yet another embodiment, T of Formula I can be:



It is understood, that the compounds can contain one of more tether moieties. In certain aspects, the tether moieties are the same. In other embodiments, the tether moieties are different.

COMPOUNDS (T-L-P)

In a first aspect, the GPCR Compound of the invention is represented by Formula I:

T-L-P,

or pharmaceutically acceptable salts thereof, wherein:

P is a peptide comprising at least three contiguous amino-acid residues of an intracellular i1, i2, i3 loop or an intracellular i4 domain of the PTHR1 receptor;

L is a linking moiety represented by C(O) and bonded to P at an N terminal nitrogen of an N-terminal amino-acid residue;

and T is a lipophilic tether moiety bonded to L.

In a second aspect, P comprises at least six contiguous amino acid residues.

In a third aspect, P comprises at least 3 contiguous amino acids of the i1 loop.

In a specific embodiment of the third aspect, the i1 loop of the PTHR1 receptor from which P is derived has the following sequence: LAYFRRLHCTRNYIHMHLFL (SEQ ID NO: 1)

In another embodiment of the third aspect, P is a sequence selected from:

LAYFRRLHSTRNYIHMH	(SEQ ID NO: 2);
LAAFRRLHSTRNYIH	(SEQ ID NO: 3);
LAYARRLHSTRNYIH	(SEQ ID NO: 4);
LAYFARLHSTRNYIH	(SEQ ID NO: 5);
LAYFKRLHSTRNYIH	(SEQ ID NO: 6);
LAYFRALHSTRNYIH	(SEQ ID NO: 7);
LAYFRKLHSTRNYIH	(SEQ ID NO: 8);
LAYFRRAHSTRNYIH	(SEQ ID NO: 9);
LAYFRRLASTRNYIH	(SEQ ID NO: 10);
LAYFRRLHATRNYIH	(SEQ ID NO: 11);
LAYFRRLHSARNYIH	(SEQ ID NO: 12);
LAYFRRLHSTANYIH	(SEQ ID NO: 13);
LAYFRRLHSTKNYIH	(SEQ ID NO: 14);
LAYFRRLHSTRAYIH	(SEQ ID NO: 15);
LAYFRRLHSTRNYAH	(SEQ ID NO: 16);
LAYFRRLHSTRNYIA	(SEQ ID NO: 17);
LAYFRRLHSTRNYIH	(SEQ ID NO: 18);
GGYFRRLHSTRNYIH	(SEQ ID NO: 19);
GSYFRRLHSTRNYIH	(SEQ ID NO: 20);
AYFRRLHSTRNYIH	(SEQ ID NO: 21);
LAYFRRLHSTRNYI	(SEQ ID NO: 22);
RRLHSTRNYIHMHL	(SEQ ID NO: 23);
SSYFRRLHSTRNYIH	(SEQ ID NO: 24);
SGRRLHSTRNYIHMH	(SEQ ID NO: 25);
LAYFRRLHSTRNY	(SEQ ID NO: 26);
RRLHSTRNYIHMH	(SEQ ID NO: 27);
LAYFRRLHSTRN	(SEQ ID NO: 28);
FRRLHSTRNYIH	(SEQ ID NO: 29);
RRLHSTRNYIHM	(SEQ ID NO: 30);
YFRRLHSTRNYIH	(SEQ ID NO: 31);
LAYFRRLHSTR	(SEQ ID NO: 32); and
RRLHSTRNYIH	(SEQ ID NO: 33).

In a fourth aspect, P comprises at least 3 contiguous amino acids of the i2 loop.

In a specific embodiment of the fourth aspect, the i2 loop of the PTHR1 receptor from which P is derived has the following sequence:

YWILVEGLYLHSLIFMAFFSEKKYLWGFT (SEQ ID NO: 34).

In another embodiment of the fourth aspect, P is a sequence selected from:

LYLHSLIFMSFFSEKK	(SEQ ID NO: 35);
LYLHSLIFMAFFSEKKYLWGFT	(SEQ ID NO: 34);
LYLHSLIFMAFFSEKKYLWG	(SEQ ID NO: 35);
LYLHSLIFMAFFSEKKYL	(SEQ ID NO: 36);
LYLHSLIFMAFFSEKK	(SEQ ID NO: 37);
YLHSLIFMAFFSEKKYLWGFT	(SEQ ID NO: 38);
LHSLIFMAFFSEKKYLWGFT	(SEQ ID NO: 39);
HSLIFMAFFSEKKYLWGFT	(SEQ ID NO: 40);
HSLIFMAFFSEKKYL	(SEQ ID NO: 41);
GSEKKYLWGFTVF	(SEQ ID NO: 42);
GSEKKYLWGFT	(SEQ ID NO: 43); and
GSEKKYLWG	(SEQ ID NO: 44).

In a fifth aspect, P comprises at least 3 contiguous amino acids of the i3 loop.

In a specific embodiment of the fifth aspect, the i3 loop of the PTHR1 receptor from which P is derived has the following sequence:

INIVRVLATKLRETNAGRCDTRQQYRKLLKSTLV (SEQ ID NO: 45).

In another embodiment of the fifth aspect, P is a sequence selected from:

NIVRVLATKLRETNAGRSD	(SEQ ID NO: 46);
NIVRVLATKLRETNAGR	(SEQ ID NO: 47);
NIVRVLATKLRE	(SEQ ID NO: 48);
SGRVLATKLRETNAGR	(SEQ ID NO: 49);
SGRVLATKLRETNA	(SEQ ID NO: 50);
SGRVLATKLRET	(SEQ ID NO: 51);
SGRVLATKLR	(SEQ ID NO: 52);
VRVLATKLRETNAGRSDTR	(SEQ ID NO: 53);
RVLATKLRETNAGR	(SEQ ID NO: 54);
VLATKLRETNAGRSDTRQQ	(SEQ ID NO: 55);
KLRETNAGRSDTRQQYRKLL	(SEQ ID NO: 56);
KLRETNAGRSDTRQQY	(SEQ ID NO: 57);
KLRETNAGRSDTRQQRKLL	(SEQ ID NO: 58);
KRETNAGRSDTRQQYRKLL	(SEQ ID NO: 59);

RETNAGRSDTRQQYRKLLKS (SEQ ID NO: 60);
RETNAGRSDTRQQYRKLLFS (SEQ ID NO: 61);
RETNAGRSDTRQQYRKLL (SEQ ID NO: 62);
RETNAGRSDTRQQYRK (SEQ ID NO: 63);
RETNAGRSDTRQQYRF (SEQ ID NO: 64);
RETNAGRSDTRQQY (SEQ ID NO: 65);
RETNAGRSDTRQQRKLLKS (SEQ ID NO: 66);
RETNAGRSDTRQ (SEQ ID NO: 67);
TNAGRSDTRQQYRKLLKSTL (SEQ ID NO: 68);
TNAGRSDTRQQYRKLLKS (SEQ ID NO: 69);
TNAGRSDTRQQYRKLLK (SEQ ID NO: 70);
TNAGRSDTRQQYRKLLFS (SEQ ID NO: 71);
TNAGRSDTRQQYRKLLFA (SEQ ID NO: 72);
TNAGRSDTRQQYRKLLF (SEQ ID NO: 73);
TNAGRSDTRQQYRKLLA (SEQ ID NO: 74);
TNAGRSDTRQQYRKLL (SEQ ID NO: 75);
TNAGRSDTRQQYRKLA (SEQ ID NO: 76);
TNAGRSDTRQQYRKAL (SEQ ID NO: 77);
TNAGRSDTRQQYRK (SEQ ID NO: 78);
TNAGRSDTRQQYRF (SEQ ID NO: 79);
TNAGRSDTRQQYRALL (SEQ ID NO: 80);
TNAGRSDTRQQYAKLL (SEQ ID NO: 81);
TNAGRSDTRQQTRF (SEQ ID NO: 82);
TNAGRSDTRQQRKLLKSTL (SEQ ID NO: 83);
TNAGRSDTRQQARKLL (SEQ ID NO: 84);
TNAGRSDTRQAYRKLL (SEQ ID NO: 85);
TNAGRSDTRAQYRKLL (SEQ ID NO: 86);
TNAGRSDTAQQYRKLL (SEQ ID NO: 87);
TNAGRSDARQQYRKLL (SEQ ID NO: 88);
TNAGRSATRQQYRKLL (SEQ ID NO: 89);
TNAGRADTRQQYRKLL (SEQ ID NO: 90);
TNAGASDTRQQYRKLL (SEQ ID NO: 91);
TNAARSDTRQQYRKLL (SEQ ID NO: 92);
AGRSDTRQQYRKLLKS (SEQ ID NO: 93);
AGRSDTRQQYRKLLFS (SEQ ID NO: 94);
AGRSDTRQQYRKLLFA (SEQ ID NO: 95);
RSDTRQQYRKLLKS (SEQ ID NO: 96);
DTRQQYRKLLKSTL (SEQ ID NO: 97);
DTRQQYRKLLKS (SEQ ID NO: 98); and
RQQYRKLLKSTL (SEQ ID NO: 99).

In a sixth aspect, P comprises at least 3 contiguous amino acids of the i4 domain.

In a specific embodiment of the sixth aspect, the i4 domain of the PTHR1 receptor from which P is derived has the following sequence:

AIIYCFCNGEVQAEIKKSWSRWTLALDFKRKAR
SGSSSYSYGPMVSHTSVTNVGPRVGLGLPLSPRLLP
TATTNGHPQLPGHAKPGTPALETLETTPPAMAAPKDD
GFLNGSCSGLDEEASGPERPPALLQEEWETVM (SEQ ID NO: 100)

In another embodiment of the sixth aspect, P is a sequence selected from:

EIKKSWSRWTLALDFKRKAR	(SEQ ID NO: 101);
KKSWSRWTLALDFKRKAR	(SEQ ID NO: 102);
NGEVQAEIKKSW	(SEQ ID NO: 103);
NGEVQAEIKKSWSR	(SEQ ID NO: 104);
NGEVQAEIKKSWSRWT	(SEQ ID NO: 105);
NGEVQAEIKKSWSRWTLA	(SEQ ID NO: 106);
NGEVQAEIKKSWSRWTLALD	(SEQ ID NO: 107);
SRWTLALDFKRKAR	(SEQ ID NO: 108);
SWSRWTLALDFKRKAR	(SEQ ID NO: 109); and
WTLALDFKRKAR	(SEQ ID NO: 110).

In a seventh aspect, T is an optionally substituted $(C_6\text{-}C_{30})$alkyl, $(C_6\text{-}C_{30})$alkenyl, $(C_6\text{-}C_{30})$alkynyl, wherein 0-3 carbon atoms are replaced with oxygen, sulfur, nitrogen or a combination thereof. This value of T is applicable to the first, second, third, fourth, fifth and sixth aspects and the specific (i.e., specific, more specific and most specific) embodiments of same.

In a specific embodiment of the seventh aspect, T is selected from: $CH_3(CH_2)_{16}$, $CH_3(CH_2)_{15}$, $CH_3(CH_2)_{14}$, $CH_3(CH_2)_{13}$, $CH_3(CH_2)_{12}$, $CH_3(CH_2)_{11}$, $CH_3(CH_2)_{10}$, $CH_3(CH_2)_9$, $CH_3(CH_2)_8$, $CH_3(CH_2)_9OPh$-, $CH_3(CH_2)_6C{=}C(CH_2)_6$, $CH_3(CH_2)_{11}O(CH_2)_3$, and $CH_3(CH_2)_9O(CH_2)_2$.

In another specific embodiment of the seventh aspect, T is a fatty acid derivative.

In a more specific embodiment of the seventh aspect, the fatty acid is selected from the group consisting of: butyric acid, caproic acid, caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, arachidic acid, behenic acid, lignoceric acid,

myristoleic acid, palmitoleic acid, oleic acid, linoleic acid, α-linolenic acid, arachidonic acid, eicosapentaenoic acid, erucic acid, docosahexaenoic acid.

In an eighth aspect, T is a bile acid derivative. This value of T is applicable to the first, second, third, fourth, fifth and sixth aspects and the specific (i.e., specific, more specific and most specific) embodiments of same.

In a specific embodiment of the eighth aspect, the bile acid is selected from the group consisting of: lithocholic acid, chenodeoxycholic acid, deoxycholic acid, cholanic acid, cholic acid, ursocholic acid, ursodeoxycholic acid, isoursodeoxycholic acid, lagodeoxycholic acid, dehydrocholic acid, hyocholic acid, and hyodeoxycholic acid.

In a ninth aspect, T is selected from sterols; progestagens; glucocorticoids; mineralcorticoids; androgens; and estrogens. This value of T is applicable to the first, second, third, fourth, fifth and sixth aspects and the specific (i.e., specific, more specific and most specific) embodiments of same.

In a tenth aspect, T-L of Formula I is represented by a moiety selected from the group consisting of:

$CH_3(CH_2)_{15}$–C(O);

$CH_3(CH_2)_{13}$– C(O);

$CH_3(CH_2)_9O(CH_2)_2C(O)$;

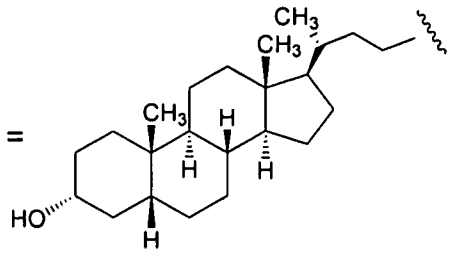
$CH_3(CH_2)_{10}O(CH_2)_2C(O)$;

$CH_3(CH_2)_6C{=}C(CH_2)_6$-C(O);

LCA-C(O); and

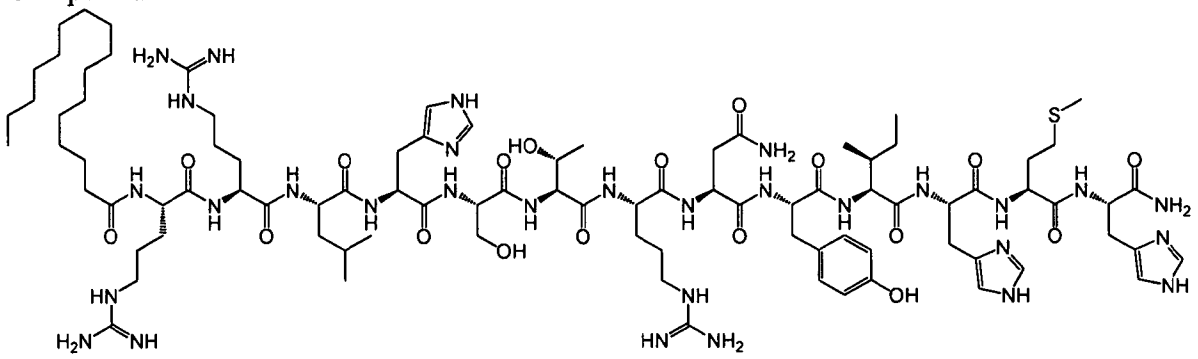
$CH_3(CH_2)_9OPh$-C(O) wherein

LCA =

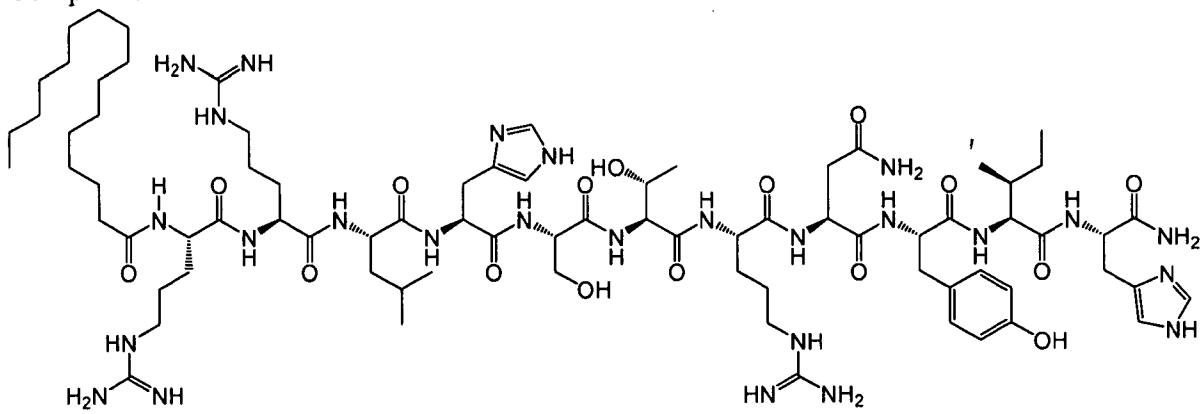


In yet another embodiment, a GPCR compound of the invention is selected from one of the following compounds or a pharmaceutically acceptable salt thereof:

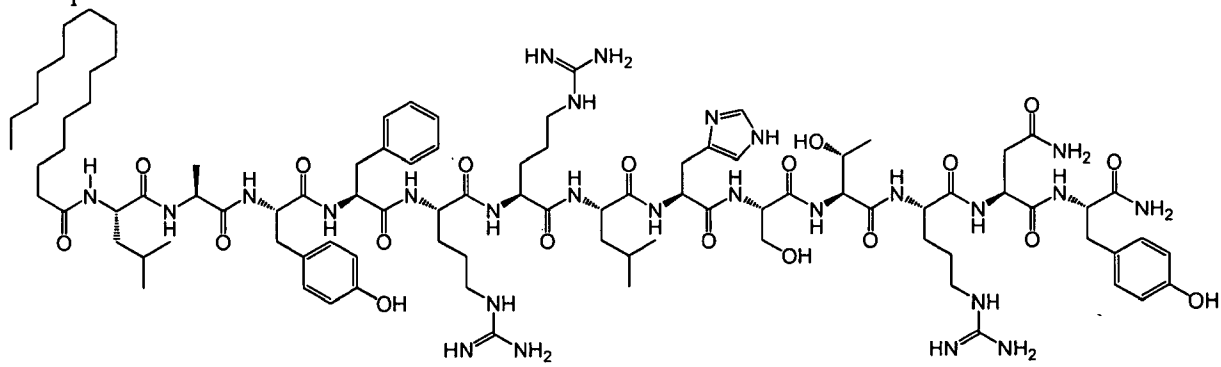
Compound 1



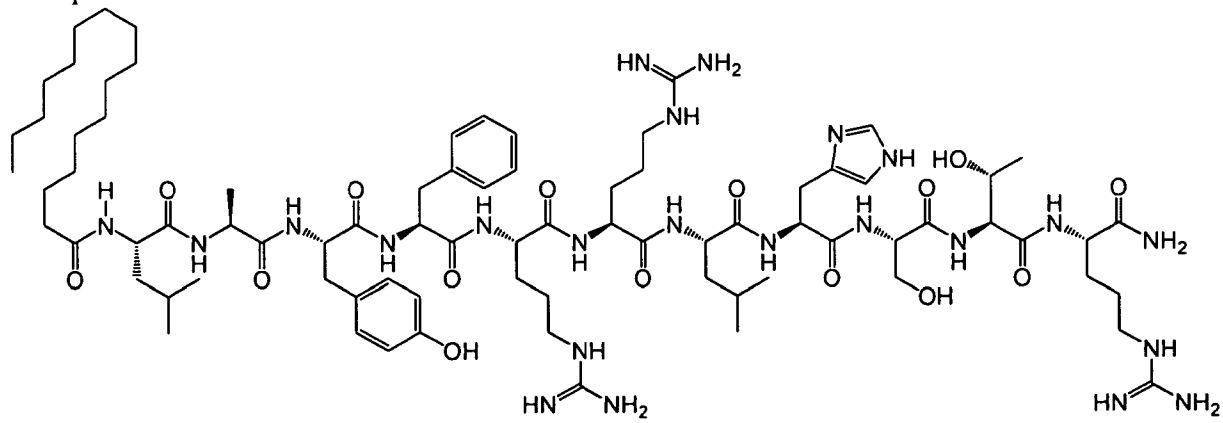
Compound 2



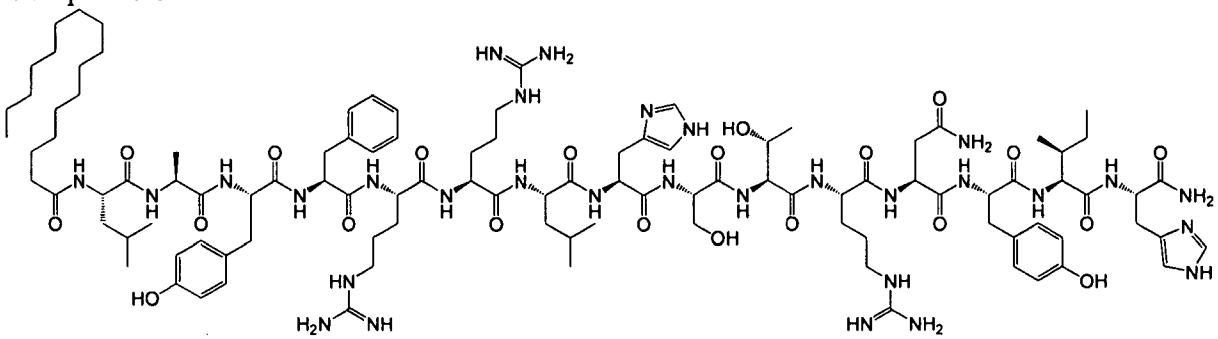
Compound 3



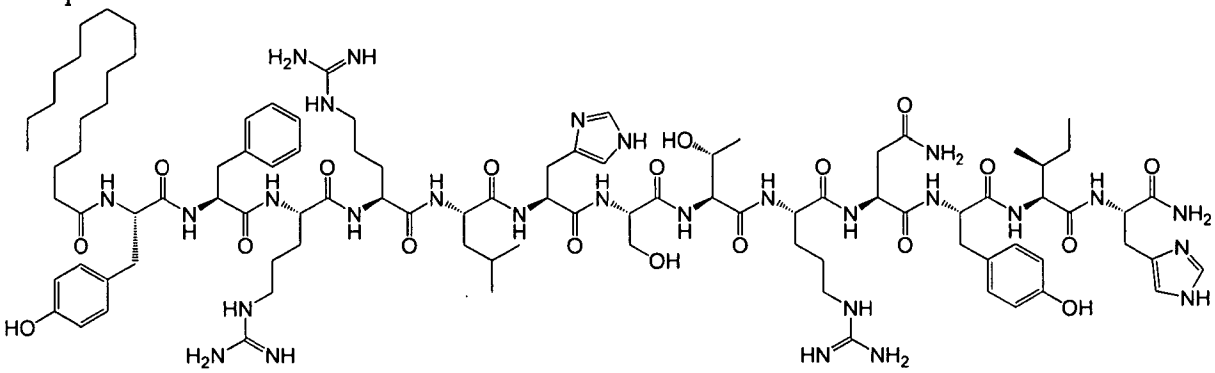
Compound 4



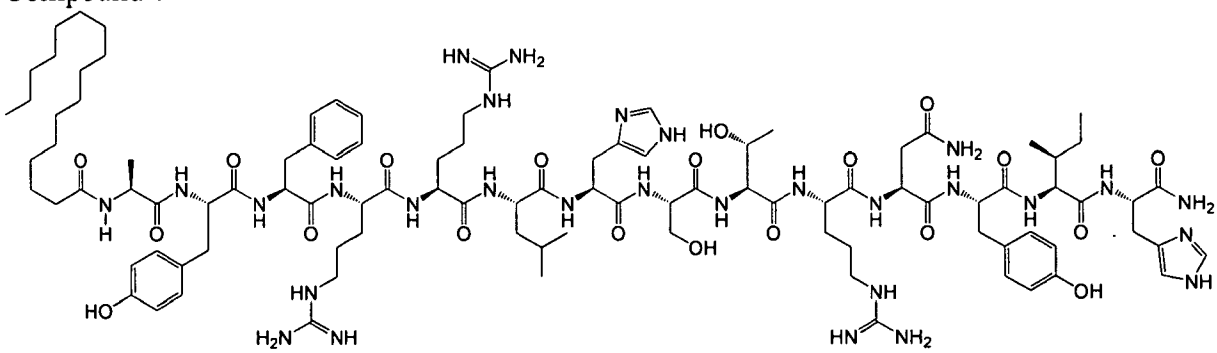
Compound 5



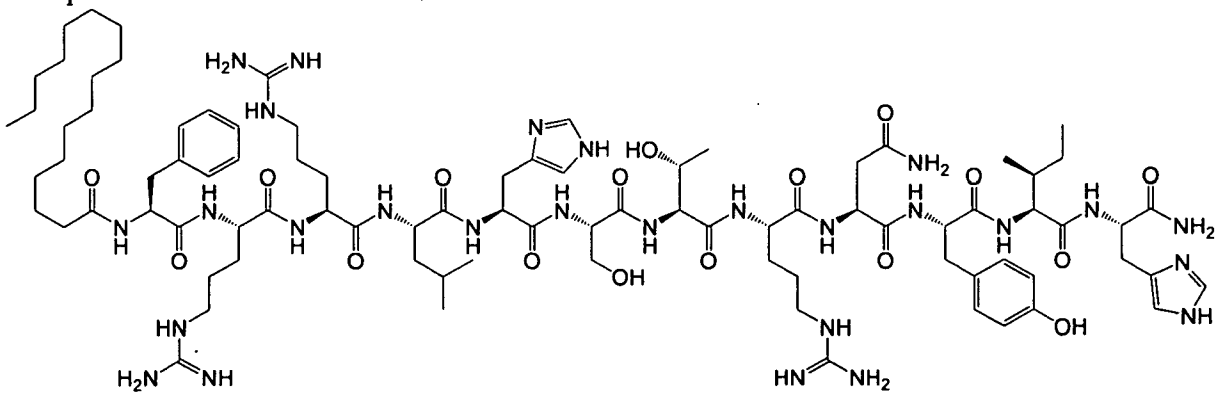
Compound 6



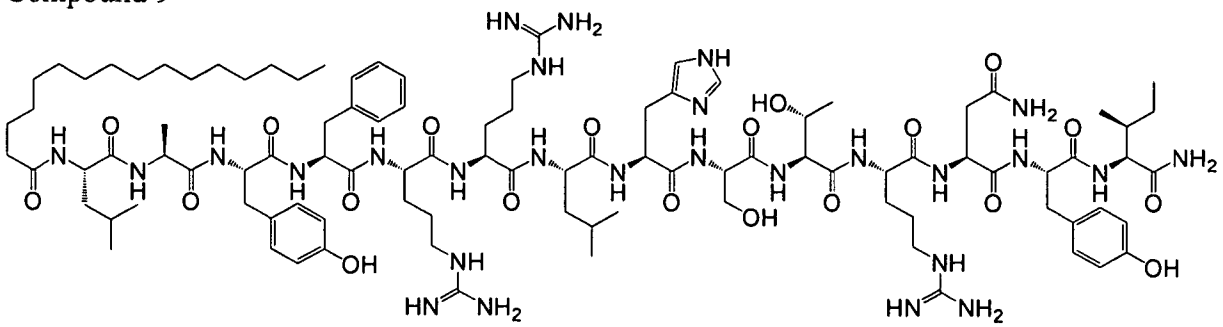
Compound 7



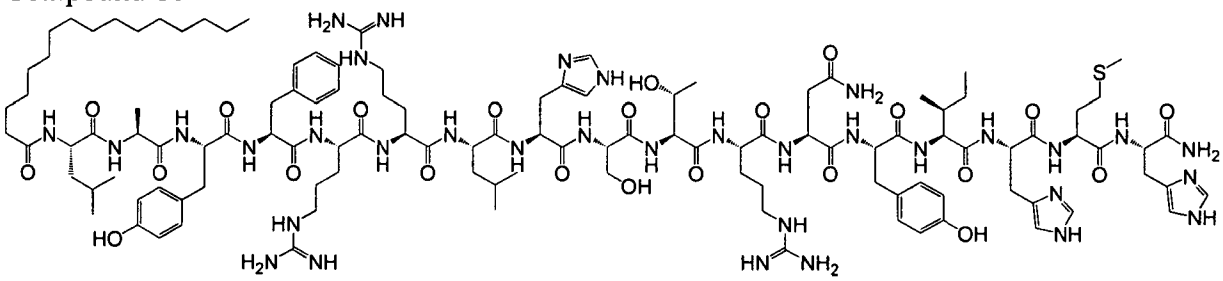
Compound 8



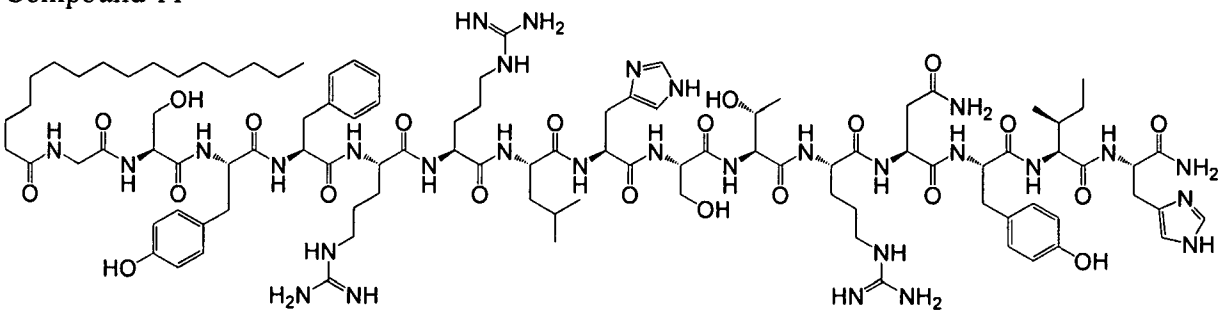
Compound 9



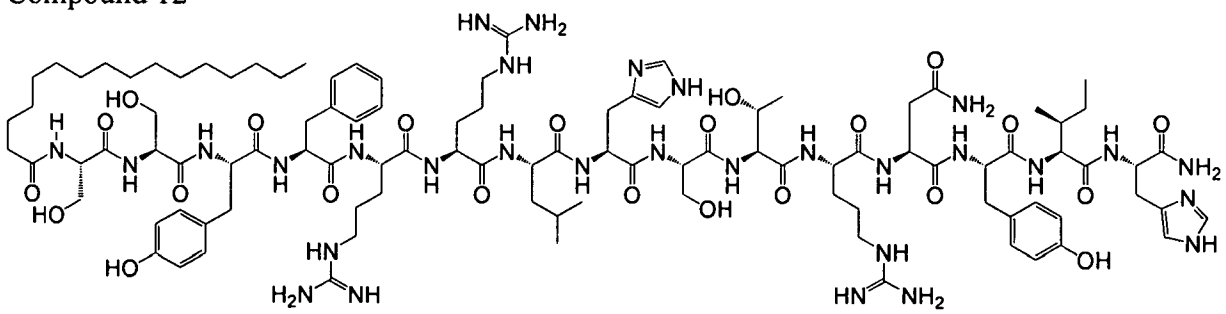
Compound 10



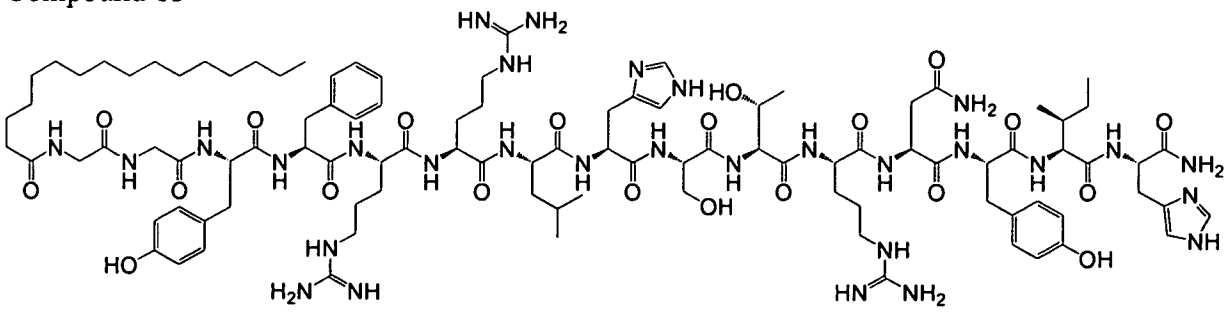
Compound 11



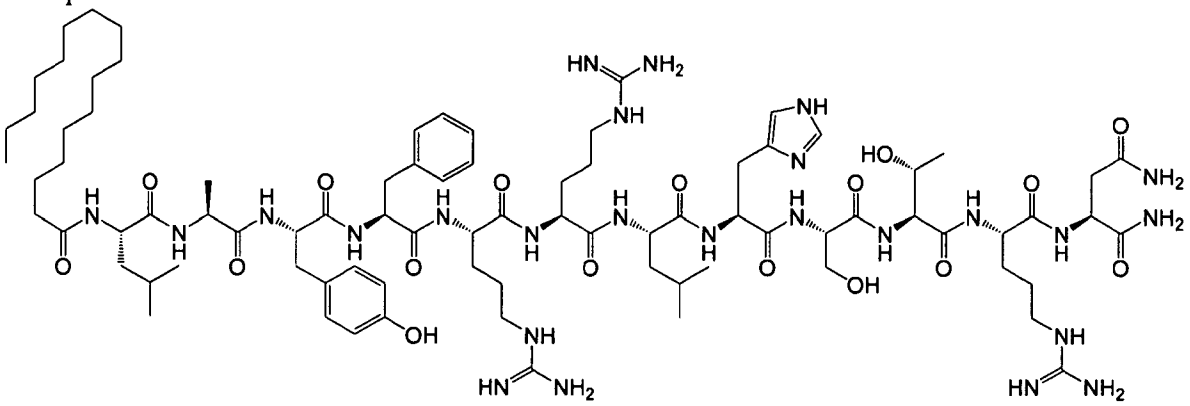
Compound 12



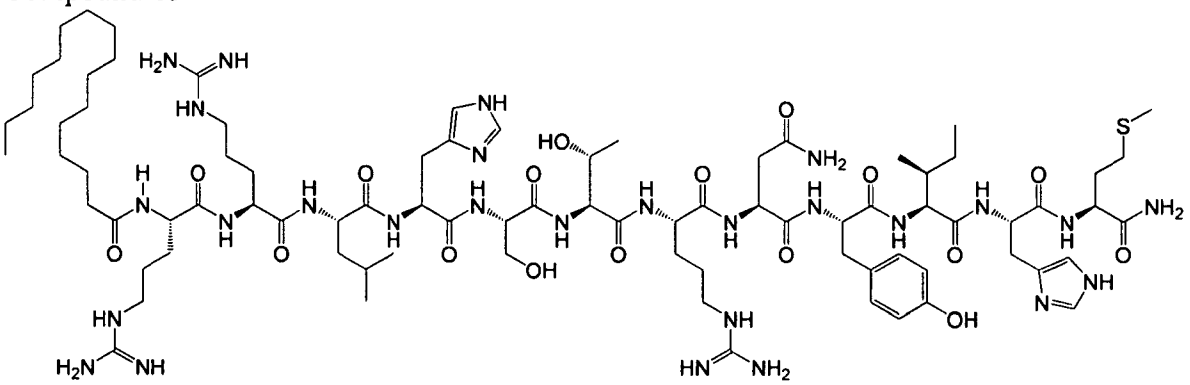
Compound 13



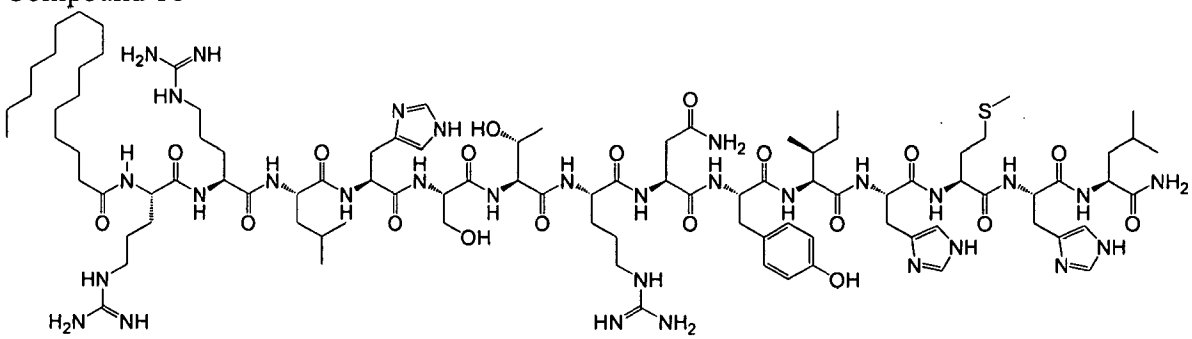
Compound 14



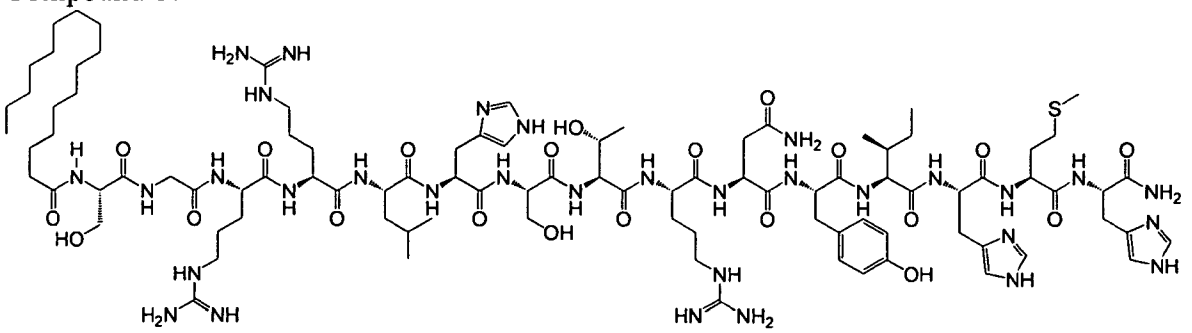
Compound 15



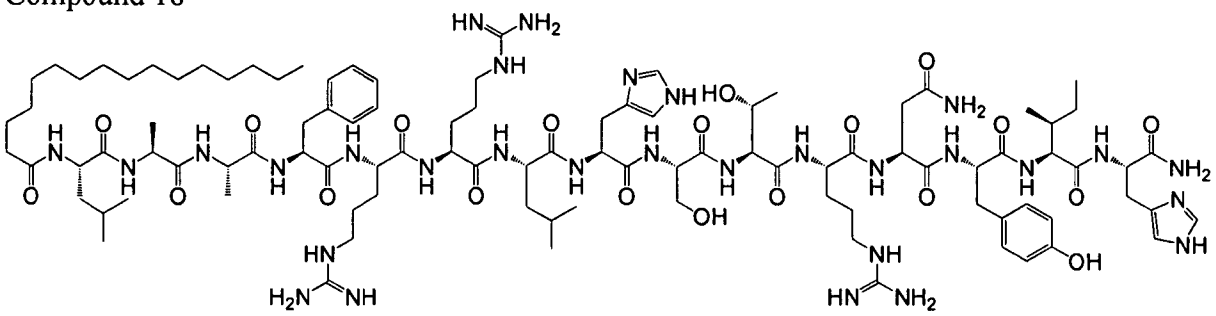
Compound 16



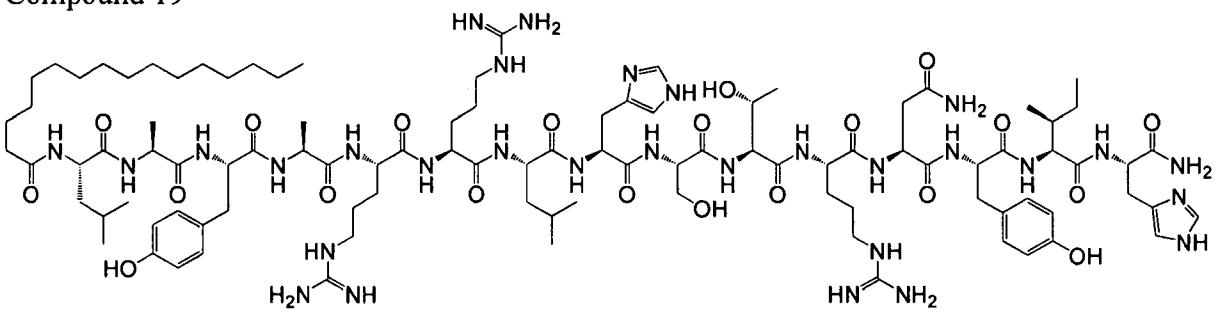
Compound 17



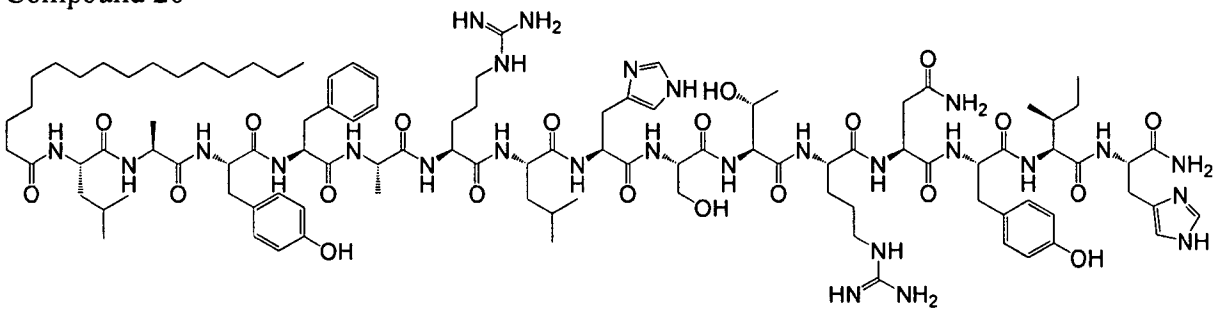
Compound 18



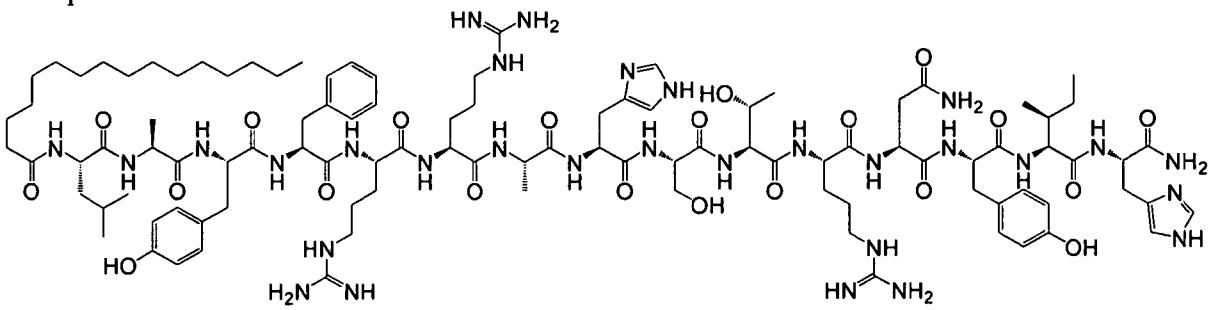
Compound 19



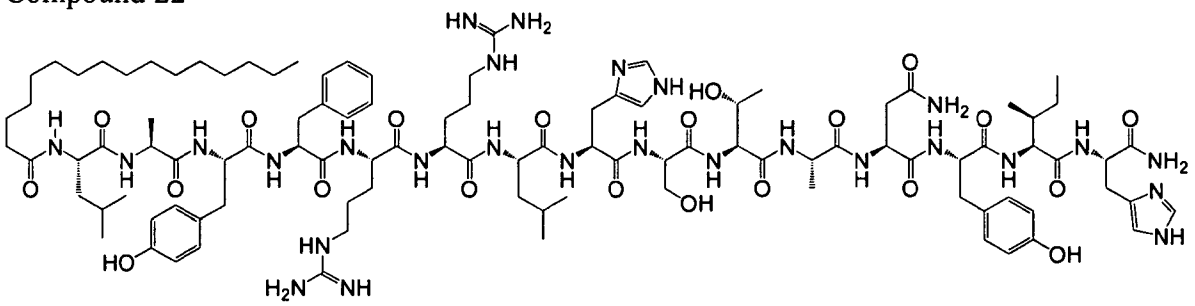
Compound 20



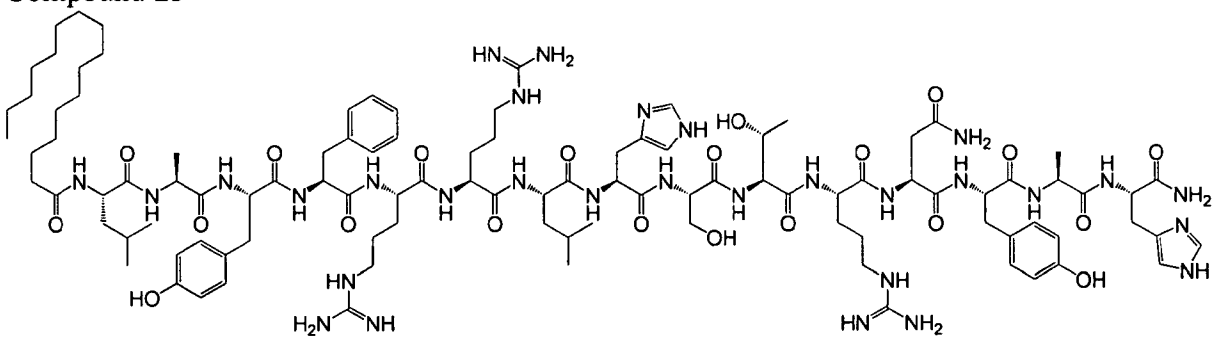
Compound 21



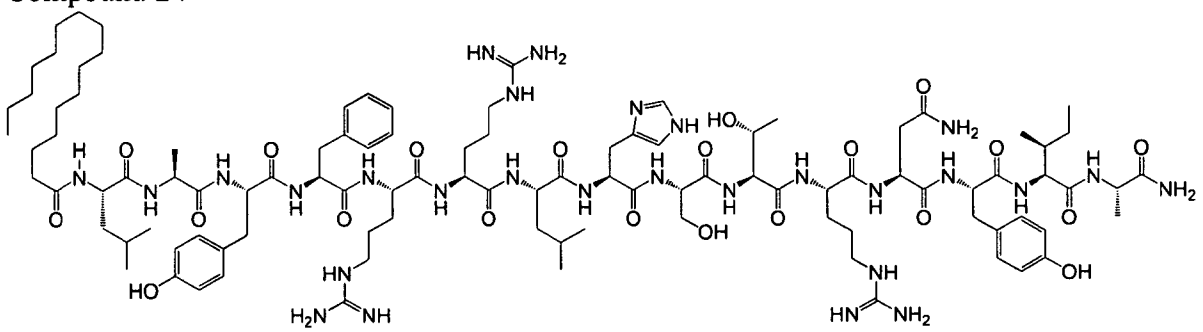
Compound 22



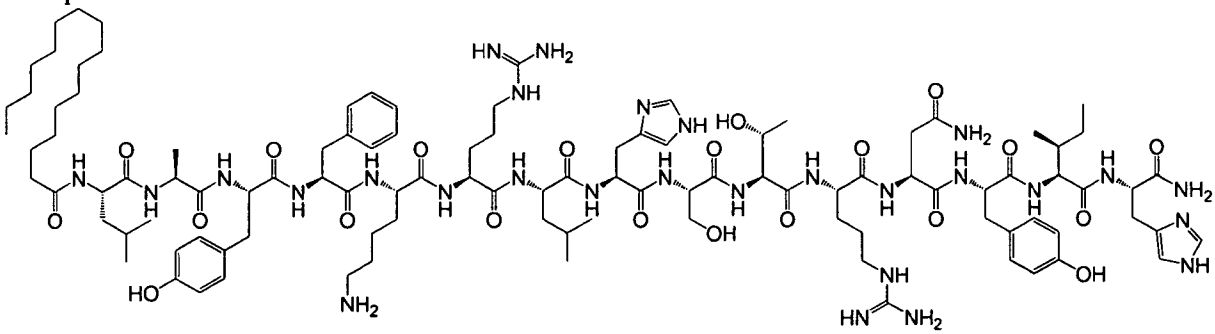
Compound 23



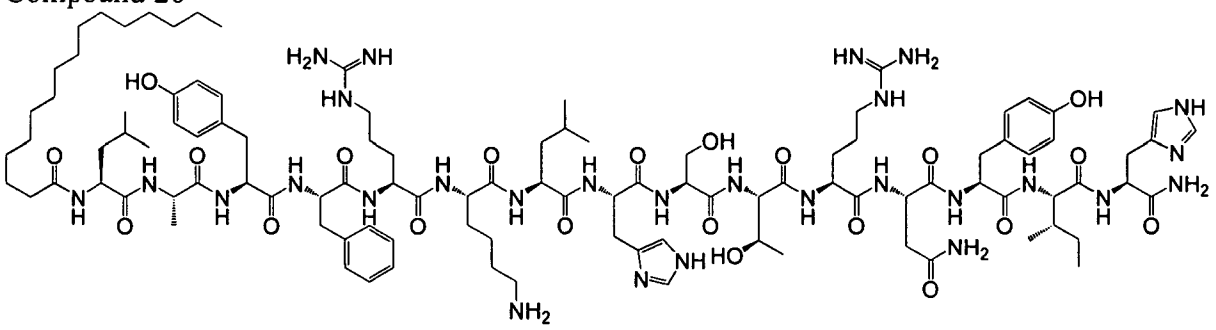
Compound 24



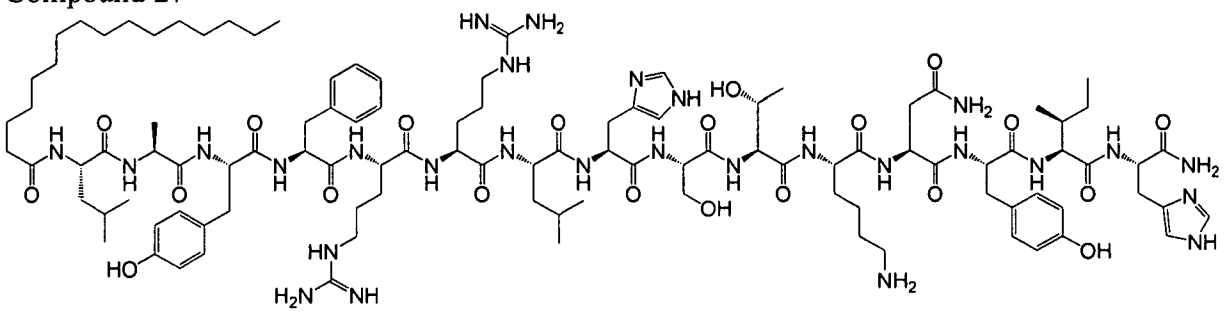
Compound 25



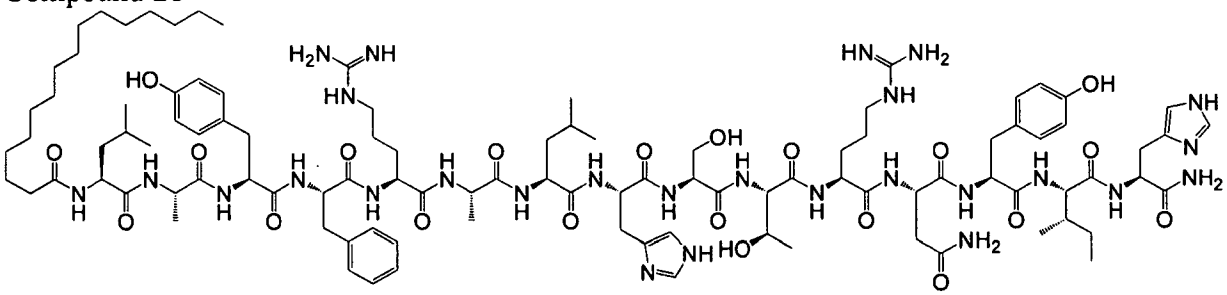
Compound 26



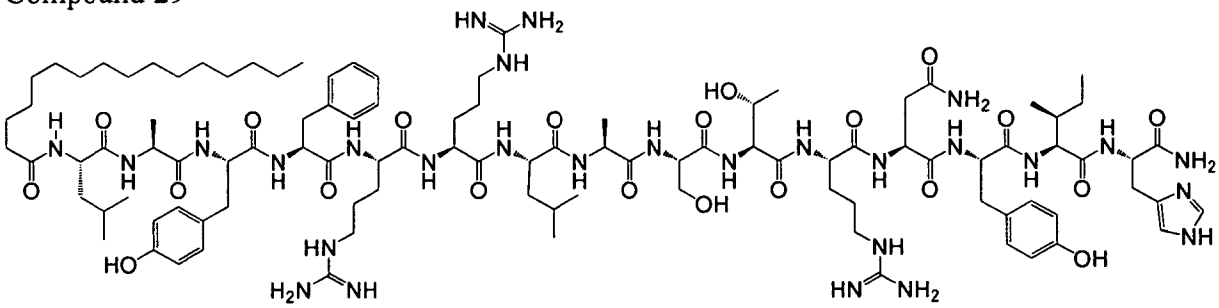
Compound 27



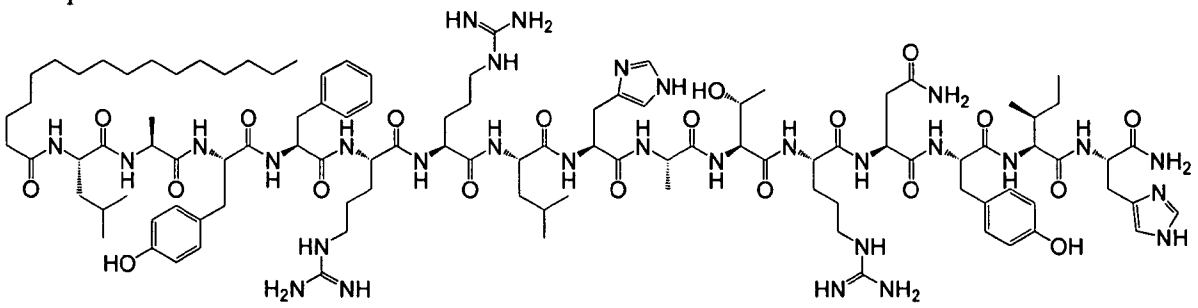
Compound 28



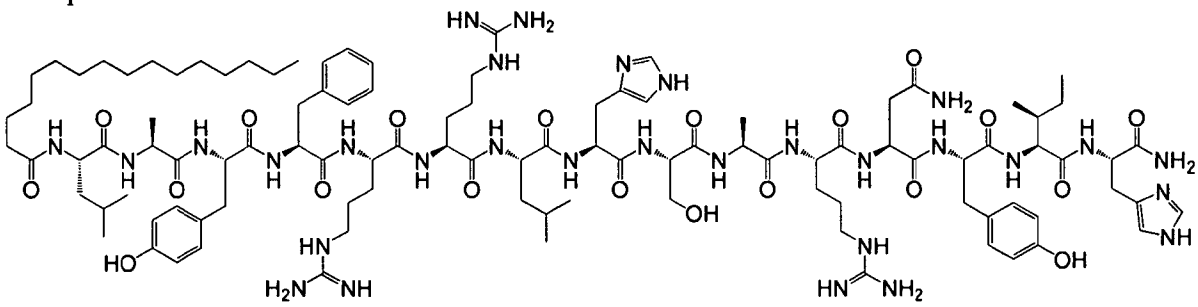
Compound 29



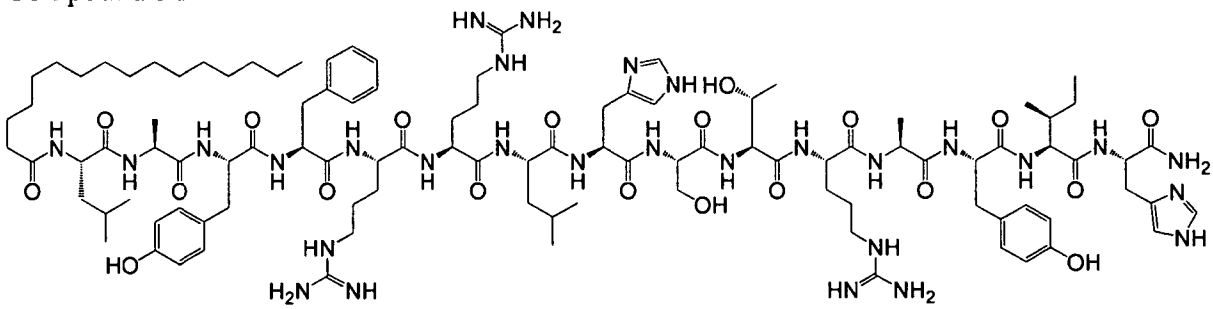
Compound 30



Compound 31

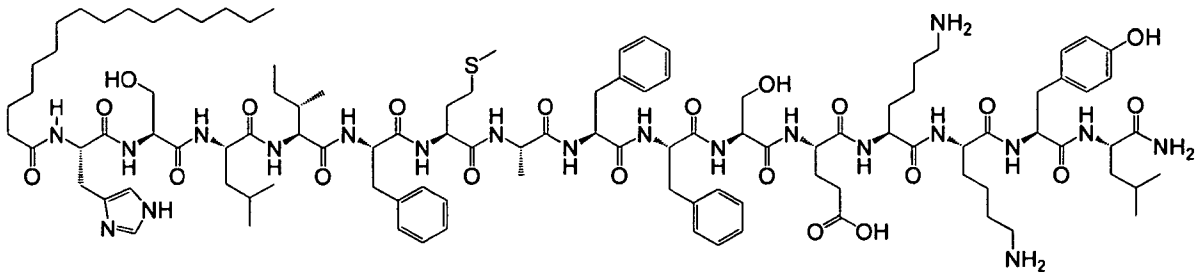


Compound 32

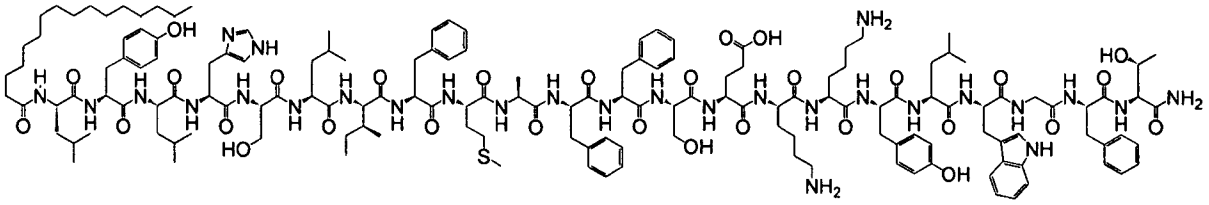


In yet another embodiment, a GPCR compound of the invention is selected from one of the following compounds or a pharmaceutically acceptable salt thereof:

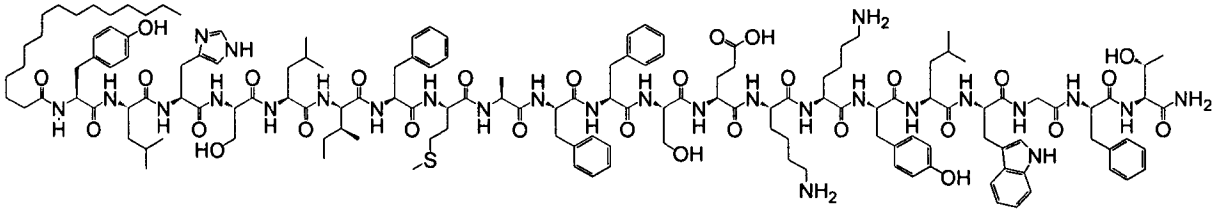
Compound 33



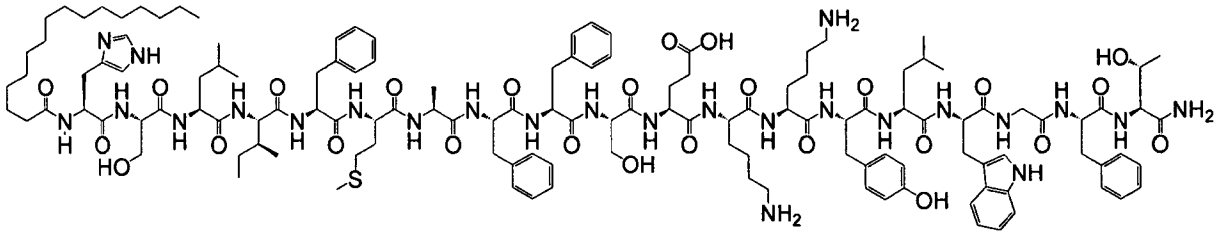
Compound 34



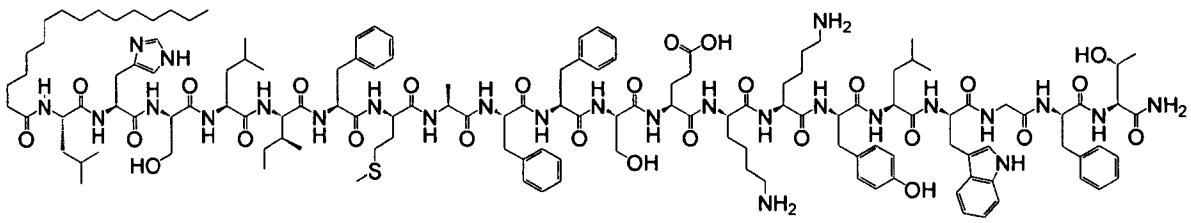
Compound 35



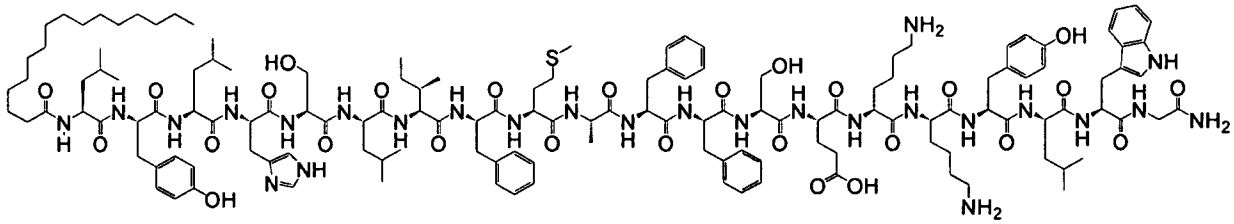
Compound 36



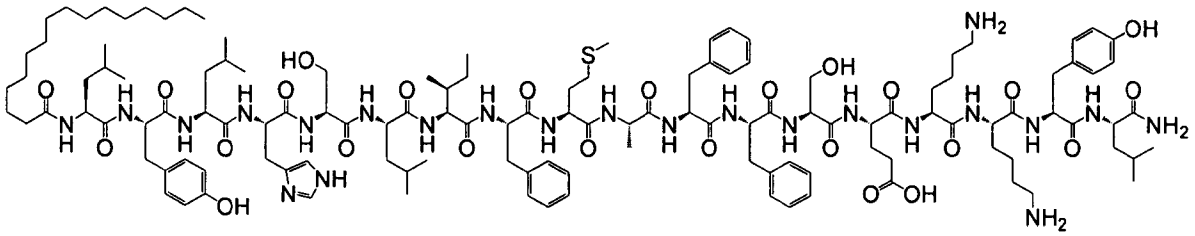
Compound 37



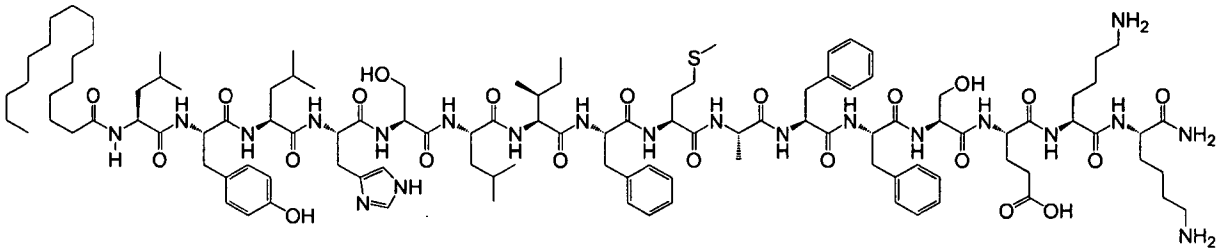
Compound 38



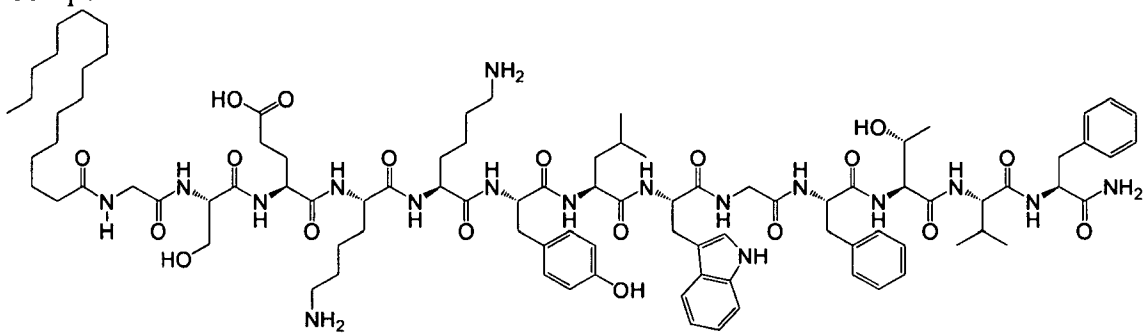
Compound 39



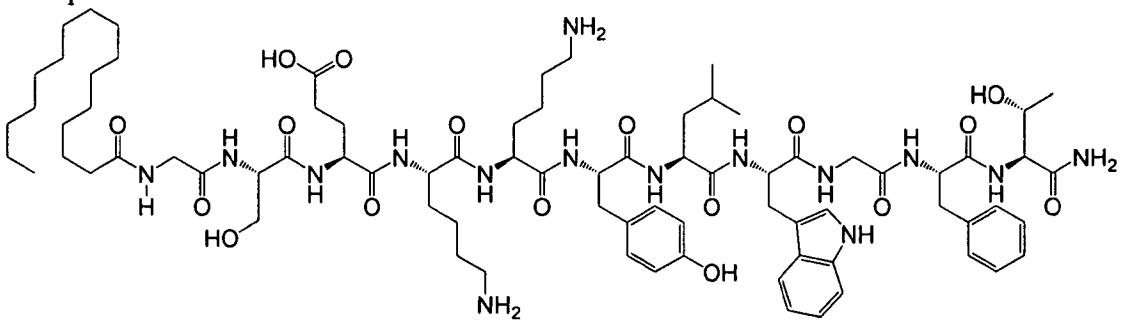
Compound 40



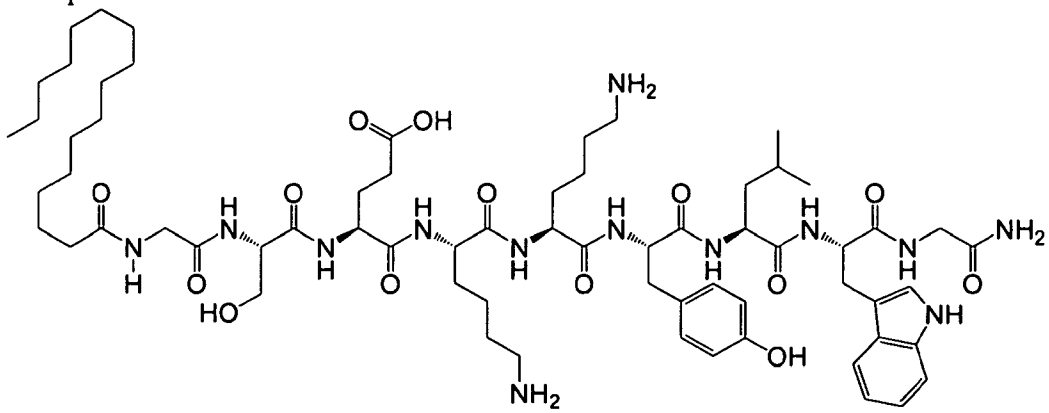
Compound 41



Compound 42

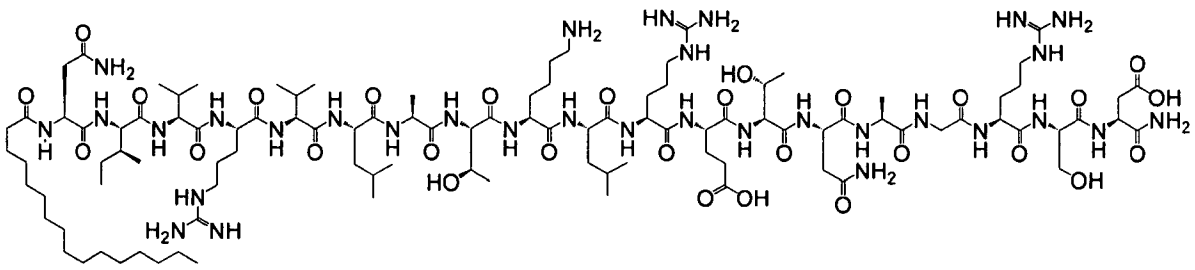


Compound 43

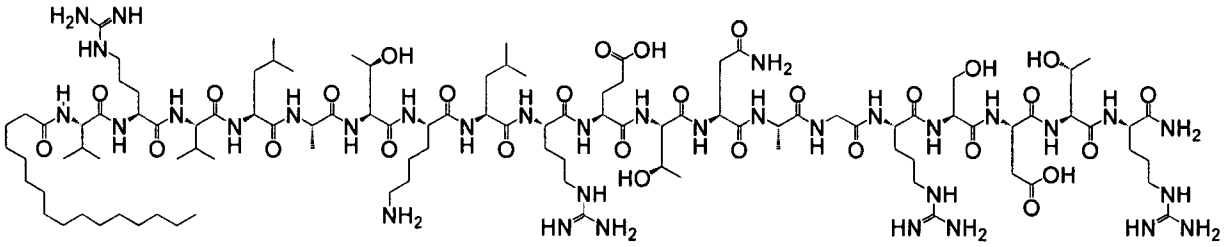


In yet another embodiment, a GPCR compound of the invention is selected from one of the following compounds or a pharmaceutically acceptable salt thereof:

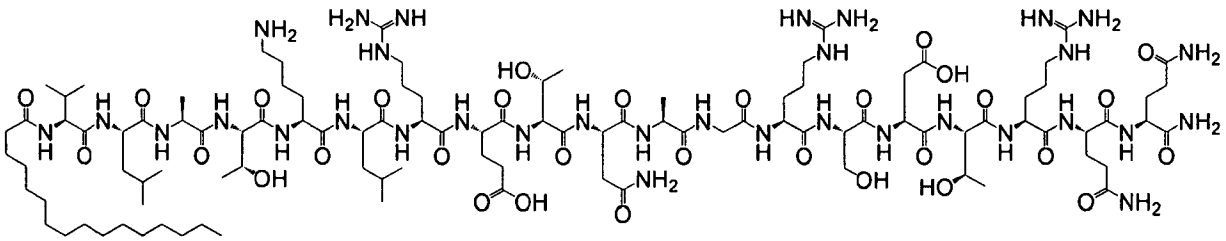
Compound 44



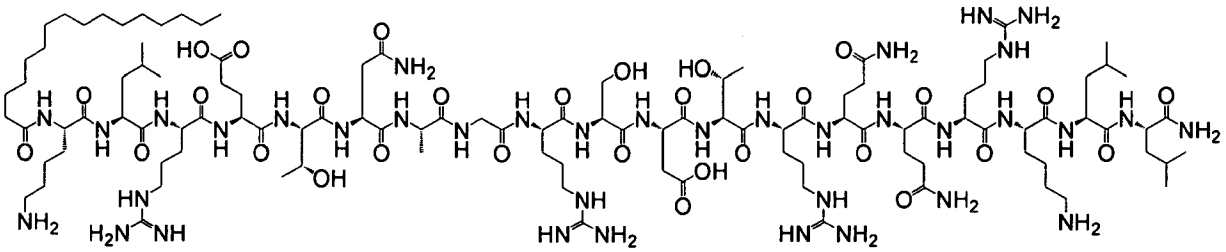
Compound 45



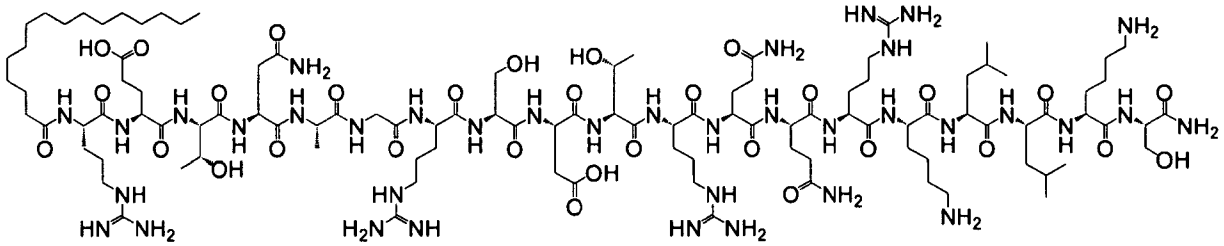
Compound 46



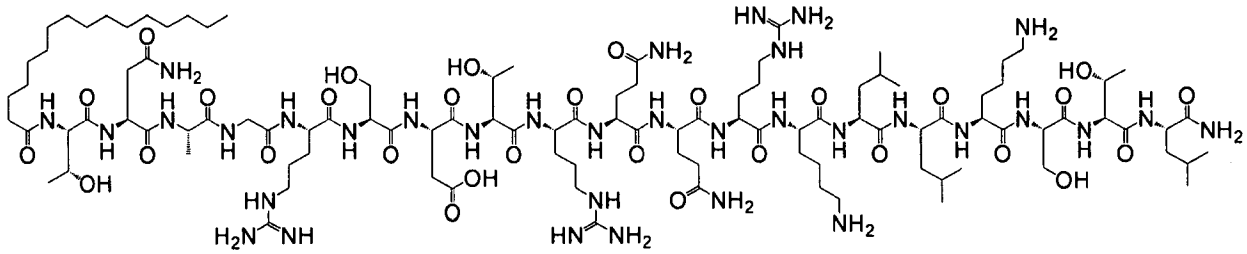
Compound 47



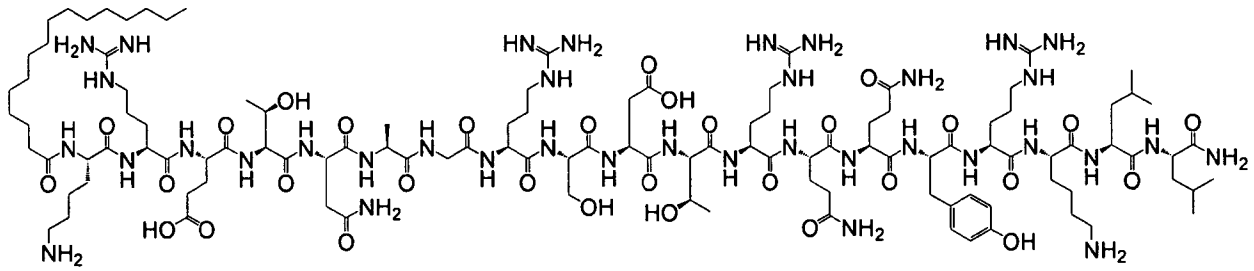
Compound 48



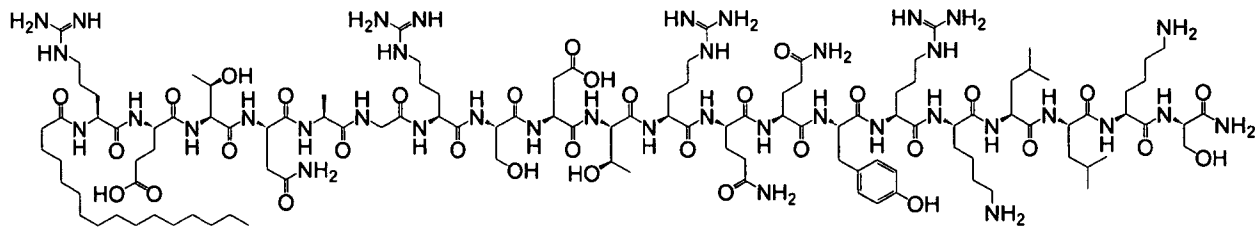
Compound 49



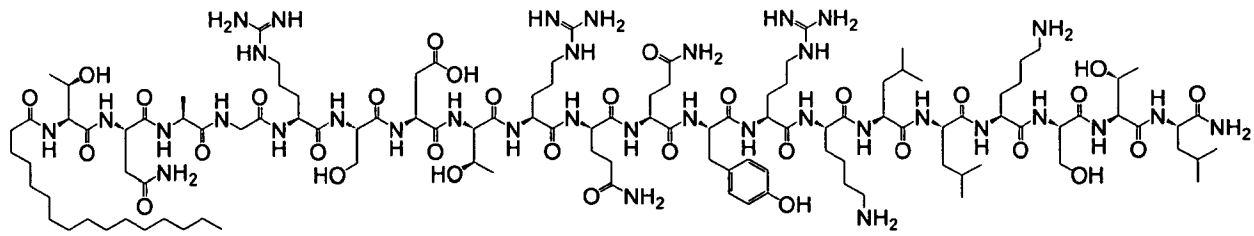
Compound 50



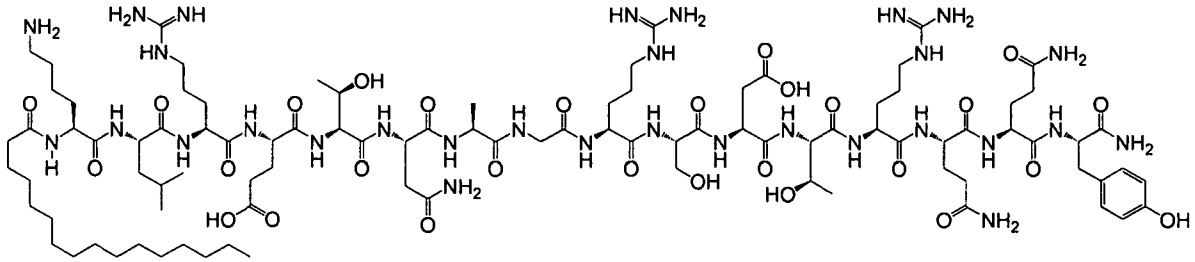
Compound 51



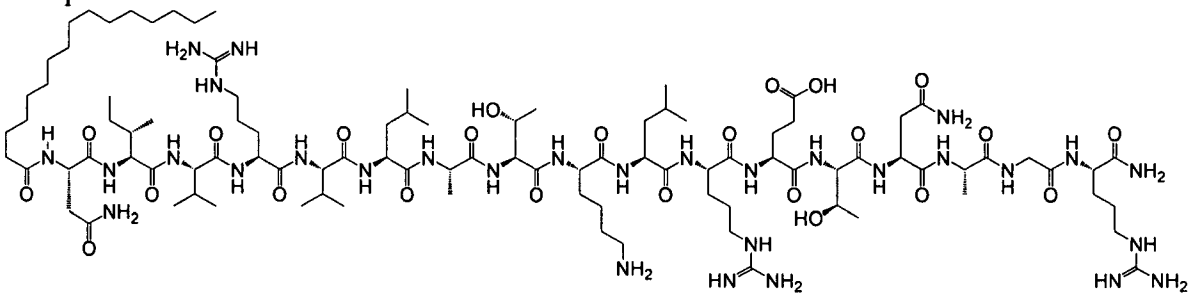
Compound 52



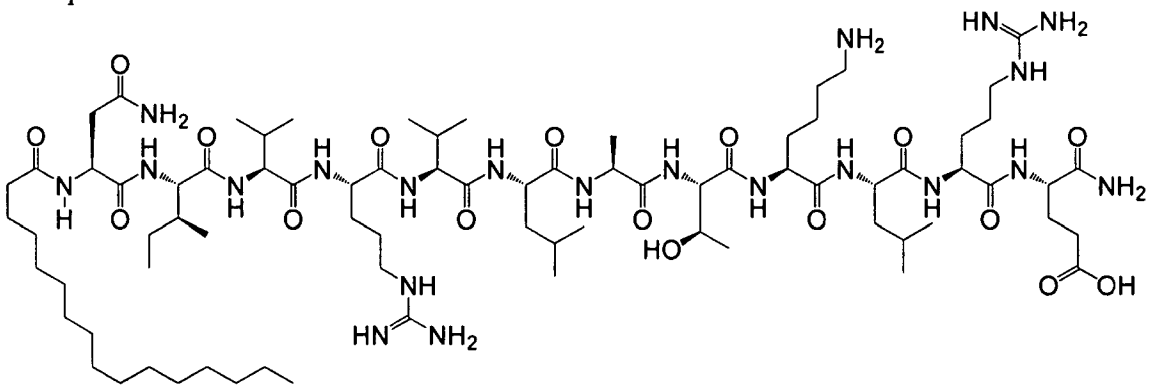
Compound 53



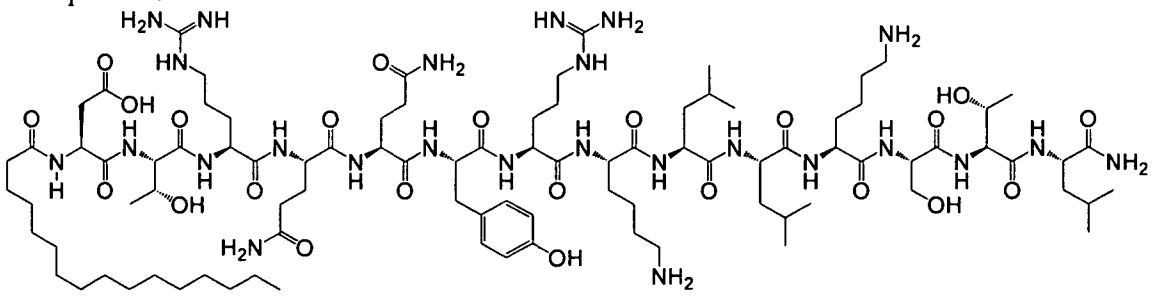
Compound 54



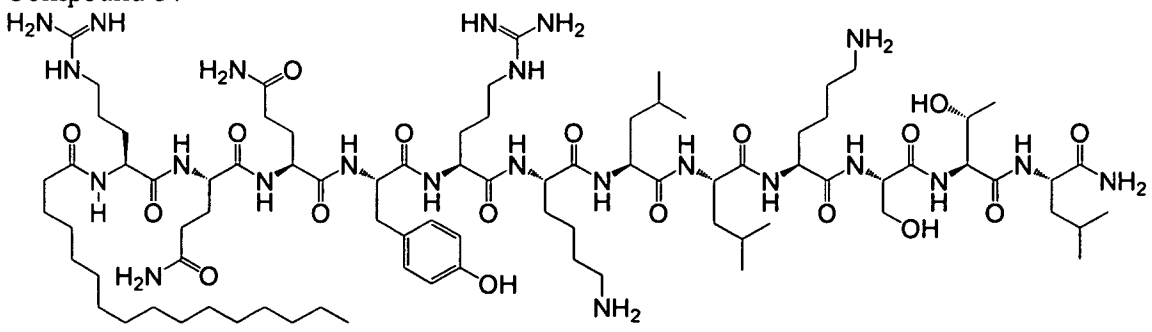
Compound 55



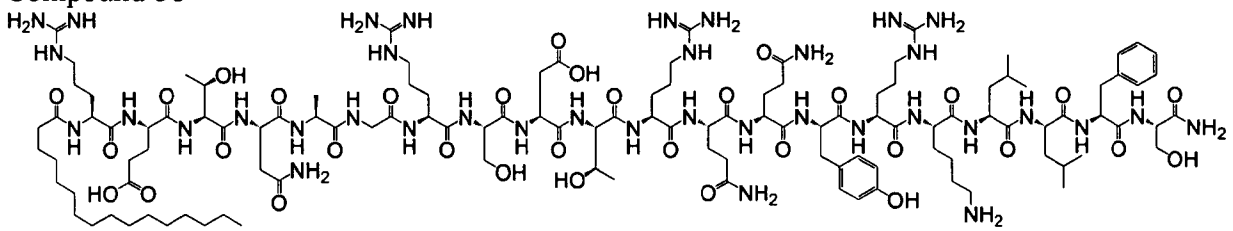
Compound 56



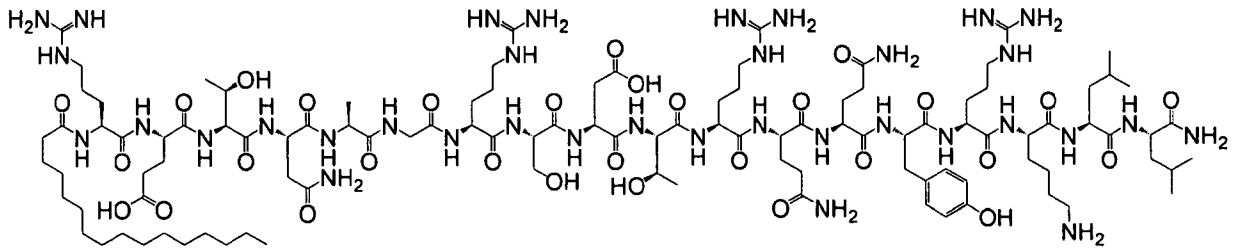
Compound 57



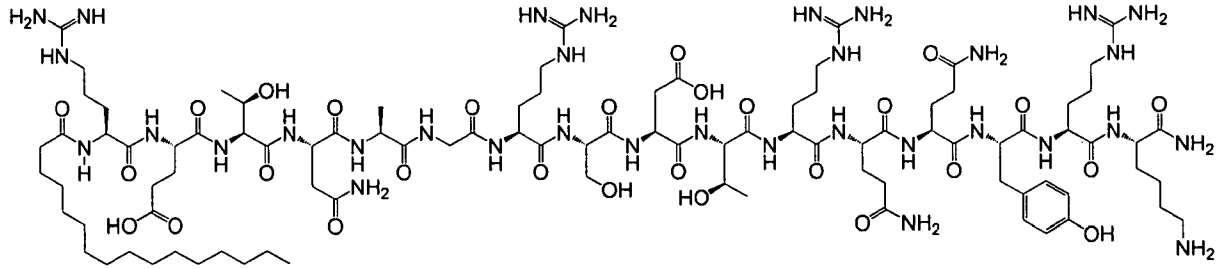
Compound 58



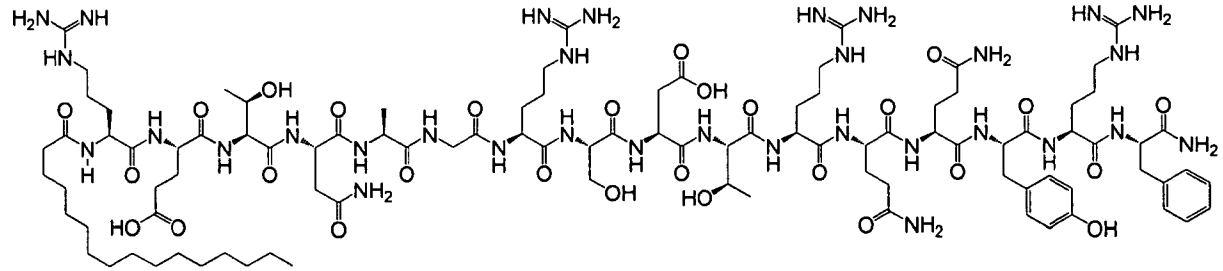
Compound 59



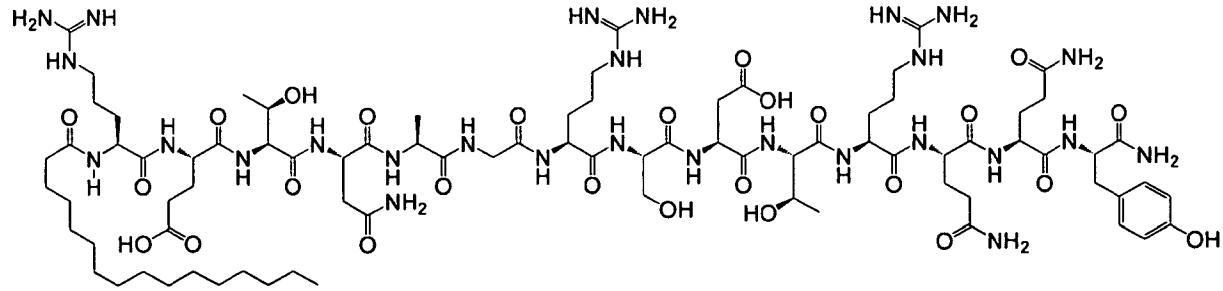
Compound 60



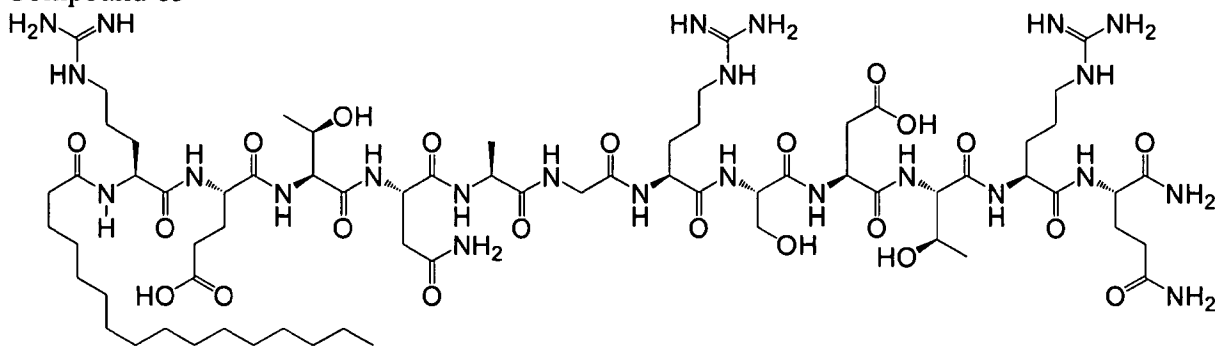
Compound 61



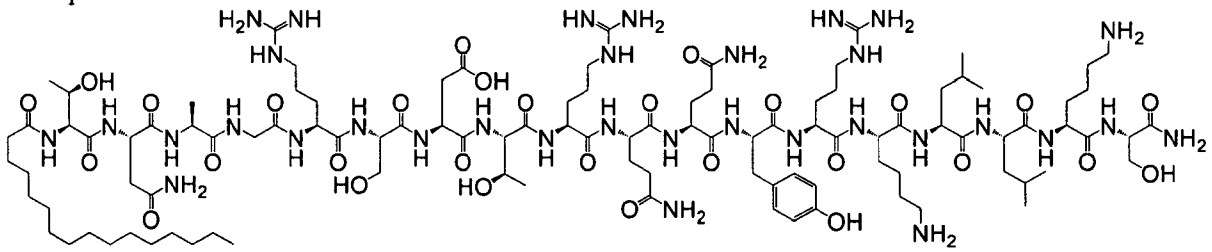
Compound 62



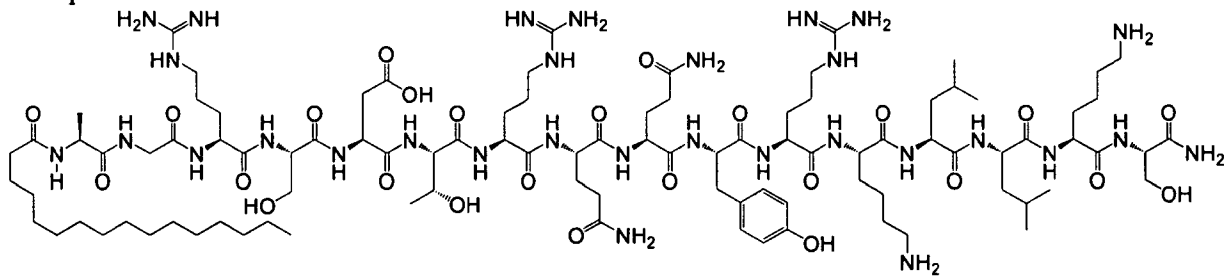
Compound 63



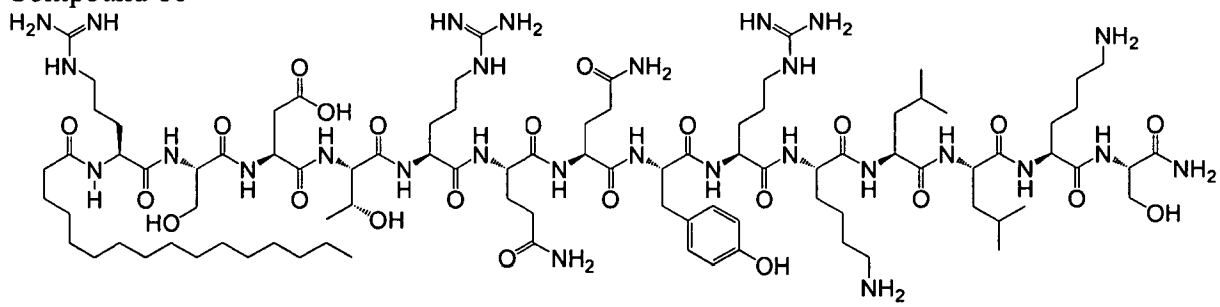
Compound 64



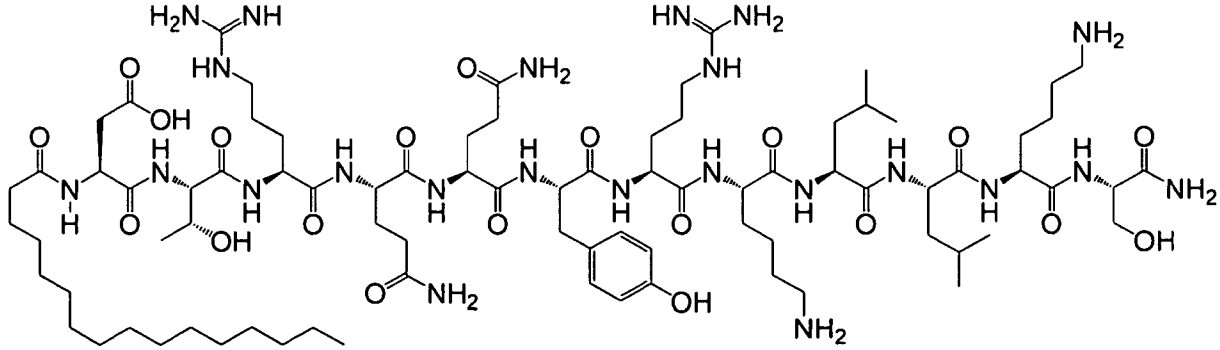
Compound 65



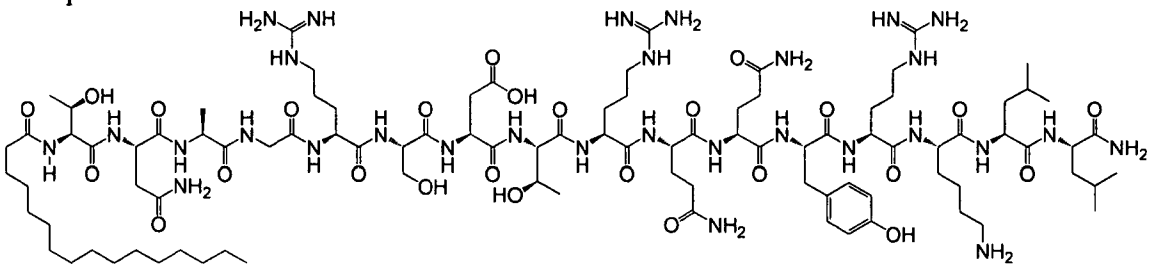
Compound 66



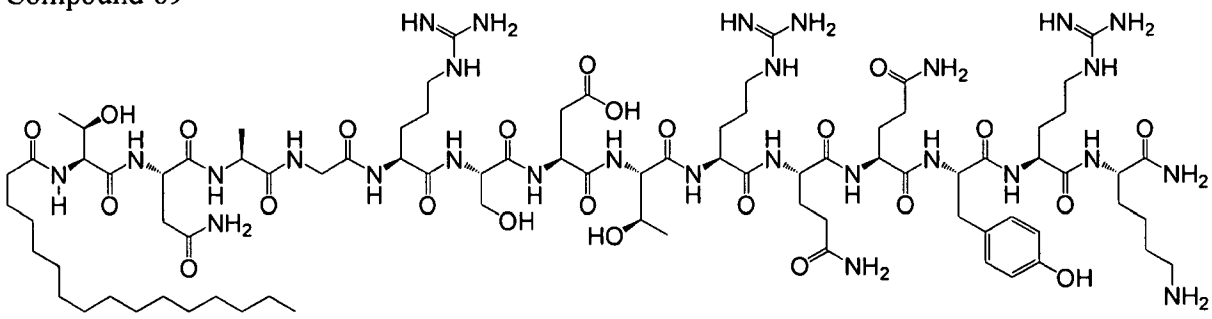
Compound 67



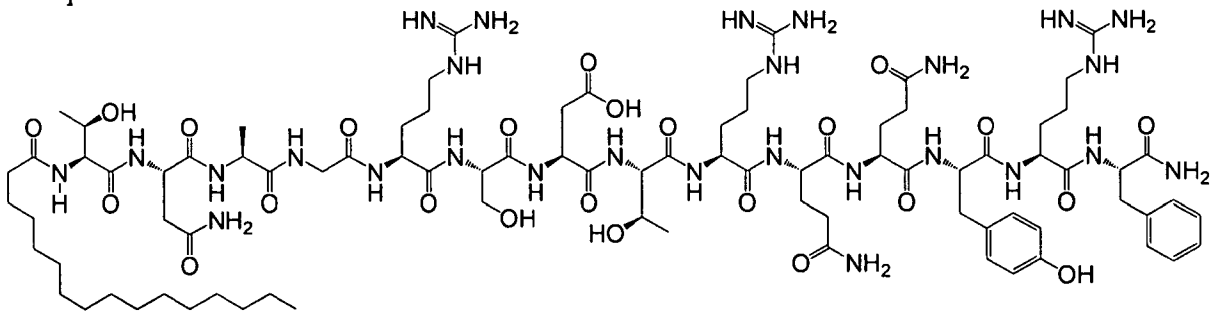
Compound 68



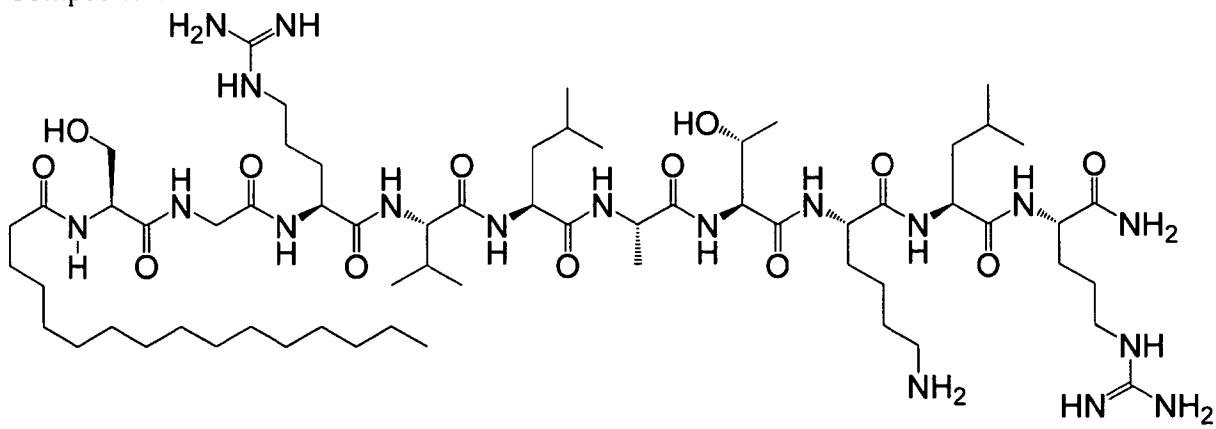
Compound 69



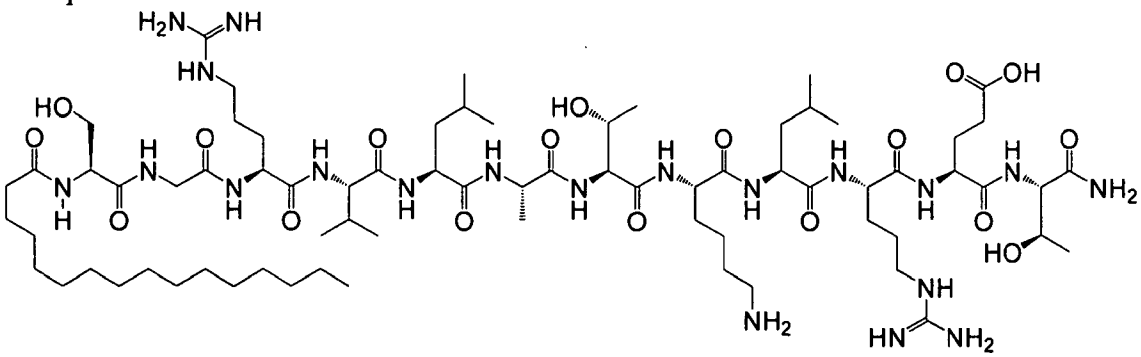
Compound 70



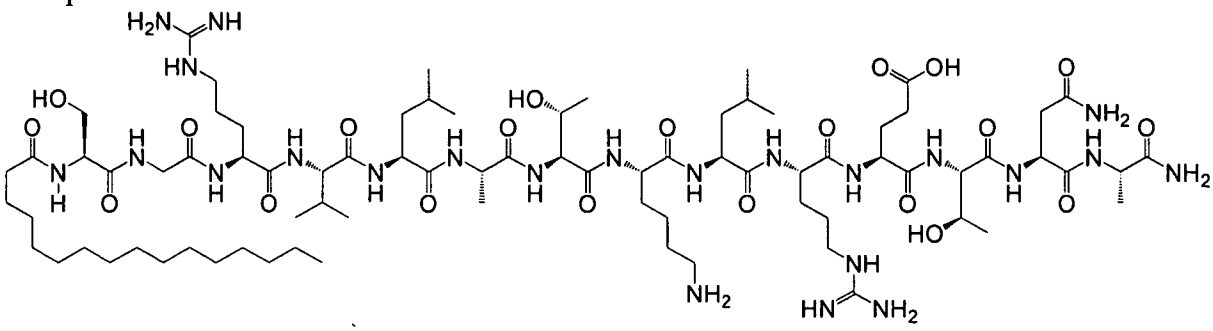
Compound 71



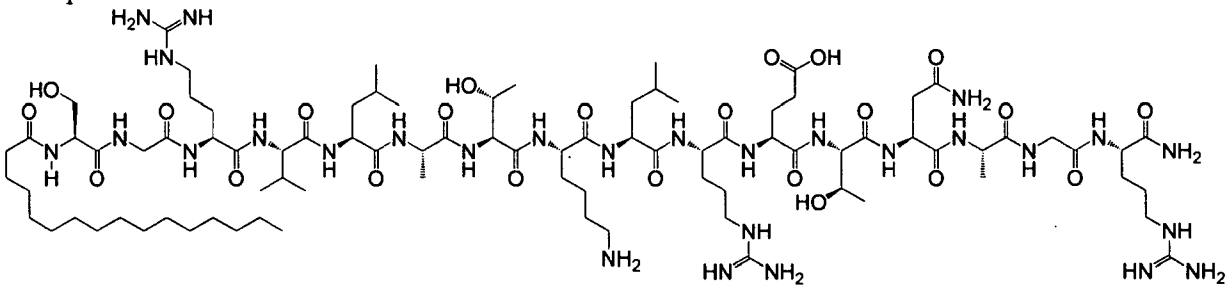
Compound 72



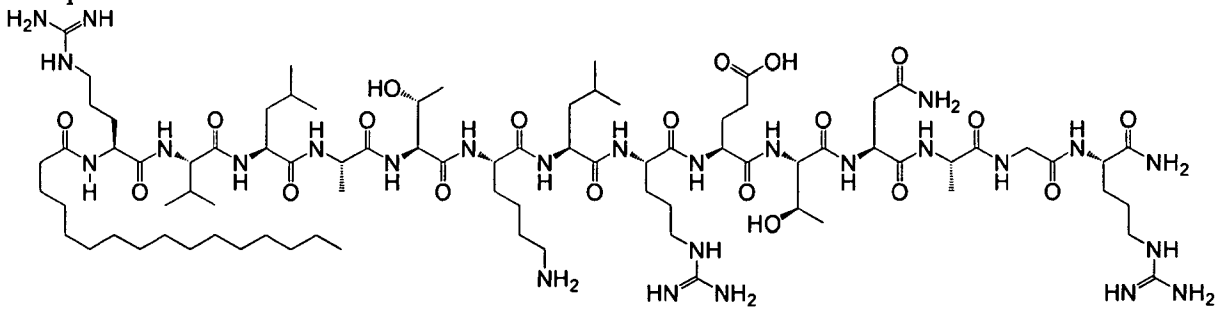
Compound 73



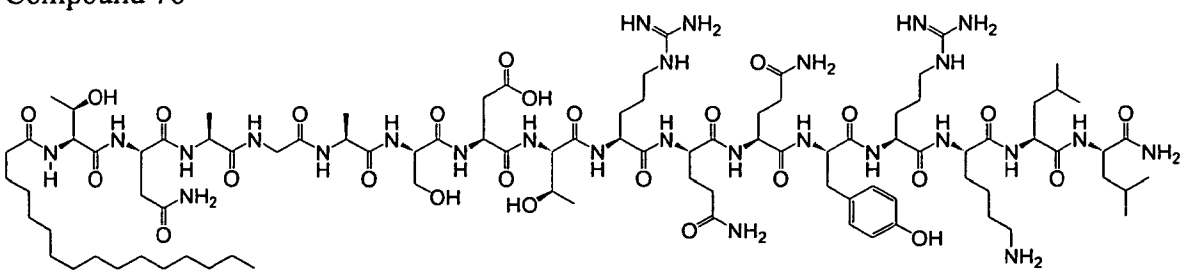
Compound 74



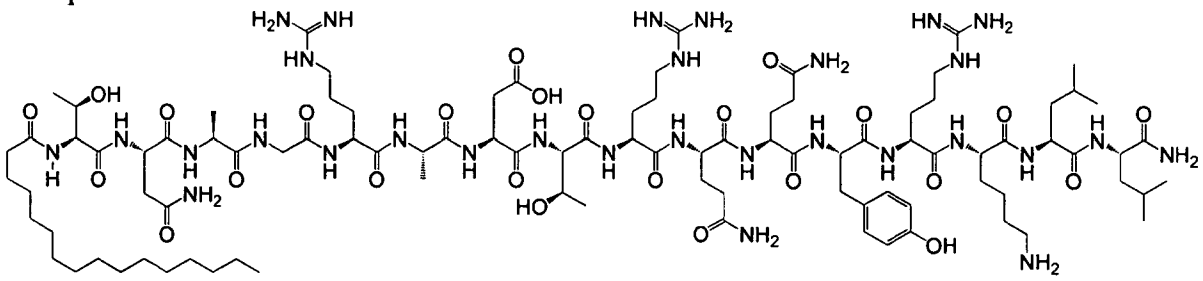
Compound 75



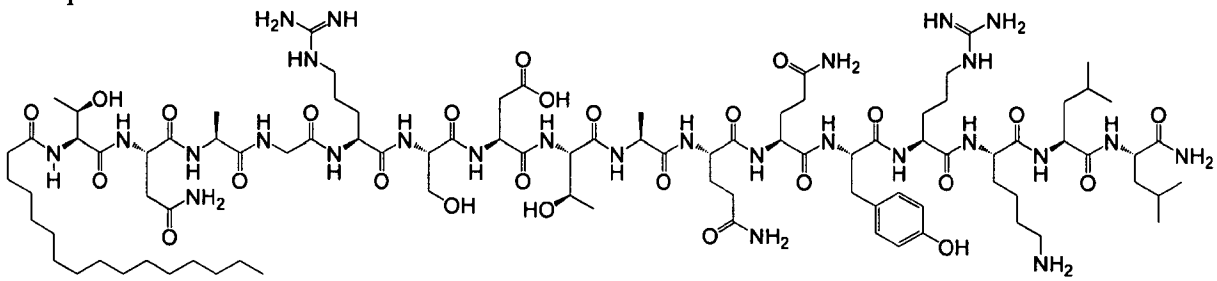
Compound 76



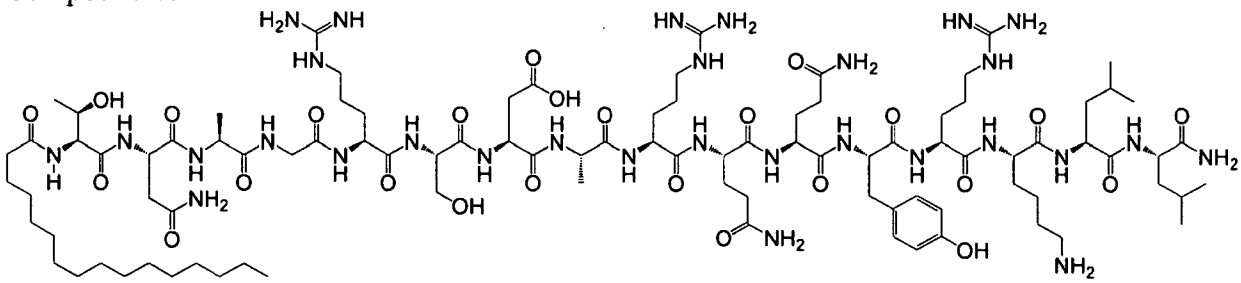
Compound 77



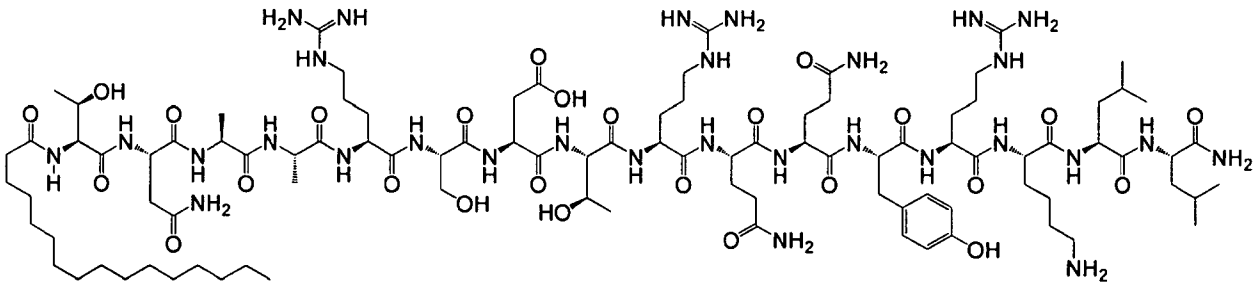
Compound 78



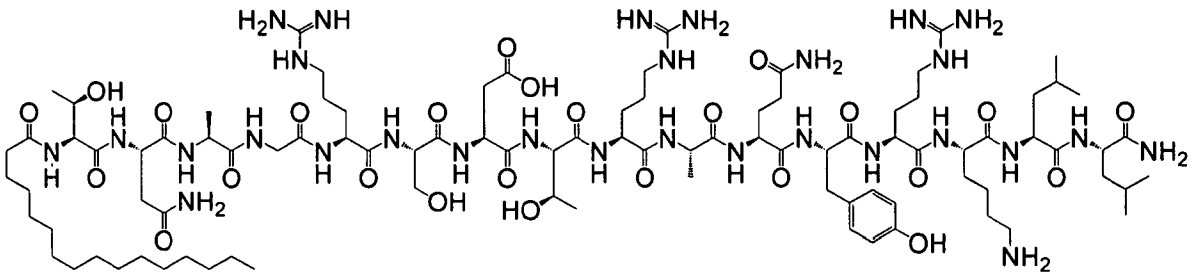
Compound 79



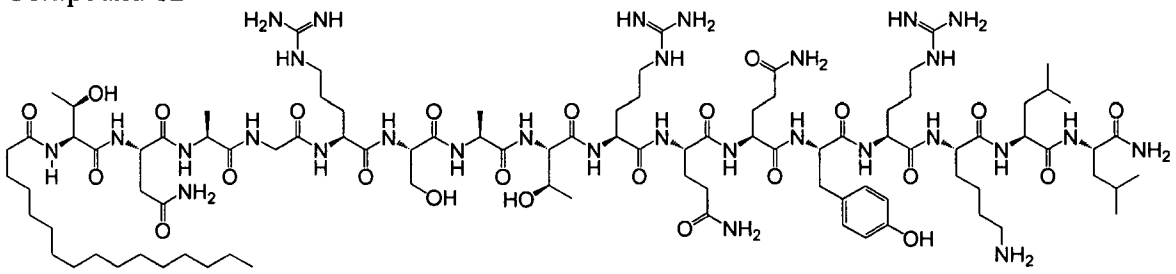
Compound 80



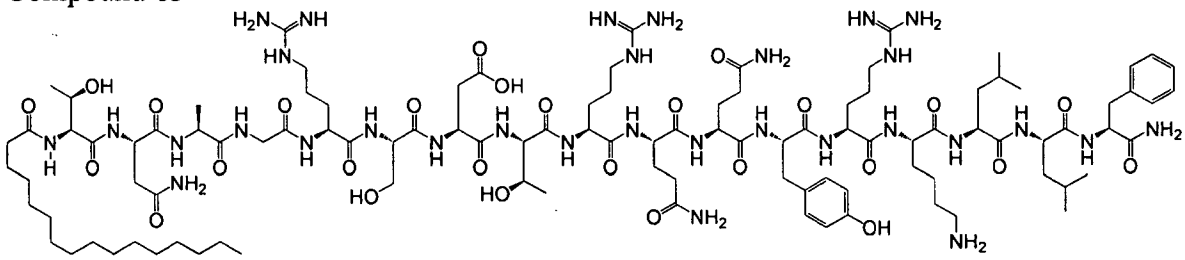
Compound 81



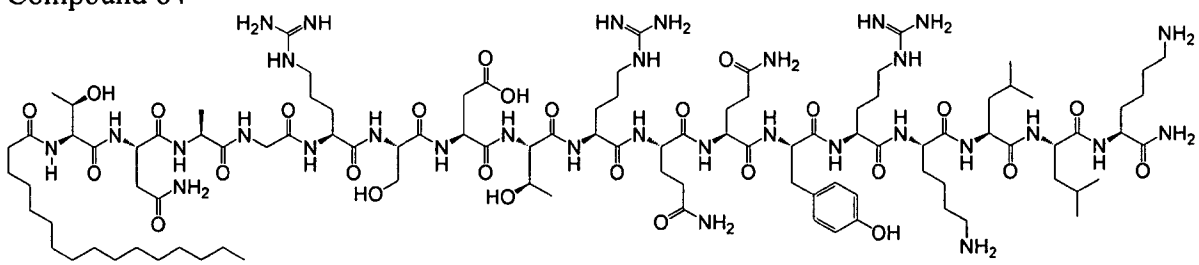
Compound 82



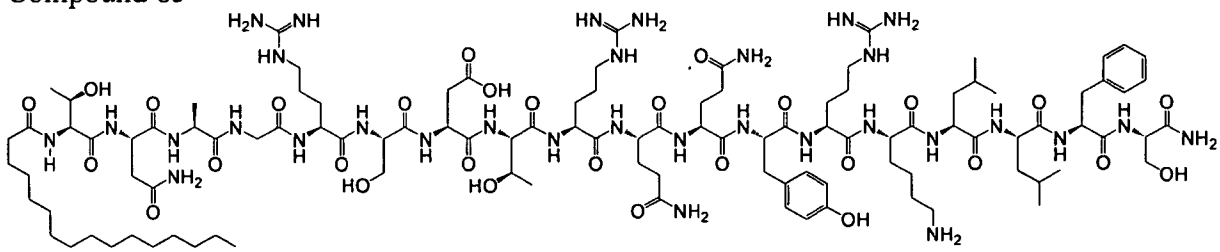
Compound 83



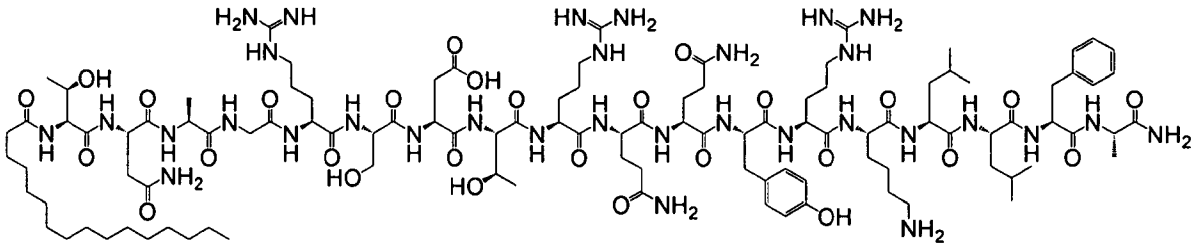
Compound 84



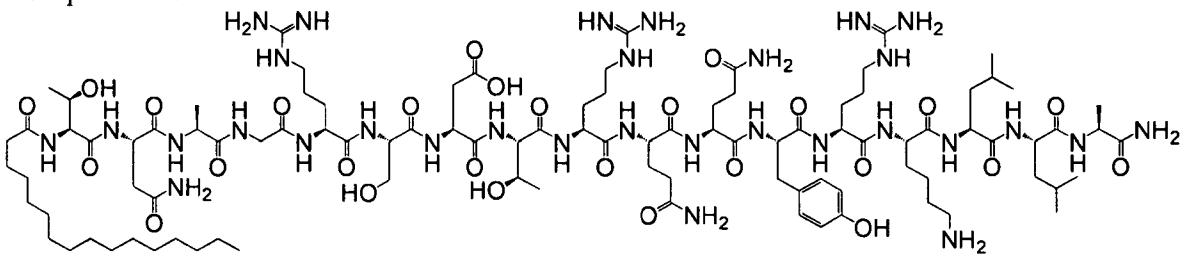
Compound 85



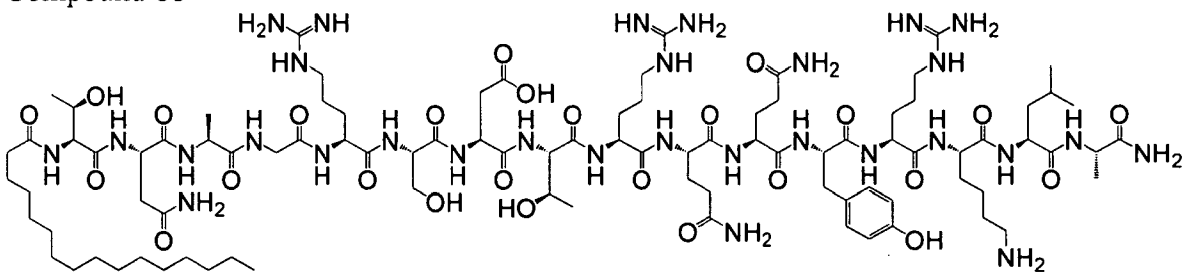
Compound 86



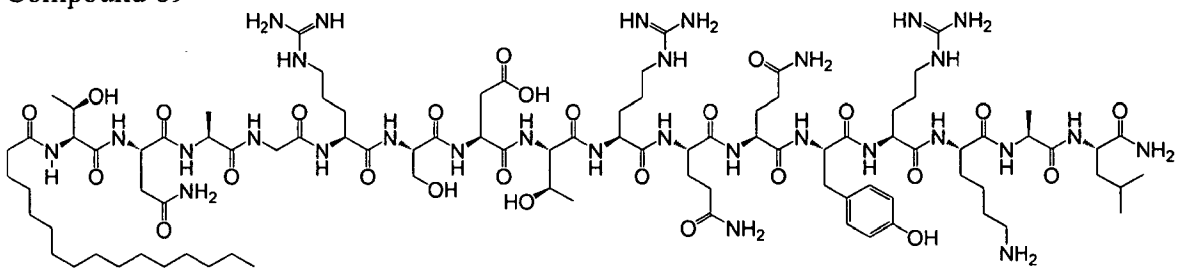
Compound 87



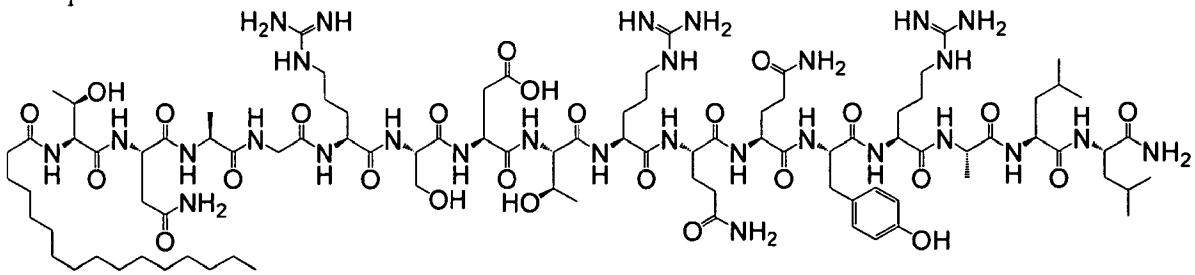
Compound 88



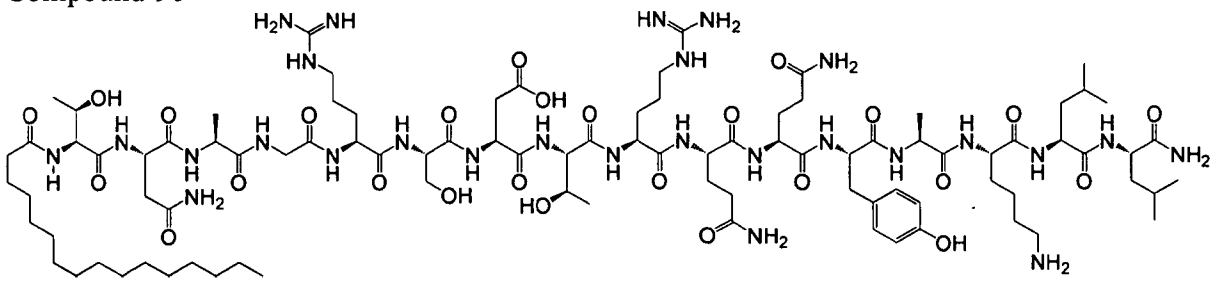
Compound 89



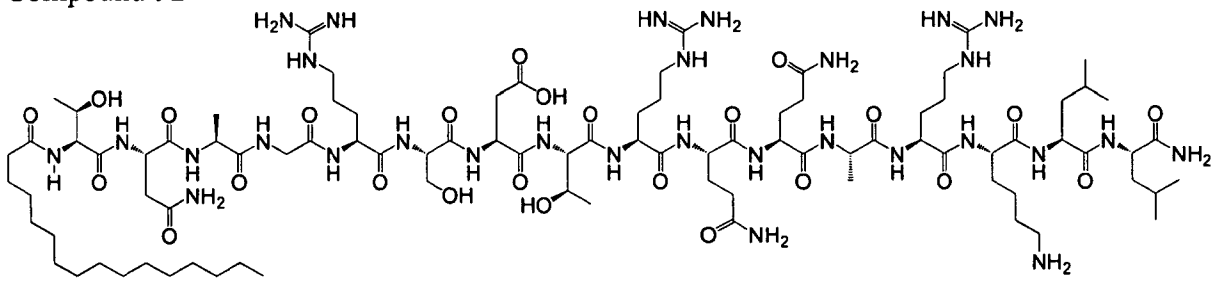
Compound 90



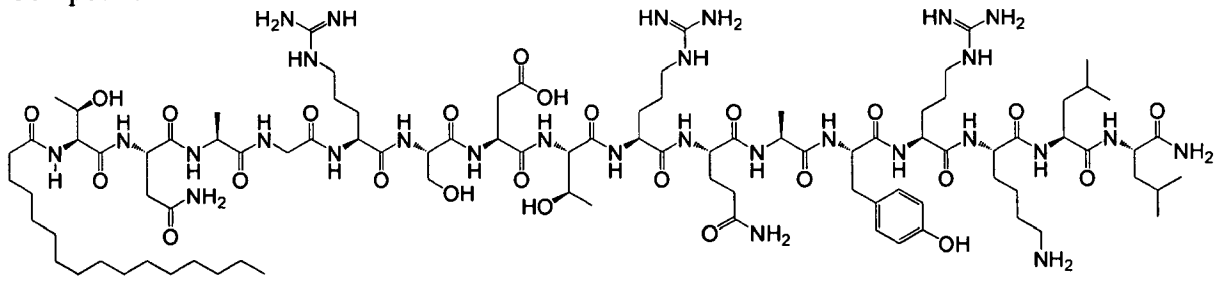
Compound 91



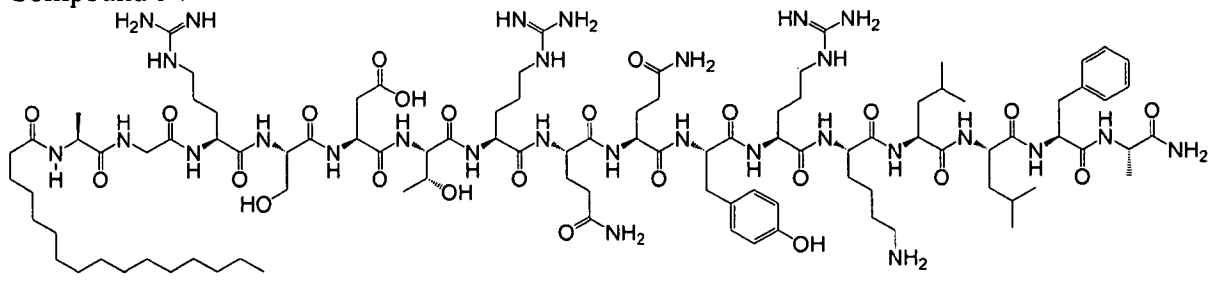
Compound 92



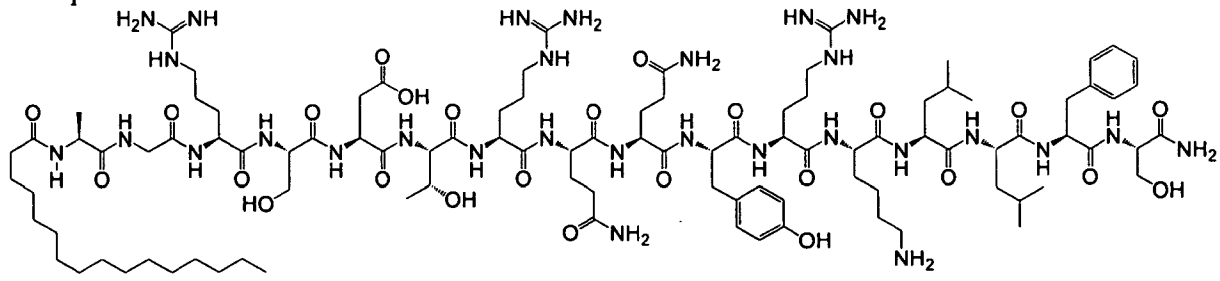
Compound 93



Compound 94

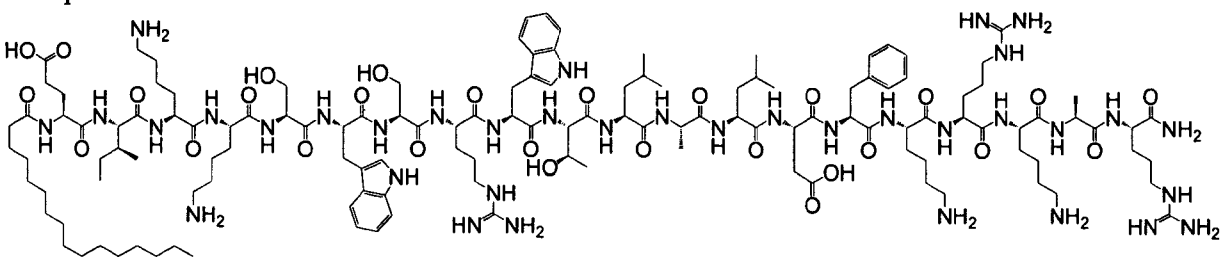


Compound 95

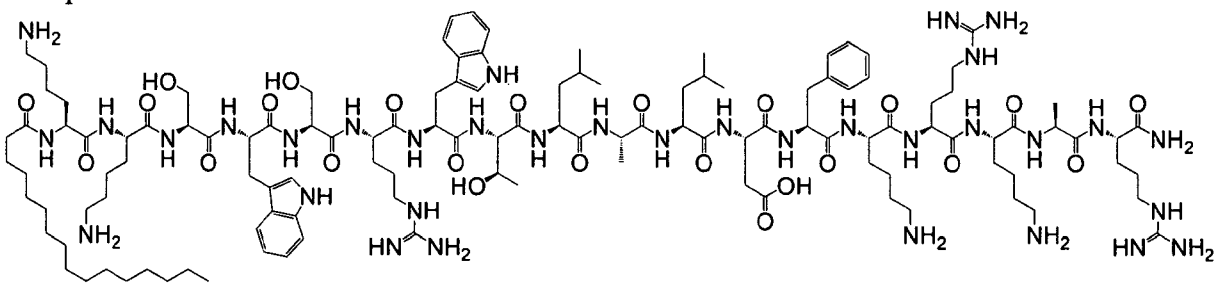


In yet another embodiment, a GPCR compound of the invention is selected from one of the following compounds or a pharmaceutically acceptable salt thereof:

Compound 96



Compound 97



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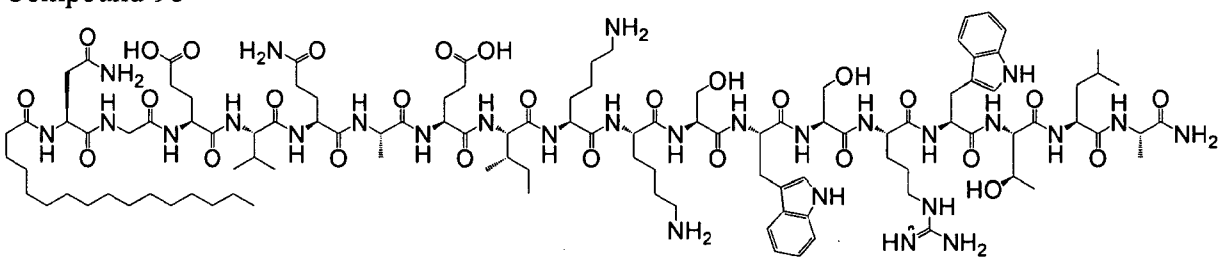

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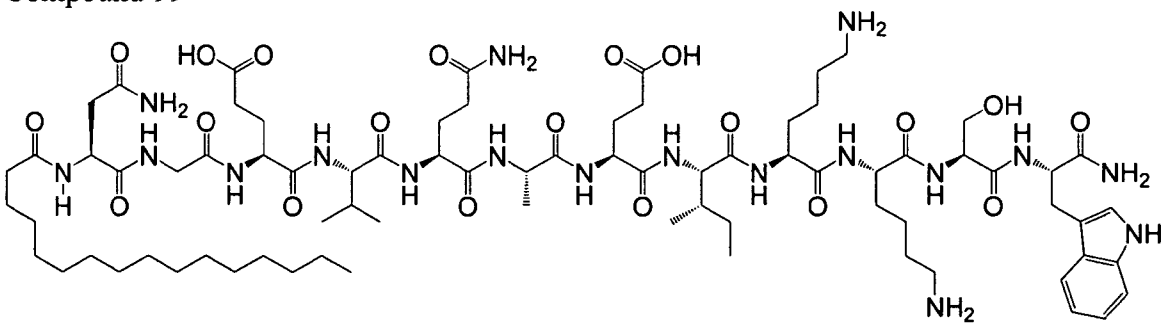

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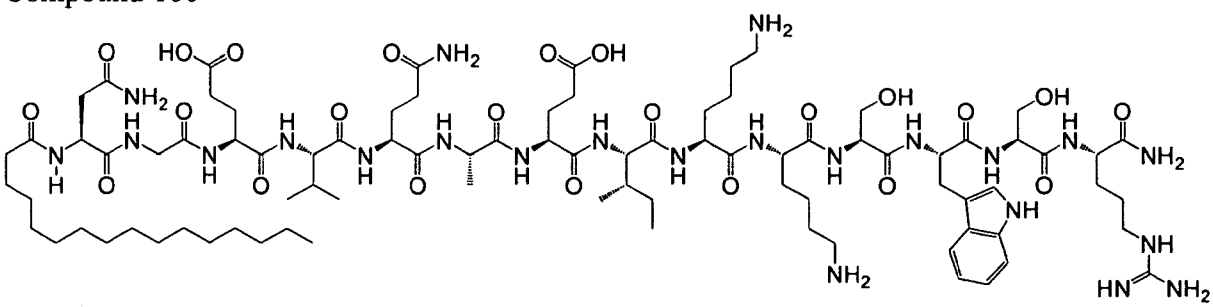

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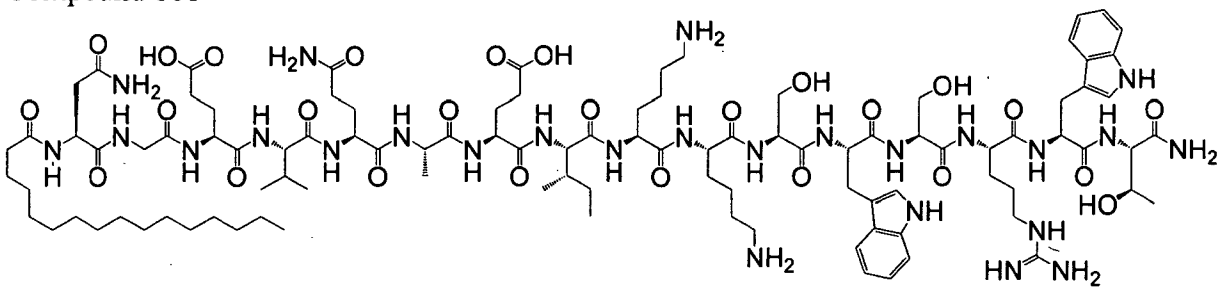

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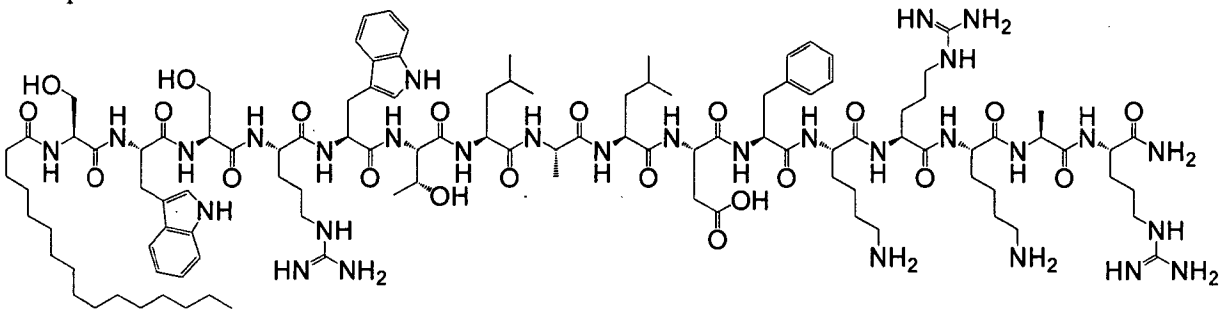

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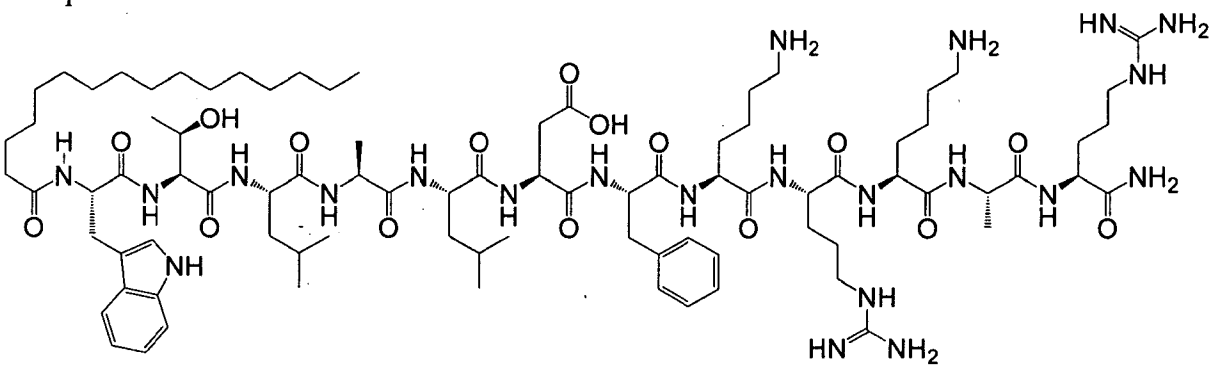

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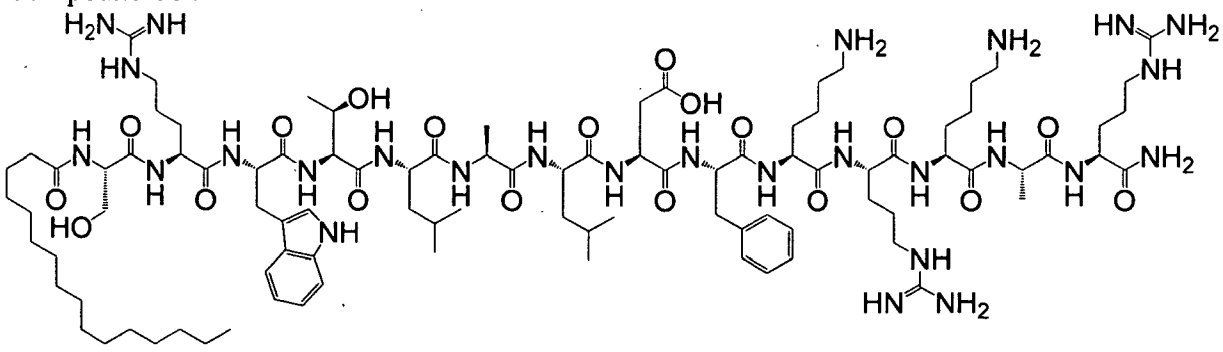

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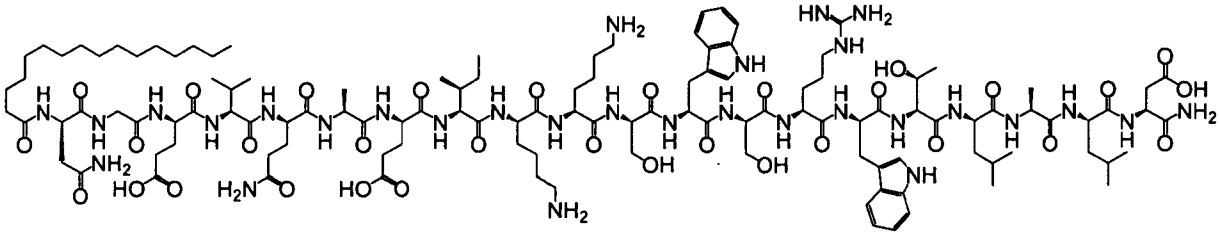


"Cycloalkyl" used alone or as part of a larger moiety such as "cycloalkylalkyl" refers to a monocyclic or polycyclic, non-aromatic ring system of 3 to 20 carbon atoms, 3 to 12 carbon atoms, or 3 to 9 carbon atoms, which may be saturated or unsaturated. Examples of cycloalkyl groups include cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cyclohexenyl, cyclohexa-1,3-dienyl, cyclooctyl, cycloheptanyl, norbornyl, adamantyl, and the like.

"Heterocycloalkyl" refers to a saturated or unsaturated, non-aromatic, monocyclic or polycyclic ring system of 3 to 20 atoms, 3 to 12 atoms, or 3 to 8 atoms, containing one to four ring heteroatoms chosen from O, N and S. Examples of heterocyclyl groups include pyrrolidine, piperidine, tetrahydrofuran, tetrahydropyran, tetrahydrothiophene, tetrahydrothiopyran, isoxazolidine, 1,3-dioxolane, 1,3-dithiolane, 1,3-dioxane, 1,4-dioxane, 1,3-dithiane, 1,4-dithiane, morpholine, thiomorpholine, thiomorpholine-1,1-dioxide, tetrahydro-2H-1,2-thiazine-1,1-dioxide, isothiazolidine-1,1-dioxide, pyrrolidin-2-one, piperidin-2-one, piperazin-2-one, and morpholin-2-one, and the like.

"Halogen" and "halo" refer to fluoro, chloro, bromo or iodo.

"Haloalkyl" refers to an alkyl group substituted with one or more halogen atoms. By analogy, "haloalkenyl", "haloalkynyl", etc., refers to the group (for example alkenyl or alkynyl) substituted by one or more halogen atomes.

"Cyano" refers to the group –CN.

"Oxo" refers to a divalent =O group.

"Thioxo" refers to a divalent =S group.

"Ph" refers to a phenyl group.

"Carbonyl" refers to a divalent –C(O)- group.

"Alkyl" used alone or as part of a larger moiety such as "hydroxyalkyl", "alkoxyalkyl", "alkylamine" refers to a straight or branched, saturated aliphatic group having the specified number of carbons, typically having 1 to 12 carbon atoms. More particularly, the aliphatic group may have 1 to 8, 1 to 6, or 1 to 4 carbon atoms. This term is exemplified

by groups such as methyl, ethyl, n-propyl, isopropyl, n-butyl, isobutyl, *tert*-butyl, n-hexyl, and the like.

"Alkenyl" refers to a straight or branched aliphatic group with at least one double bond. Typically, alkenyl groups have from 2 to 12 carbon atoms, from 2 to 8, from 2 to 6, or from 2 to 4 carbon atoms. Examples of alkenyl groups include ethenyl (-CH=CH_2), n-2-propenyl (allyl, -CH_2CH=CH_2), pentenyl, hexenyl, and the like.

"Alkynyl" refers to a straight or branched aliphatic group having at least 1 site of alkynyl unsaturation. Typically, alkynyl groups contain 2 to 12, 2 to 8, 2 to 6 or 2 to 4 carbon atoms. Examples of alkynyl groups include ethynyl (-C≡CH), propargyl (-CH_2C≡CH), pentynyl, hexynyl, and the like.

"Alkylene" refers to a bivalent saturated straight-chained hydrocarbon, e.g., C_1-C_6 alkylene includes –$(CH_2)_6$-, -CH_2-CH-$(CH_2)_3CH_3$, and the like. "Bivalent means that the alkylene group is attached to the remainder of the molecule through two different carbon atoms.

"Alkenylene" refers to an alkylene group with in which one carbon-carbon single bond is replaced with a double bond.

"Alkynylene" refers to an alkylene group with in which one carbon-carbon single bond is replaced with a triple bond.

"Aryl" used alone or as part of a larger moiety as in "aralkyl" refers to an aromatic carbocyclic group of from 6 to 14 carbon atoms having a single ring or multiple condensed rings. The term "aryl" also includes aromatic carbocycle(s) fused to cycloalkyl or heterocycloalkyl groups. Examples of aryl groups include phenyl, benzo[*d*][1,3]dioxole, naphthyl, phenantrenyl, and the like.

"Aryloxy" refers to an –OAr group, wherein O is an oxygen atom and Ar is an aryl group as defined above.

"Aralkyl" refers to an alkyl having at least one alkyl hydrogen atom replaced with an aryl moiety, such as benzyl, -$(CH_2)_2$phenyl, -$(CH_2)_3$phenyl, -CH$(phenyl)_2$, and the like.

"Alkyl cycloalkyl" refers to an alkyl having at least one alkyl hydrogen atom replaced with a cycloalkyl moiety, such as –CH_2-cyclohexyl, -CH_2-cyclohexenyl, and the like.

"Heteroaryl" used alone or a part of a larger moiety as in "heteroaralkyl" refers to a 5 to 14 membered monocyclic, bicyclic or tricyclic heteroaromatic ring system, containing one

to four ring heteroatoms independently selected from nitrogen, oxygen and sulfur. The term "heteroaryl" also includes heteroaromatic ring(s) fused to cycloalkyl or heterocycloalkyl groups. Particular examples of heteroaryl groups include optionally substituted pyridyl, pyrrolyl, pyrimidinyl, furyl, thienyl, imidazolyl, oxazolyl, isoxazolyl, thiazolyl, isothiazolyl, pyrazolyl, 1,2,3-triazolyl, 1,2,4-triazolyl, 1,2,3-oxadiazolyl, 1,2,4-oxadiazolyl, 1,2,5-oxadiazolyl, 1,3,4-oxadiazolyl,1,3,4-triazinyl, 1,2,3-triazinyl, benzofuryl, [2,3-dihydro]benzofuryl, isobenzofuryl, benzothienyl, benzotriazolyl, isobenzothienyl, indolyl, isoindolyl, 3H-indolyl, benzimidazolyl, imidazo[1,2-a]pyridyl, benzothiazolyl, benzoxazolyl, quinolizinyl, quinazolinyl, pthalazinyl, quinoxalinyl, cinnolinyl, napthyridinyl, pyrido[3,4-b]pyridyl, pyrido[3,2-b]pyridyl, pyrido[4,3-b]pyridyl, quinolyl, isoquinolyl, tetrazolyl, 1,2,3,4-tetrahydroquinolyl, 1,2,3,4-tetrahydroisoquinolyl, purinyl, pteridinyl, carbazolyl, xanthenyl, benzoquinolyl, and the like.

"Heteroaryloxy" refers to an –OHet group, wherein O is an oxygen atom and Het is a heteroaryl group as defined above.

"Heteroaralkyl" refers to an alkyl having at least one alkyl hydrogen atom replaced with a heteroaryl moiety, such as $-CH_2$-pyridinyl, $-CH_2$-pyrimidinyl, and the like.

"Alkoxy" refers to the group –O-R where R is "alkyl", "cycloalkyl", "alkenyl", or "alkynyl". Examples of alkoxy groups include for example, methoxy, ethoxy, ethenoxy, and the like.

"Alkyl heterocycloalkyl" refers to an alkyl having at least one alkyl hydrogen atom replaced with a heterocycloalkyl moiety, such as $-CH_2$-morpholino, $-CH_2$-piperidyl and the like.

"Alkoxycarbonyl" refers to the group –C(O)OR where R is "alkyl", "alkenyl", "alkynyl", "cycloalkyl", "heterocycloalkyl", "aryl", or "heteroaryl".

"Hydroxyalkyl" and "alkoxyalkyl" are alky groups substituted with hydroxyl and alkoxy, respectively.

"Amino" means $-NH_2$; "alkylamine" and "dialkylamine" mean –NHR and $-NR_2$, respectively, wherein R is an alkyl group. "Cycloalkylamine" and "dicycloalkylamine" mean –NHR and $-NR_2$, respectively, wherein R is a cycloalkyl group. "Cycloalkylalkylamine"

means –NHR wherein R is a cycloalkylalkyl group. "[Cycloalkylalkyl][alkyl]amine" means –$N(R)_2$ wherein one R is cycloalkylalkyl and the other R is alkyl.

Haloalkyl and halocycloalkyl include mono, poly, and perhaloalkyl groups where the halogens are independently selected from fluorine, chlorine, bromine and iodine.

Suitable substituents for "alkyl", "alkenyl", "alkynyl", "cycloalkyl", "heterocycloalkyl", "aryl", or "heteroaryl", etc., are those which will form a stable compound of the invention. Examples of suitable substituents are those selected from the group consisting of halogen, -CN, -OH, $-NH_2$, (C_1-C_4)alkyl, (C_1-C_4)haloalkyl, aryl, heteroaryl, (C_3-C_7)cycloalkyl, (5-7 membered) heterocycloalkyl, $-NH(C_1-C_6)$alkyl, $-N((C_1-C_6)alkyl)_2$, (C_1-C_6)alkoxy, (C_1-C_6)alkoxycarbonyl, $-CONH_2$, $-OCONH_2$, $-NHCONH_2$, $-N(C_1-C_6)alkylCONH_2$, $-N(C_1-C_6)alkylCONH(C_1-C_6)$alkyl, $-NHCONH(C_1-C_6)$alkyl, $-NHCON((C_1-C_6)alkyl)_2$, $-N(C_1-C_6)alkylCON((C_1-C_6)alkyl)_2$, $-NHC(S)NH_2$, $-N(C_1-C_6)alkylC(S)NH_2$, $-N(C_1-C_6)alkylC(S)NH(C_1-C_6)$alkyl, $-NHC(S)NH(C_1-C_6)$alkyl, $-NHC(S)N((C_1-C_6)alkyl)_2$, $-N(C_1-C_6)alkylC(S)N((C_1-C_6)alkyl)_2$, $-CONH(C_1-C_6)$alkyl, $-OCONH(C_1-C_6)$alkyl $-CON((C_1-C_6)alkyl)_2$, $-C(S)(C_1-C_6)$alkyl, $-S(O)_p(C_1-C_6)$alkyl, $-S(O)_pNH_2$, $-S(O)_pNH(C_1-C_6)$alkyl, $-S(O)_pN((C_1-C_6)alkyl)_2$, $-CO(C_1-C_6)$alkyl, $-OCO(C_1-C_6)$alkyl, $-C(O)O(C_1-C_6)$alkyl, $-OC(O)O(C_1-C_6)$alkyl, -C(O)H or $–CO_2H$. More particularly, the substituents are selected from halogen, -CN, -OH, $-NH_2$, (C_1-C_4)alkyl, (C_1-C_4)haloalkyl, (C_1-C_4)alkoxy, phenyl, and (C_3-C_7)cycloalkyl. Within the framework of this invention, said "substitution" is also meant to encompass situations where a hydrogen atom is replaced with a deuterium atom. p is an integer with a value of 1 or 2.

Pharmaceutically acceptable salts of the compounds disclosed herein are included in the present invention. For example, an acid salt of a compound containing an amine or other basic group can be obtained by reacting the compound with a suitable organic or inorganic acid, resulting in pharmaceutically acceptable anionic salt forms. Examples of anionic salts include the acetate, benzenesulfonate, benzoate, bicarbonate, bitartrate, bromide, calcium edetate, camsylate, carbonate, chloride, citrate, dihydrochloride, edetate, edisylate, estolate, esylate, fumarate, glyceptate, gluconate, glutamate, glycollylarsanilate, hexylresorcinate, hydrobromide, hydrochloride, hydroxynaphthoate, iodide, isethionate, lactate, lactobionate, malate, maleate, mandelate, mesylate, methylsulfate, mucate, napsylate, nitrate, pamoate, pantothenate, phosphate/diphospate, polygalacturonate, salicylate, stearate, subacetate, succinate, sulfate, tannate, tartrate, teoclate, tosylate, and triethiodide salts.

Salts of the compounds containing an acidic functional group can be prepared by reacting with a suitable base. Such a pharmaceutically acceptable salt can be made with a base which affords a pharmaceutically acceptable cation, which includes alkali metal salts (especially sodium and potassium), alkaline earth metal salts (especially calcium and magnesium), aluminum salts and ammonium salts, as well as salts made from physiologically acceptable organic bases such as trimethylamine, triethylamine, morpholine, pyridine, piperidine, picoline, dicyclohexylamine, N,N'-dibenzylethylenediamine, 2-hydroxyethylamine, bis-(2-hydroxyethyl)amine, tri-(2-hydroxyethyl)amine, procaine, dibenzylpiperidine, dehydroabietylamine, N,N'-bisdehydroabietylamine, glucamine, N-methylglucamine, collidine, quinine, quinoline, and basic amino acids such as lysine and arginine.

PHARMACEUTICAL COMPOSITIONS

The invention also provides pharmaceutical compositions comprising an effective amount of a compound Formula I (e.g., including any of the formulae herein), or a pharmaceutically acceptable salt of said compound; and a pharmaceutically acceptable carrier. The carrier(s) are "pharmaceuticallyacceptable" in that they are not deleterious to the recipient thereof in an amount used in the medicament.

Pharmaceutically acceptable carriers, adjuvants and vehicles that may be used in the pharmaceutical compositions of this invention include, but are not limited to, ion exchangers, alumina, aluminum stearate, lecithin, serum proteins, such as human serum albumin, buffer substances such as phosphates, glycine, sorbic acid, potassium sorbate, partial glyceride mixtures of saturated vegetable fatty acids, water, salts or electrolytes, such as protamine sulfate, disodium hydrogen phosphate, potassium hydrogen phosphate, sodium chloride, zinc salts, colloidal silica, magnesium trisilicate, polyvinyl pyrrolidone, cellulose-based substances, polyethylene glycol, sodium carboxymethylcellulose, polyacrylates, waxes, polyethylene-polyoxypropylene-block polymers, polyethylene glycol and wool fat.

If required, the solubility and bioavailability of the compounds of the present invention in pharmaceutical compositions may be enhanced by methods well-known in the art. One method includes the use of lipid excipients in the formulation. See "Oral Lipid-Based Formulations: Enhancing the Bioavailability of Poorly Water-Soluble Drugs (Drugs and the Pharmaceutical Sciences)," David J. Hauss, ed. Informa Healthcare, 2007;

and "Role of Lipid Excipients in Modifying Oral and Parenteral Drug Delivery: Basic Principles and Biological Examples," Kishor M. Wasan, ed. Wiley-Interscience, 2006.

Another known method of enhancing bioavailability is the use of an amorphous form of a compound of this invention optionally formulated with a poloxamer, such as LUTROL™ and PLURONIC™ (BASF Corporation), or block copolymers of ethylene oxide and propylene oxide. See United States patent 7,014,866; and United States patent publications 20060094744 and 20060079502.

The pharmaceutical compositions of the invention include those suitable for oral, rectal, nasal, topical (including buccal and sublingual), pulmonary, vaginal or parenteral (including subcutaneous, intramuscular, intravenous and intradermal) administration. In certain embodiments, the compound of the formulae herein is administered transdermally (e.g., using a transdermal patch or iontophoretic techniques). Other formulations may conveniently be presented in unit dosage form, e.g., tablets, sustained release capsules, and in liposomes, and may be prepared by any methods well known in the art of pharmacy. See, for example, Remington's Pharmaceutical Sciences, Mack Publishing Company, Philadelphia, PA (17th ed. 1985).

Such preparative methods include the step of bringing into association with the molecule to be administered ingredients such as the carrier that constitutes one or more accessory ingredients. In general, the compositions are prepared by uniformly and intimately bringing into association the active ingredients with liquid carriers, liposomes or finely divided solid carriers, or both, and then, if necessary, shaping the product.

In certain embodiments, the compound is administered orally. Compositions of the present invention suitable for oral administration may be presented as discrete units such as capsules, sachets, or tablets each containing a predetermined amount of the active ingredient; a powder or granules; a solution or a suspension in an aqueous liquid or a non-aqueous liquid; an oil-in-water liquid emulsion; a water-in-oil liquid emulsion; packed in liposomes; or as a bolus, etc. Soft gelatin capsules can be useful for containing such suspensions, which may beneficially increase the rate of compound absorption.

In the case of tablets for oral use, carriers that are commonly used include lactose and corn starch. Lubricating agents, such as magnesium stearate, are also typically added. For oral administration in a capsule form, useful diluents include lactose and dried cornstarch. When aqueous suspensions are administered orally, the active ingredient is combined with

emulsifying and suspending agents. If desired, certain sweetening and/or flavoring and/or coloring agents may be added.

Compositions suitable for oral administration include lozenges comprising the ingredients in a flavored basis, usually sucrose and acacia or tragacanth; and pastilles comprising the active ingredient in an inert basis such as gelatin and glycerin, or sucrose and acacia.

Compositions suitable for parenteral administration include aqueous and non-aqueous sterile injection solutions which may contain anti-oxidants, buffers, bacteriostats and solutes which render the formulation isotonic with the blood of the intended recipient; and aqueous and non-aqueous sterile suspensions which may include suspending agents and thickening agents. The formulations may be presented in unit-dose or multi-dose containers, for example, sealed ampules and vials, and may be stored in a freeze dried (lyophilized) condition requiring only the addition of the sterile liquid carrier, for example water for injections, immediately prior to use. Extemporaneous injection solutions and suspensions may be prepared from sterile powders, granules and tablets.

Such injection solutions may be in the form, for example, of a sterile injectable aqueous or oleaginous suspension. This suspension may be formulated according to techniques known in the art using suitable dispersing or wetting agents (such as, for example, Tween 80) and suspending agents. The sterile injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally-acceptable diluent or solvent, for example, as a solution in 1,3-butanediol. Among the acceptable vehicles and solvents that may be employed are mannitol, water, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium. For this purpose, any bland fixed oil may be employed including synthetic mono- or diglycerides. Fatty acids, such as oleic acid and its glyceride derivatives are useful in the preparation of injectables, as are natural pharmaceutically-acceptable oils, such as olive oil or castor oil, especially in their polyoxyethylated versions. These oil solutions or suspensions may also contain a long-chain alcohol diluent or dispersant.

The pharmaceutical compositions of this invention may be administered in the form of suppositories for rectal administration. These compositions can be prepared by mixing a compound of this invention with a suitable non-irritating excipient which is solid at room temperature but liquid at the rectal temperature and therefore will melt in the rectum to

release the active components. Such materials include, but are not limited to, cocoa butter, beeswax and polyethylene glycols.

The pharmaceutical compositions of this invention may be administered by nasal aerosol or inhalation. Such compositions are prepared according to techniques well-known in the art of pharmaceutical formulation and may be prepared as solutions in saline, employing benzyl alcohol or other suitable preservatives, absorption promoters to enhance bioavailability, fluorocarbons, and/or other solubilizing or dispersing agents known in the art. See, e.g.: Rabinowitz JD and Zaffaroni AC, US Patent 6,803,031, assigned to Alexza Molecular Delivery Corporation.

Topical administration of the pharmaceutical compositions of this invention is especially useful when the desired treatment involves areas or organs readily accessible by topical application. For topical application topically to the skin, the pharmaceutical composition should be formulated with a suitable ointment containing the active components suspended or dissolved in a carrier. Carriers for topical administration of the compounds of this invention include, but are not limited to, mineral oil, liquid petroleum, white petroleum, propylene glycol, polyoxyethylene polyoxypropylene compound, emulsifying wax, and water. Alternatively, the pharmaceutical composition can be formulated with a suitable lotion or cream containing the active compound suspended or dissolved in a carrier. Suitable carriers include, but are not limited to, mineral oil, sorbitan monostearate, polysorbate 60, cetyl esters wax, cetearyl alcohol, 2-octyldodecanol, benzyl alcohol, and water. The pharmaceutical compositions of this invention may also be topically applied to the lower intestinal tract by rectal suppository formulation or in a suitable enema formulation. Topically-transdermal patches and iontophoretic administration are also included in this invention.

Application of the patient therapeutics may be local, so as to be administered at the site of interest. Various techniques can be used for providing the patient compositions at the site of interest, such as injection, use of catheters, trocars, projectiles, pluronic gel, stents, sustained drug release polymers or other device which provides for internal access.

Thus, according to yet another embodiment, the compounds of this invention may be incorporated into compositions for coating an implantable medical device, such as prostheses, artificial valves, vascular grafts, stents, or catheters. Suitable coatings and the general preparation of coated implantable devices are known in the art and are exemplified in US

Patents 6,099,562; 5,886,026; and 5,304,121. The coatings are typically biocompatible polymeric materials such as a hydrogel polymer, polymethyldisiloxane, polycaprolactone, polyethylene glycol, polylactic acid, ethylene vinyl acetate, and mixtures thereof. The coatings may optionally be further covered by a suitable topcoat of fluorosilicone, polysaccharides, polyethylene glycol, phospholipids or combinations thereof to impart controlled release characteristics in the composition. Coatings for invasive devices are to be included within the definition of pharmaceutically acceptable carrier, adjuvant or vehicle, as those terms are used herein.

According to another embodiment, the invention provides a method of coating an implantable medical device comprising the step of contacting said device with the coating composition described above. It will be obvious to those skilled in the art that the coating of the device will occur prior to implantation into a mammal.

According to another embodiment, the invention provides a method of impregnating an implantable drug release device comprising the step of contacting said drug release device with a compound or composition of this invention. Implantable drug release devices include, but are not limited to, biodegradable polymer capsules or bullets, non-degradable, diffusible polymer capsules and biodegradable polymer wafers.

According to another embodiment, the invention provides an implantable medical device coated with a compound or a composition comprising a compound of this invention, such that said compound is therapeutically active.

According to another embodiment, the invention provides an implantable drug release device impregnated with or containing a compound or a composition comprising a compound of this invention, such that said compound is released from said device and is therapeutically active.

Where an organ or tissue is accessible because of removal from the patient, such organ or tissue may be bathed in a medium containing a composition of this invention, a composition of this invention may be painted onto the organ, or a composition of this invention may be applied in any other convenient way.

In another embodiment, a composition of this invention further comprises a second therapeutic agent. In one embodiment, the second therapeutic agent is one or more additional compounds of the invention.

In another embodiment, the second therapeutic agent may be selected from any compound or therapeutic agent known to have or that demonstrates advantageous properties when administered with a compound having the same mechanism of action as the PTHR1 receptor compound of Formula I.

In a particular embodiment, the second therapeutic is an agent useful in the treatment or prevention of a disease or condition selected from osteoporosis; humoral hypercalcemia of malignancy; osteolytic and osteoblastic metastasis to bone; primary and secondary hyperparathyroidism associated increase in bone absorption; vascular calcification; psychiatric disorders and cognitive disorders associated with hyperparathyroidism; dermatological disorders; and excess hair growth.

In another embodiment, the second therapeutic is an agent useful in the treatment or prevention of a disease or condition selected from humoral hypercalcemia of malignancy and primary and secondary hyperparathyroidism associated increase in bone absorption.

In one embodiment, the invention provides separate dosage forms of a compound of this invention and one or more of any of the above-described second therapeutic agents, wherein the compound and second therapeutic agent are associated with one another. The term "associated with one another" as used herein means that the separate dosage forms are packaged together or otherwise attached to one another such that it is readily apparent that the separate dosage forms are intended to be sold and administered together (within less than 24 hours of one another, consecutively or simultaneously).

In the pharmaceutical compositions of the invention, the compound of the present invention is present in an effective amount. As used herein, the term "effective amount" refers to an amount which, when administered in a proper dosing regimen, is sufficient to treat (therapeutically or prophylactically) the target disorder. For example, and effective amount is sufficient to reduce or ameliorate the severity, duration or progression of the disorder being treated, prevent the advancement of the disorder being treated, cause the regression of the disorder being treated, or enhance or improve the prophylactic or therapeutic effect(s) of another therapy. Preferably, the compound is present in the composition in an amount of from 0.1 to 50wt.%, more preferably from 1 to 30 wt.%, most preferably from 5 to 20wt.%.

The interrelationship of dosages for animals and humans (based on milligrams per meter squared of body surface) is described in Freireich et al., (1966) Cancer Chemother.

Rep 50: 219. Body surface area may be approximately determined from height and weight of the patient. See, e.g., Scientific Tables, Geigy Pharmaceuticals, Ardsley, N.Y., 1970, 537.

For pharmaceutical compositions that comprise a second therapeutic agent, an effective amount of the second therapeutic agent is between about 20% and 100% of the dosage normally utilized in a monotherapy regime using just that agent. Preferably, an effective amount is between about 70% and 100% of the normal monotherapeutic dose. The normal monotherapeutic dosages of these second therapeutic agents are well known in the art. *See*, e.g., Wells et al., eds., Pharmacotherapy Handbook, 2nd Edition, Appleton and Lange, Stamford, Conn. (2000); PDR Pharmacopoeia, Tarascon Pocket Pharmacopoeia 2000, Deluxe Edition, Tarascon Publishing, Loma Linda, Calif. (2000), each of which references are incorporated herein by reference in their entirety.

The compounds for use in the method of the invention can be formulated in unit dosage form. The term "unit dosage form" refers to physically discrete units suitable as unitary dosage for subjects undergoing treatment, with each unit containing a predetermined quantity of active material calculated to produce the desired therapeutic effect, optionally in association with a suitable pharmaceutical carrier. The unit dosage form can be for a single daily treatment dose or one of multiple daily treatment doses (e.g., about 1 to 4 or more times per day). When multiple daily treatment doses are used, the unit dosage form can be the same or different for each dose.

METHODS OF TREATMENT

As used herein the term "subject" and "patient" typically means a human, but can also be an animal in need of treatment, *e.g.*, companion animals (dogs, cats, and the like), farm animals (cows, pigs, horses, sheep, goats, and the like) and laboratory animals (rats, mice, guinea pigs, and the like).

The terms "treat" and "treating" are used interchangeably and include both therapeutic treatment and prophylactic treatment (reducing the likelihood of development). Both terms mean decrease, suppress, attenuate, diminish, arrest, or stabilize the development or progression of a disease (e.g., a disease or disorder delineated herein), lessen the severity of the disease or improve the symptoms associated with the disease.

"Disease" means any condition or disorder that damages or interferes with the normal function of a cell, tissue, or organ.

As used herein, the term "effective amount" refers to an amount which, when administered in a proper dosing regimen, is sufficient to treat (therapeutically or prophylactically) the target disorder. For example, and effective amount is sufficient to reduce or ameliorate the severity, duration or progression of the disorder being treated, prevent the advancement of the disorder being treated, cause the regression of the disorder being treated, or enhance or improve the prophylactic or therapeutic effect(s) of another therapy.

The invention also includes methods of treating diseases, disorders or pathological conditions which benefit from modulation of the PTHR1 receptor comprising administering an effective amount of an PTHR1 receptor compound of the invention to a subject in need thereof. Diseases and conditions which can benefit from modulation (inhibition or activation) of the PTHR1 receptor include, but are not limited to, osteoporosis; humoral hypercalcemia of malignancy; osteolytic and osteoblastic metastasis to bone; primary and secondary hyperparathyroidism associated increase in bone absorption; vascular calcification; psychiatric disorders and cognitive disorders associated with hyperparathyroidism; dermatological disorders; and excess hair growth.

Humoral hypercalcemia of malignancy is caused by secretion of parathyroid hormone related protein (PTHrP) by malignant tumor cell. PTHrP binds to PTH receptor leading to increase in bone turnover and hypercalcemia. PTHR1 receptor compounds of the invention having antagonist activity can block the effect of PTHrP at PTH receptor being suitable for use in treating symptoms associated with hypercalemia of malignancy.

PTHR1 receptor compounds of the invention having antagonist activity can be used to block the effect of uncontrolled secretion of PTH and thus control/reduce the symptoms of hyperparathyroidism and slow the progression from secondary hyperthyroidism to tertiary. In addition, PTHR1 receptor compounds of the invention can be used for treating psychiatric and cognitive disoreder associated with hyperparathyroidism (Curr Opin Oncol. 2007 Jan;19(1):1-5).

Although continuous elevation of PTH leads to bone loss, intermittent short elevation of this hormone can be anabolic for bone. Clinical benefit of PTH peptide in osteoporosis was established in 2001 and therapeutic use of PTH for osteoporosis was approved by U.S.

FDA in 2002. The success of PTH has raised the question if a purely anabolic PTH-related ligand can be achieved (Ann N Y Acad Sci. 2007 Nov;1117:196-208). PTHR1 receptor compounds of the invention can provide the unique opportunity to selectively modulate downstream effectors from inside of the receptor.

PTHR1 receptor antagonist compounds can also be used for preventing or treating tumor growth stimulated by PTHrP (recent reference: Int J Cancer. 2008 Aug 26), for treating dermatological disorders and for hair growth promotion (Endocrinology. 2007 Mar;148(3):1167-70).

In one embodiment, an effective amount of a compound of this invention can range from about .005 mg to about 5000 mg per treatment. In more specific embodiments, the range is from about .05 mg to about 1000 mg, or from about 0.5 mg to about 500 mg, or from about 5 mg to about 50 mg. Treatment can be administered one or more times per day (for example, once per day, twice per day, three times per day, four times per day, five times per day, etc.). When multiple treatments are used, the amount can be the same or different. It is understood that a treatment can be administered every day, every other day, every 2 days, every 3 days, every 4 days, every 5 days, etc. For example, with every other day administration, a treatment dose can be initiated on Monday with a first subsequent treatment administered on Wednesday, a second subsequent treatment administered on Friday, etc. Treatment is typically administered from one to two times daily. Effective doses will also vary, as recognized by those skilled in the art, depending on the diseases treated, the severity of the disease, the route of administration, the sex, age and general health condition of the patient, excipient usage, the possibility of co-usage with other therapeutic treatments such as use of other agents and the judgment of the treating physician.

Alternatively, the effective amount of a compound of the invention is from about 0.01 mg/kg/day to about 1000 mg/kg/day, from about 0.1 mg/kg/day to about 100 mg/kg/day, from about 0.5 mg/kg/day to about 50 mg/kg/day, or from about 1 mg/kg/day to 10 mg/kg/day.

In another embodiment, any of the above methods of treatment comprises the further step of co-administering to said patient one or more second therapeutic agents. The choice of second therapeutic agent may be made from any second therapeutic agent known to be useful for co-administration with a compound that modulates the PTHR1 receptor. The choice of

second therapeutic agent is also dependent upon the particular disease or condition to be treated. Examples of second therapeutic agents that may be employed in the methods of this invention are those set forth above for use in combination compositions comprising a compound of this invention and a second therapeutic agent.

The term "co-administered" as used herein means that the second therapeutic agent may be administered together with a compound of this invention as part of a single dosage form (such as a composition of this invention comprising a compound of the invention and an second therapeutic agent as described above) or as separate, multiple dosage forms. Alternatively, the additional agent may be administered prior to, consecutively with, or following the administration of a compound of this invention. In such combination therapy treatment, both the compounds of this invention and the second therapeutic agent(s) are administered by conventional methods. The administration of a composition of this invention, comprising both a compound of the invention and a second therapeutic agent, to a subject does not preclude the separate administration of that same therapeutic agent, any other second therapeutic agent or any compound of this invention to said subject at another time during a course of treatment.

In one embodiment of the invention, where a second therapeutic agent is administered to a subject, the effective amount of the compound of this invention is less than its effective amount would be where the second therapeutic agent is not administered. In another embodiment, the effective amount of the second therapeutic agent is less than its effective amount would be where the compound of this invention is not administered. In this way, undesired side effects associated with high doses of either agent may be minimized. Other potential advantages (including without limitation improved dosing regimens and/or reduced drug cost) will be apparent to those of skill in the art.

KITS

The present invention also provides kits for use to treat the target disease, disorder or condition. These kits comprise (a) a pharmaceutical composition comprising a compound of Formula I, or a salt thereof, wherein said pharmaceutical composition is in a container; and (b) instructions describing a method of using the pharmaceutical composition to treat the target disease, disorder or condition.

The container may be any vessel or other sealed or sealable apparatus that can hold said pharmaceutical composition. Examples include bottles, ampules, divided or multi-chambered holders bottles, wherein each division or chamber comprises a single dose of said composition, a divided foil packet wherein each division comprises a single dose of said composition, or a dispenser that dispenses single doses of said composition. The container can be in any conventional shape or form as known in the art which is made of a pharmaceutically acceptable material, for example a paper or cardboard box, a glass or plastic bottle or jar, a re-sealable bag (for example, to hold a "refill" of tablets for placement into a different container), or a blister pack with individual doses for pressing out of the pack according to a therapeutic schedule. The container employed can depend on the exact dosage form involved, for example a conventional cardboard box would not generally be used to hold a liquid suspension. It is feasible that more than one container can be used together in a single package to market a single dosage form. For example, tablets may be contained in a bottle, which is in turn contained within a box. In one embodiment, the container is a blister pack.

The kits of this invention may also comprise a device to administer or to measure out a unit dose of the pharmaceutical composition. Such device may include an inhaler if said composition is an inhalable composition; a syringe and needle if said composition is an injectable composition; a syringe, spoon, pump, or a vessel with or without volume markings if said composition is an oral liquid composition; or any other measuring or delivery device appropriate to the dosage formulation of the composition present in the kit.

In certain embodiment, the kits of this invention may comprise in a separate vessel of container a pharmaceutical composition comprising a second therapeutic agent, such as one of those listed above for use for co-administration with a compound of this invention.

GENERAL METHODS FOR PREPARING PTHR1 RECEPTOR COMPOUNDS

Synthesis of Peptides

The peptide component (P) of the compounds of the invention can be synthesized by incorporating orthogonally protected amino acids in a step-wise fashion. Any suitable synthetic methods can be used. Traditional Fmoc or Boc chemistry can be easily adapted to provide the desired peptide component (P) of the compounds of the invention. Fmoc is generally preferred, because the cleavage of the Fmoc protecting group is milder than the

acid deprotection required for Boc cleavage, which requires repetitive acidic deprotections that lead to alteration of sensitive residues, and increase acid catalyzed side reactions.(G. B. FIELDS et al. in *Int. J. Pept. Protein,* 1990, 35, 161).

The peptides can be assembled linearly via Solid Phase Peptide Synthesis (SPPS), can be assembled in solution using modular condensations of protected or unprotected peptide components or a combination of both.

Solid Phase Peptide Synthesis

For SPPS, an appropriate resin is chosen that will afford the desired moiety on the C-terminus upon cleavage. For example upon cleavage of the linear peptide, a Rink amide resin will provide a primary amide on the C-terminus, whereas a Rink acid resin will provide an acid. Rink acid resins are more labile than Rink amide resins and the protected peptide could also be cleaved and subsequently the free acid activated to react with amines or other nucleophiles. Alternatively, other resins could provide attachment of other moieties prior to acylation, leading to cleavage of an alkylated secondary amide, ester or other desired C-terminal modification. A review of commonly used resins and the functional moiety that results after cleavage can be found in manufacturer literature such as NovaBiochem or Advanced Chemtech catalogues.

Typically a resin is chosen such that after cleavage the C-terminus is an amide bond. Rink amide resin is a resin that results in a C-terminal amide during cleavage. The orthogonally protected Fmoc amino acids are added stepwise using methods well known in literature (Bodansky M. Principles of Peptide synthesis (1993) 318p; Peptide Chemistry, a Practical Textbook (1993); Spinger-Verlag). These procedures could be done manually or by using automated peptide synthesizers.

The process involves activating the acid moiety of a protected amino acid, using activating agents such as HBTU, HATU, PyBop or simple carbodiimides. Often an additive is used to decrease racemization during coupling such as HOBt or HOAt (M. SCHNÖLZER et al., *Int. J. Pept. Protein Res.,* 1992, 40, 180). Manually, the coupling efficiency can be determined photometrically using a ninhydrin assay. If the coupling efficiency is below 98%, a second coupling may be desired. After the second coupling a capping step may be employed to prevent long deletion sequences to form, simplifying the purification of the

desired final compound. With automation, second couplings are not commonly required, unless a residue is known to be problematic such as Arginine.

Deprotection of the Fmoc is most commonly accomplished using piperidine (20%) in dimethylformamide (DMF). Alternatively other secondary amines may also be used such as morpholine, diethylamine or piperazine. This reaction is facile and normally is accomplished within 20 minutes using piperidine. After deprotection the resin is washed several times with DMF and DCM prior to coupling with the next residue. This process is repeated, assembling the peptide linearly until the sequence is complete. The final Fmoc is removed, which allows for coupling with the tether moiety.

In a preferred synthesis, the peptide is formed by SPPS accomplished manually or in an automated fashion using a commercially available synthesizer such as the CEM Microwave peptide synthesizer, Rainin Symphony synthesizer, or ABI 433 flow-through synthesizer. Commercially available Rink Amide resin is used for synthesizing the C-terminal amide peptides (*Rink, H. Tetrahedron Lett, 28, 4645, 1967*). Peptide synthesis reagents (coupling, deprotection agents) are commercially available and include HOBT, HBTU (Novabiochem) as well as DMF, DCM, Piperidine, NMP, and DIEA (Sigma-Aldrich). Suitably protected amino acids for use in solid phase peptide synthesis are commercially available from many sources, including Sigma-Aldrich and CEM Corporation.

For example, a convenient preparation of peptides on a 0.1mmol or 0.25 mmol scale uses Rink amide solid-phase resin with a substitution of about 0.6mmol/g. Linear attachment of the amino acids is accomplished on a ABI continuous flow automated synthesizer using 5 eq of orthogonally protected amino acid (AA), and using HBTU/HOBt coupling protocol, (5 eq. of each reagent). In another preferred synthesis, peptides can be synthesized using a microwave instrument using 10 eq of reagents. Deprotection of Fmoc can be accomplished with 20% piperidine in DMF followed by washing with DMF and DCM.

In both cases (i.e., Rink acid and Rink amide resins), final Fmoc deprotection of the N-terminus would leave a free amine after cleavage from the resin unless it is modified prior to cleavage. In the compounds of the invention, tether moieties are attached through amide bonds.

Solution Phase Synthesis of Peptides

For solution phase synthesis the desired peptide is generally broken down into peptide fragments in units of 2-4 amino acids. The selected unit is dependent on the sequence, the stability of the fragment to racemization, and the ease of assembly. As each amino acid is added, only 1-1.5eq of the residue is required, versus the 5-10 equivalents of reagent required for SSPS. Preactivated amino acids such as OSu active ester and acid fluorides also can be used, requiring only a base for completion of the reaction.

Coupling times require 1.5-2 hours for each step. Two fragments are condensed in solution, giving a larger fragment that then can be further condensed with additional fragments until the desired sequence is complete. The solution phase protocol uses only 1eq of each fragment and will use coupling reagents such as carbodiimides (DIC). For racemized prone fragments, PyBop or HBTU/HOBt can be used. Amino acids with Bsmoc/tBu or Fmoc/tBu and Boc/Benzyl protection are equally suitable for use.

When Fmoc is used, the use of 4-(aminomethyl) piperidine or tris(2-aminoethyl)amine as the deblocking agent can avoid undesired side reactions. The resulting Fmoc adduct can be extracted with a phosphate aqueous buffer of pH 5.5 (Organic Process Research & Development 2003, *7*, 2837). If Bsmoc is used, no buffer is required, only aqueous extractions are needed. Deprotections using these reagents occur in 30-60 minutes. Deblocking of the Fmoc group on the N-terminal residue provides a free terminal amine that is used for attachment of the tether moiety. In the compounds of the invention, tether moieties are attached through amide bonds to the N-terminal amine.

One advantage of solution phase synthesis is the ability to monitor the compound after every coupling step by mass spectrometry to see that the product is forming. In addition, a simple TLC system could be used to determine completion of reaction.

Attachment of Tethers

Tethers are attached to the terminal nitrogen of the N-terminal amino acid of the peptide chain using amide bond coupling:

The tether can be attached using solid phase procedures or in solution using an amide bond coupling. After the N-terminus is suitably coupled, the final compound is cleaved from the resin using an acidic cocktail (Peptide Synthesis and Applications, John Howl, Humana Press, 262p, 2005) . Typically these cocktails use concentrated trifluoroacetic acid (80-95%) and various scavengers to trap carbocations and prevent side chain reactions. Typical scavengers include isopropylsilanes, thiols, phenols and water. The cocktail mixture is determined by the residues of the peptide. Special care needs to be taken with sensitive residues, such as methionine, aspartic acid, and cysteine. Typical deprotection occurs over 2-5 hours in the cocktail. A preferred deprotection cocktail include the use of triisopropylsilane (TIS), Phenol, thioanisole, dodecanethiol (DDT) and water. Methane sulfonic acid (MSA) may also be used in the cocktail (4.8%). A more preferred cocktail consists of (TFA:MSA:TIS:DDT:Water 82: 4.5:4.5:4.5:4.5; 10 mL/0.1 mmol resin).

After deprotection, the resin is removed via filtration, and the final compound is isolated via precipitation from an organic solvent such as diethyl ether, m-*tert*-butyl ether, or ethyl acetate and the resulting solid collected via filtration or lyophilized to a powder. Purification of the peptide using reverse phase HPLC may be required to achieve sufficient purity. Generally, a gradient of aqueous solvent with an organic solvent will provide sufficient separation from impurities and deletion sequences. Typically 0.1%TFA is used as the aqueous and organic modifier, however, other modifiers such as ammonium acetate can also be used. After purification, the compound is collected, analyzed and fractions of sufficient purity are combined and lyophilized, providing the compound as a solid.

Amino acid reagents

The following commercially available orthogonally protected amino acids used can be used in the synthesis of compounds of the invention: Fmoc-Tyr(tBu)-OH, Fmoc-Ala-OH*H_2O, Fmoc-Arg(Pbf)-OH, Fmoc, Asn(Trt)-OH, Fmoc-Asp(tBu), Fmoc-Cys(tBu)-OH, Fmoc-Glu(tBu)-OH, Fmoc-Glx(Pbf)-OH, Fmoc-Gly-OH, Fmoc-His(Trt)-OH, Fmoc-Leu-OH, Fmoc-Ile-OH, Fmoc, Lys(tBu)-OH, Fmoc-Met-OH, Fmoc-Phe-OH, Fmoc-Ser(tBu)-OH, Fmoc-Thr(tBu)-OH, Fmoc-Typ-OH, and Fmoc-Val-OH. Additional amino acids suitable for incorporation into the compounds of the invention (e.g., D amino acids, substituted amino

acids and other protecting group variations) are also commercially available or synthesized by methods known in the art.

Analytical Methods

The compounds of the invention are analyzed for purity by HPLC using the methods listed below. Purification is achieved by preparative HPLC.

Fast LC/MS Method

Column:	Phenomenex Luna C-5 20x 30mm
Flow:	1.0 ml/min
Solvent A:	0.1 % TFA in Type I water
Solvent B:	0.1% TFA in Acetonitrile
UV	220 nm
Injection:	20 ul
Gradient	5-95%B (7 minutes); 95-5%B (1 minute); 5% B (4 minutes)

Analytical Purity Method

Column:	Phenomenex Luna C-5 20x 30mm
Flow:	1.0 ml/min
Solvent A:	0.1 % TFA in Type I water
Solvent B:	0.1% TFA in Acetonitrile
UV:	220 nm
Injection:	20 ul
Gradient:	2-95%B (10 minutes); 95-2%B (2 minutes); 2% B (2 minutes)

Preparative LC/MS Method

Column:	Phenomenex Luna C-5 250x 150mm
Flow:	5.0 ml/min
Solvent A:	0.1 % TFA in Type I water
Solvent B:	0.1% TFA in Acetonitrile
UV:	220 nm
Injection:	900 ul
Gradient:	35%B (5 minutes); 35-85%B (13 minutes); 85-35% B (0.5 minutes); 35%B (1.5 minutes)

Synthesis of Selected Compounds

Compound 82 Pal- TNAGRSATRQQYRKLL-amide

Compound 82 was synthesized as described above on Rink amide resin at 0.1 mmol scale. Amino acids were coupled sequentially as described above. Following deprotection of the Fmoc group on the N-terminal residue serine, the N-terminal amine was capped with

palmitic acid (10 eq.), HBTU (10 eq.) and DIEA (10 eq.) as described above. The pepducin was cleaved from the resin by TFA containing MS, TIS, DDT, and water (82: 4.5:4.5:4.5:4.5; 10 mL), filtered through a Medium frit Buchner full, triturated with ether and the resulting precipitate collected by centrifugation. Crude peptide was taken up in minimum amount of DMSO and purified by RP-HPLC as described previously. Fractions with correct MW were pooled and lyophilized and analyzed for purity using Method A. The yield of representative lots is illustrated in the following table.

Lot #	Yield (mg)
1	7.3

Compound 41 Pal- GSEKKYLWGFTVF-amide

Compound 41 was synthesized as described for Compound 82. The yield of representative lots is illustrated in the following table.

Lot #	Yield (mg)
1	2.6

Compound 105 Pal- NGEVQAEIKKSWSRWTLALD-amide

Compound 105 was synthesized as described for Compound 82. The yield of representative lots is illustrated in the following table.

Lot #	Yield (mg)
1	0.6

Additional compounds that were synthesized following the above-described method are listed in Tables below.

Compound #	Loop	Sequence	MS Theoretical	MS Observed
Compound 1	il	RRLHSTRNYIHMH	980.192	979.7
Compound 2	il	RRLHSTRNYIH	846.024	746
Compound 3	il	LAYFRRLHSTRNY	968.167	968.2
Compound 4	il	LAYFRRLHSTR	829.529	829.5

Compound 5	il	LAYFRRLHSTRNYIH	729.210	729
Compound 6	il	YFRRLHSTRNYIH	2000.160	2000.04
Compound 7	il	AYFRRLHSTRNYIH	2071.200	2071.15
Compound 8	il	FRRLHSTRNYIH	1837.100	1837.52
Compound 9	il	LAYFRRLHSTRNYI	2047.230	2047.31
Compound 10	il	LAYFRRLHSTRNYIHMH	818.655	818.3
Compound 11	il	GSYFRRLHSTRNYIH	715.841	715.5
Compound 12	il	SSYFRRLHSTRNYIH	725.850	725.5
Compound 13	il	GGYFRRLHSTRNYIH	705.832	705.3
Compound 14	il	LAYFRRLHSTRN	886.580	866.1
Compound 15	il	RRLHSTRNYIHM	911.622	911.7
Compound 16	il	RRLHSTRNYIHMHL	691.513	691.5
Compound 17	il	SGRRLHSTRNYIHMH	701.837	701.7
Compound 18	il	LAAFRRLHSTRNYIH	698.512	698.45
Compound 19	il	LAYARRLHSTRNYIH	703.845	703.75
Compound 20	il	LAYFARLHSTRNYIH	700.841	700.75
Compound 21	il	LAYFRRAHSTRNYIH	715.183	715.2
Compound 22	il	LAYFRRLHSTANYIH	700.841	700.8
Compound 23	il	LAYFRRLHSTRNYAH	715.183	715.2
Compound 24	il	LAYFRRLHSTRNYIA	707.190	707.2
Compound 25	il	LAYFKRLHSTRNYIH	719.872	719.85
Compound 26	il	LAYFRKLHSTRNYIH	719.872	719.8
Compound 27	il	LAYFRRLHSTKNYIH	719.872	719.95
Compound 28	il	LAYFRALHSTRNYIH	700.841	700.8
Compound 29	il	LAYFRRLASTRNYIH	707.190	707.1
Compound 30	il	LAYFRRLHATRNYIH	723.877	723.8
Compound 31	il	LAYFRRLHSARNYIH	719.201	719.1
Compound 32	il	LAYFRRLHSTRAYIH	714.868	714.8

Compound	Loop	Sequence	MS Theoretical	MS Observed

Compound 33	i2	HSLIFMAFFSEKKYL	700.544	700.4
Compound 34	i2	LYLHSLIFMAFFSEKKYLWGFT	993.554	993.7
Compound 35	i2	YLHSLIFMAFFSEKKYLWGFT	955.857	955.9
Compound 36	i2	HSLIFMAFFSEKKYLWGFT	1295.215	1295.5
Compound 37	i2	LHSLIFMAFFSEKKYLWGFT	902.110	901.9
Compound 38	i2	LYLHSLIFMAFFSEKKYLWG	911.461	911.3
Compound 39	i2	LYLHSLIFMAFFSEKKYL	1244.220	1244.3
Compound 40	i2	LYLHSLIFMSFFSEKK	1106.900	1106.6
Compound 41	i2	GSEKKYLWGFTVF	900.601	900
Compound 42	i2	GSEKKYLWGFT	777.448	777
Compound 43	i2	GSEKKYLWG	653.310	652.9

Compound	Loop	Sequence	MS Theoretical	MS Observed
Compound 44	i3	NIVRVLATKLRETNAGRSD	2351.802	2351.68
Compound 45	i3	VRVLATKLRETNAGRSDTR	2381.832	2381.42
Compound 46	i3	VLATKLRETNAGRSDTRQQ	2382.773	2382
Compound 47	i3	KLRETNAGRSDTRQQRKLL	2508.976	2508.42
Compound 48	i3	RETNAGRSDTRQQRKLLKS	2482.896	2482.61
Compound 49	i3	TNAGRSDTRQQRKLLKSTL	2411.858	2411.1
Compound 50	i3	KRETNAGRSDTRQQYRKLL	853.167	853.5
Compound 51	i3	RETNAGRSDTRQQYRKLLKS	882.690	882.6
Compound 52	i3	TNAGRSDTRQQYRKLLKSTL	859.010	859
Compound 53	i3	KLRETNAGRSDTRQQY	721.159	721
Compound 54	i3	NIVRVLATKLRETNAGR	717.213	717.2
Compound 55	i3	NIVRVLATKLRE	550.705	550.55
Compound 56	i3	DTRQQYRKLLKSTL	663.477	663.4
Compound 57	i3	RQQYRKLLKSTL	591.414	591.3
Compound 58	i3	RETNAGRSDTRQQYRKLLFS	889.024	889.3
Compound 59	i3	RETNAGRSDTRQQYRKLL	810.940	811.05

Compound 60	i3	RETNAGRSDTRQQYRK	735.501	735.4
Compound 61	i3	RETNAGRSDTRQQYRF	741.835	741.8
Compound 62	i3	RETNAGRSDTRQQY	640.715	640.3
Compound 63	i3	RETNAGRSDTRQ	543.615	543.55
Compound 64	i3	TNAGRSDTRQQYRKLLKS	787.590	787.4
Compound 65	i3	AGRSDTRQQYRKLLKS	715.854	715.8
Compound 66	i3	RSDTRQQYRKLLKS	673.145	673
Compound 67	i3	DTRQQYRKLLKS	592.057	592.15
Compound 68	i3	TNAGRSDTRQQYRKLL	715.840	715.7
Compound 69	i3	TNAGRSDTRQQYRK	640.402	640.3
Compound 70	i3	TNAGRSDTRQQYRF	646.750	646.5
Compound 71	i3	SGRVLATKLR	446.913	447.2
Compound 72	i3	SGRVLATKLRET	523.653	523.5
Compound 73	i3	SGRVLATKLRETNA	585.379	585.2656.3
Compound 74	i3	SGRVLATKLRETNAGR	656.458	656.3
Compound 75	i3	RVLATKLRETNAGR	911.585	911.5
Compound 76	i3	TNAGASDTRQQYRKLL	1030.706	1030.7
Compound 77	i3	TNAGRADTRQQYRKLL	710.507	710.2
Compound 78	i3	TNAGRSDTAQQYRKLL	1030.706	1031
Compound 79	i3	TNAGRSDARQQYRKLL	705.831	706
Compound 80	i3	TNAARSDTRQQYRKLL	720.515	720.1
Compound 81	i3	TNAGRSDTRAQYRKLL	696.823	696.5
Compound 82	i3	TNAGRSATRQQYRKLL	701.170	701
Compound 83	i3	TNAGRSDTRQQYRKLLF	764.898	764.8
Compound 84	i3	TNAGRSDTRQQYRKLLK	758.564	758.45
Compound 85	i3	TNAGRSDTRQQYRKLLFS	793.924	793.85
Compound 86	i3	TNAGRSDTRQQYRKLLFA	788.591	788.5
Compound 87	i3	TNAGRSDTRQQYRKLLA	739.533	739.8
Compound 88	i3	TNAGRSDTRQQYRKLA	701.813	701.7
Compound 89	i3	TNAGRSDTRQQYRKAL	701.813	702.15

Compound 90	i3	TNAGRSDTRQQYRALL	696.809	696.25
Compound 91	i3	TNAGRSDTRQQYAKLL	687.471	687.35
Compound 92	i3	TNAGRSDTRQQARKLL	685.142	685.1
Compound 93	i3	TNAGRSDTRQAYRKLL	696.823	696.8
Compound 94	i3	AGRSDTRQQYRKLLFA	716.855	716.75
Compound 95	i3	AGRSDTRQQYRKLLFS	722.188	722.1

Compound	Loop	Sequence	MS Theoretical	MS Observed
Compound 96	i4	EIKKSWSRWTLALDFKRKAR	920.123	919.8
Compound 97	i4	KKSWSRWTLALDFKRKAR	838.850	839.1
Compound 98	i4	NGEVQAEIKKSWSRWTLA	781.254	780.9
Compound 99	i4	NGEVQAEIKKSW	813.975	814
Compound 100	i4	NGEVQAEIKKSWSR	935.606	935
Compound 101	i4	NGEVQAEIKKSWSRWT	1078.600	1078.6
Compound 102	i4	SWSRWTLALDFKRKAR	753.917	753.7
Compound 103	i4	WTLALDFKRKAR	581.734	581.6
Compound 104	i4	SRWTLALDFKRKAR	662.821	662.6
Compound 105	i4	NGEVQAEIKKSWSRWTLALD	1285.503	1285.2

METHODS OF SCREENING

FUNCTIONAL ASSAYS

Functional assays suitable for use in detecting and characterizing GPCR signaling include Gene Reporter Assays and Calcium Flux assays, cAMP and kinase activation assays. Several suitable assays are described in detail below.

Gene Reporter Assays

Cells expressing the GPCR of interest can be transiently or stably transfected with a reporter gene plasmid construct containing an enhancer element which responds to activation of a second messenger signaling pathway or pathways, thereby controlling transcription of a cDNA encoding a detectable reporter protein. GPCR expression can be the result of endogenous expression on a cell line or cell type or the result of stable or transient

transfection of DNA encoding the receptor of interest into a cell line by means commonly used in the art. Immortalized cell lines or primary cell cultures can be used.

If the activated pathway is stimulatory (e.g., Gs or Gq for PTHR1), agonist activity results in activation of transcription factors, in turn causing an increase in reporter gene transcription, detectable by an increase in reporter activity. To test for agonist or inverse agonist activity, cells expressing the GPCR and the reporter gene construct can be challenged by the test compound for a predetermined period of time (e.g., 2-12 hours, typically 4 hours). Cells can then be assessed for levels of reporter gene product.Inverse agonists will suppress levels of reporter to below basal levels in a dose dependent manner.To test for antagonist or inhibitory activity through a stimulatory pathway, cells expressing both the GPCR and the reporter gene construct can be activated by a receptor agonist to increase gene reporter product levels. Treatment with antagonists will counter the effect of agonist stimulation in a dose- and receptor-dependent manner.

To test for agonist activity on receptor signaling through an inhibitory pathway, cells can be treated with a systematic activator (e.g., forskolin) to increase levels of reporter gene product. Activation of Gi by treatment with receptor agonist will inhibit this expression by inhibiting adenylyl cyclase. To screen for antagonist activity, test compounds can be assessed for the ability to counter agonist inhibition of adenylyl cyclase, resulting in increase reporter transcription.

Alternatively, a plasmid construct expressing the promiscuous G-protein Ga16 can be used to obtain a positive signal from a GPCR which normally couples to an inhibitory G-protein. Co-expression of the chimeric G-protein Gaq/Gai5 (Coward et al. Analytical Biochemistry 270, 242–248 (1999)) allows coupling to Gi-coupled receptors and conversion of second messenger signaling from the inhibitory Gi pathway to the stimulatory Gq pathway. Agonist and antagonist assessment in these systems is the same as the stimulatory pathways. Well-to-well variation caused by such factors as transfection efficiency, unequal plating of cells, and cell survival rates can be normalized in transient transfection assays by co-transfecting a constitutively expressing reporter gene with a non-interfering signal independent of the regulated reporter.

Calcium Flux Assay

Calcium Flux Assay is one of the most popular cell-based GPCR functional assays. It most often uses calcium sensing fluorescent dyes such as fura2 AM, fluo-4 and Calcium-4 to

measure changes in intracellular calcium concentration. It is used mainly to detect GPCR signaling via Gαq subunit. Activation of these Gq-coupled GPCRs leads to activation of phospholipase C, which subsequently leads to increase in inositol phosphate production. IP3 receptors on endoplasmic reticulum sense the change then release calcium into cytoplasm. Intracellular calcium binding to the fluorescent dyes can be detected by instruments that quantify fluorescent intensities, such as FLIPR Tetra, Flexstation (MDS) and FDSS (Hamamatsu). In additional to assess Gq-couple receptor signaling, calcium flux assay can also be used to study Gs and Gi couple receptors by co-expressing CNG (cycic nucleotide gated calcium channel) or chimeric G-proteins (Gqi5, Gsi5 for example). Activation of some Gi-coupled receptors can also be detected by calcium flux assay via Gβγ mediated phospholipase C activation.

HTRF cAMP Assay and IP-One Assay (Cisbio)

HTRF (homogeneous time resolved fluorescence) is a technology developed by Cisbio Bioassays based on TR-FRET (time-resolved fluorescence resonance energy transfer). Cisbio Bioassays has developed a wide selection of HTRF-based assays compatible with whole cells, thereby enabling functional assays run under more physiological conditions. cAMP kits are based on a competitive immunoassay using cryptate-labeled anti-cAMP antibody and d2-labeled cAMP. This assay allows the measurement of increase in intracellular cAMP upon Gs-coupled receptor activation as well as decrease in forskolin stimulated increase in cAMP upon Gi-coupled receptor activation. The IP-One assays are competitive immunoassays that use cryptate-labeled anti-IP1 monoclonal antibody and d2-labeled IP1. IP1 is a relatively stable downstream metabolite of IP3, and accumulates in cells following Gq receptor activation.

cAMP screening assay using DiscoveRX XS+ kit

UMR-106 cells were seeded in 96-well white plates at 10K cells/well in growth media. Twenty four hours after seeding, cell media was removed by gentle dumping and replaced with 30 μL of compounds diluted to 10 μM final concentration in assay buffer (Hank's balanced Salt Solution, 20mM HEPES, pH 7.4, 0.1μM IBMX). After 30 minute incubation

at room temperature, 10 μL human PTH1-34 serial diluted in assay buffer was added. Cells were incubated at 37°C for 15 minutes before 10 μL of water soluble analog of forskolin, NKH477 was added to final concentration of 10 μM followed by 60 minute incubation at room temperature. DiscoveRX cAMP XS+ kit reagents were then added following manufacture protocol. Briefly, 10 μL of antibody was added to each well followed by 40 μL of ED/Lysis buffer mix (1/5/19 for Galacon-star/Emerald/Lysis buffer and then 1:1 with ED). After 1 hour incubation, 40 μL of EA reagent was added followed by at least 1 hour incubation before the plates were read on TopCount reader. Data was analyzed using GraphPad Prism. PTH1-34 dose-response curves were fitted using non-linear curve fit (Y=Bottom + (Top-Bottom)/(1+10^((LogEC50-X)*HillSlope))). PTH1-34 EC50 values calculated in the presence of compounds were compared to that in the presence of vehicle control. The ratio of the EC50 values were calculated and presented as fold shift (EC50 compound/EC50 vehicle). The effect of compounds on PTH1-34 stimulated maximal response was also assessed and was presented percent inhibition (1-(Emax compound/Emax vehicle)).

Table 5. PTHR1 pepducin in vitro screening data (UMR cells, cAMP)

Compound	Loop	Sequence	Fold Shift of EC50	Inhibition of Maximal Response
Compound 1	i1	RRLHSTRNYIHMH	1.3	5.50%
Compound 33	i2	HSLIFMAFFSEKKYL	1.1	27.50%
Compound 2	i1	RRLHSTRNYIH	1.9	-24.90%
Compound 3	i1	LAYFRRLHSTRNY	0.7	-51.40%
Compound 4	i1	LAYFRRLHSTR	0.9	-74.20%
Compound 5	i1	LAYFRRLHSTRNYIH	1	13.00%
Compound 6	i1	YFRRLHSTRNYIH	1	10.80%
Compound 34	i2	LYLHSLIFMAFFSEKKYLWGFT	1.2	4.20%
Compound 35	i2	YLHSLIFMAFFSEKKYLWGFT	1.1	24.50%
Compound 36	i2	HSLIFMAFFSEKKYLWGFT	1.2	5.90%
Compound 44	i3	NIVRVLATKLRETNAGRSD	2	-8.30%
Compound 45	i3	VRVLATKLRETNAGRSDTR	1.4	9.10%

Compound 46	i3	VLATKLRETNAGRSDTRQQ	0.9	20.30%
Compound 47	i3	KLRETNAGRSDTRQQRKLL	2.7	8.30%
Compound 48	i3	RETNAGRSDTRQQRKLLKS	7	-3.30%
Compound 7	i1	AYFRRLHSTRNYIH	1.2	36.00%
Compound 9	i1	LAYFRRLHSTRNYI	1.1	12.70%
Compound 49	i3	TNAGRSDTRQQRKLLKSTL	1.5	26.10%
Compound 49	i3	TNAGRSDTRQQRKLLKSTL	2.3	-9.50%
Compound 51	i3	RETNAGRSDTRQQYRKLLKS	12.45	-0.015
Compound 52	i3	TNAGRSDTRQQYRKLLKSTL	11.1	1.70%
Compound 53	i3	KLRETNAGRSDTRQQY	2.1	6.40%
Compound 37	i2	LHSLIFMAFFSEKKYLWGFT	1.1	35.60%
Compound 38	i2	LYLHSLIFMAFFSEKKYLWG	1.1	21.90%
Compound 39	i2	LYLHSLIFMAFFSEKKYL	1.3	29.30%
Compound 10	i1	LAYFRRLHSTRNYIHMH	1.1	24.10%
Compound 11	i1	GSYFRRLHSTRNYIH	1.1	19.80%
Compound 12	i1	SSYFRRLHSTRNYIH	0.9	26.60%
Compound 13	i1	GGYFRRLHSTRNYIH	0.7	29.20%
Compound 14	i1	LAYFRRLHSTRN	0.7	15.20%
Compound 40	i2	LYLHSLIFMAFFSEKK	1.5	28.30%
Compound 54	i3	NIVRVLATKLRETNAGR	0.7	17.50%
Compound 55	i3	NIVRVLATKLRE	0.6	26.10%
Compound 56	i3	DTRQQYRKLLKSTL	1.9	-30.40%
Compound 57	i3	RQQYRKLLKSTL	15.6	-38.80%
Compound 58	i3	RETNAGRSDTRQQYRKLLFS	17.74	0.0367
Compound 59	i3	RETNAGRSDTRQQYRKLL	5.2	3.00%
Compound 60	i3	RETNAGRSDTRQQYRK	3.3	4.30%
Compound 61	i3	RETNAGRSDTRQQYRF	0.7	-47.60%
Compound 62	i3	RETNAGRSDTRQQY	1.1	-5.40%
Compound 63	i3	RETNAGRSDTRQ	1.1	1.20%
Compound 64	i3	TNAGRSDTRQQYRKLLKS	7	-0.10%

Compound 65	i3	AGRSDTRQQYRKLLKS	14.65	-0.359
Compound 66	i3	RSDTRQQYRKLLKS	16.4	-39.00%
Compound 67	i3	DTRQQYRKLLKS	6	6.90%
Compound 68	i3	TNAGRSDTRQQYRKLL	12.36	-0.0684
Compound 69	i3	TNAGRSDTRQQYRK	5.2	2.50%
Compound 70	i3	TNAGRSDTRQQYRF	2	15.10%
Compound 71	i3	SGRVLATKLR	0.8	13.50%
Compound 72	i3	SGRVLATKLRET	0.7	22.30%
Compound 73	i3	SGRVLATKLRETNA	0.8	32.30%
Compound 74	i3	SGRVLATKLRETNAGR	1.9	22.20%
Compound 75	i3	RVLATKLRETNAGR	1.1	12.60%
Compound 15	i1	RRLHSTRNYIHM	1.7	1.50%
Compound 16	i1	RRLHSTRNYIHMHL	2.45	0.0965
Compound 17	i1	SGRRLHSTRNYIHMH	2.8	39.40%
Compound 41	i2	GSEKKYLWGFTVF	0.9	17.70%
Compound 42	i2	GSEKKYLWGFT	1.3	40.80%
Compound 43	i2	GSEKKYLWG	1.4	5.70%
Compound 96	i4	EIKKSWSRWTLALDFKRKAR	1.1	7.00%
Compound 97	i4	KKSWSRWTLALDFKRKAR	4.7	37.20%
Compound 98	i4	NGEVQAEIKKSWSRWTLA	1	0.50%
Compound 99	i4	NGEVQAEIKKSW	1	17.50%
Compound 100	i4	NGEVQAEIKKSWSR	1.8	26.80%
Compound 101	i4	NGEVQAEIKKSWSRWT	1.2	25.30%
Compound 102	i4	SWSRWTLALDFKRKAR	1	-25.40%
Compound 103	i4	WTLALDFKRKAR	1.2	-7.30%
Compound 104	i4	SRWTLALDFKRKAR	1.2	26.80%
Compound 105	i4	NGEVQAEIKKSWSRWTLALD	1.1	34.00%
Compound 76	i3	TNAGASDTRQQYRKLL	16.9	2.40%
Compound 77	i3	TNAGRADTRQQYRKLL	6.9	9.90%
Compound 78	i3	TNAGRSDTAQQYRKLL	12.7	-29.20%

Compound 79	i3	TNAGRSDARQQYRKLL	7.2	1.20%
Compound 80	i3	TNAARSDTRQQYRKLL	2.3	0.80%
Compound 81	i3	TNAGRSDTRAQYRKLL	13.8	-20.80%
Compound 82	i3	TNAGRSATRQQYRKLL	23.4	-21.60%
Compound 83	i3	TNAGRSDTRQQYRKLLF	7	-5.80%
Compound 84	i3	TNAGRSDTRQQYRKLLK	9.1	-25.20%
Compound 85	i3	TNAGRSDTRQQYRKLLFS	6.9	19.50%
Compound 86	i3	TNAGRSDTRQQYRKLLFA	3.6	5.20%
Compound 87	i3	TNAGRSDTRQQYRKLLA	4.1	8.00%
Compound 88	i3	TNAGRSDTRQQYRKLA	2.8	-25.00%
Compound 89	i3	TNAGRSDTRQQYRKAL	2.3	-20.40%
Compound 90	i3	TNAGRSDTRQQYRALL	2	21.00%
Compound 91	i3	TNAGRSDTRQQYAKLL	2	9.80%
Compound 92	i3	TNAGRSDTRQQARKLL	5	8.30%
Compound 93	i3	TNAGRSDTRQAYRKLL	13.4	-67.00%
Compound 94	i3	AGRSDTRQQYRKLLFA	18.7	-24.40%
Compound 95	i3	AGRSDTRQQYRKLLFS	19	-5.70%

AlphaScreen cellular kinase assays

GPCR activation results in modulation of downstream kinase systems and is often used to probe GPCR function and regulation. TGR Bioscience and PerkinElmer have developed Surefire cellular kinase assay kits that are HTS capable and useful in screening kinase regulation. Such kits enable the monitoring of Gi regulated downstream kinases like ERK1/2. The assay allows the measurement of increases in ERK1/2 kinase phosphorylation upon Gi coupled receptor activation and this signal in turn can be used to assay Gi coupled receptor modulator. Similar kits are also availibel to assay other pathway dependent siganlling kinases such as MAP and BAD.

IN VIVO ASSAYS

The G-protein coupled receptor PTHR1 is important in several therapeutic areas including osteoporosis; humoral hypercalcemia of malignancy; osteolytic and osteoblastic metastasis to bone; primary and secondary hyperparathyroidism associated increase in bone absorption; vascular calcification; psychiatric disorders and cognitive disorders; dermatological disorders and excess hair growth. PTHR1 receptor compounds of the present invention (agonists, antagonists, modulators) can be assessed using suitable *in vivo* models. Such *in vivo* models include PTH induced rapid response in kidney by measuring urinary excretion of phosphate and cyclic AMP in thyroparathyroidectomized rats. A more relavant in bone and calcemic effects of PTH can be assessed using a similar model. Uremic rat model (5/6 nephrectomy) can be used as a disease model for secondary hyperparathyroidism.

The thyroparathyroidectomized rat model is a useful acute model in assessing antagonist actions at PTHR1 receptor compounds of the invention. The measurements can be rapid increase in urinary excretion of phosphate and cyclic AMP. The more clinical relavant properties of a PTHR1 antagonist should include the bone and calcemic effects of PTH. Rats that are on calcium free diet for a week prior to experiments and coadministered a small amount of calcium with PTH provide a sensitive and reliable system to assess PTHR1 antagonist action *in vivo* (Proc. Natl. Acad. Sci. USA 1986: Vol. 83, pp. 7557-7560).

Renal insufficient rat models can be established by surgically remove one kidney followed by ligation of both poles of the other. This has been used as a model system for secondary hyperparathryroidism. Bone resorption and tissue calcification can then be assessed.

An animal model of humoral hypercalcemia of malignancy can be established by serially carrying a human squamous cell lung cancer in athymic mice, which leads to hypercalcemia (Endocrinonogy 1994:vol 134 p2184-2188).

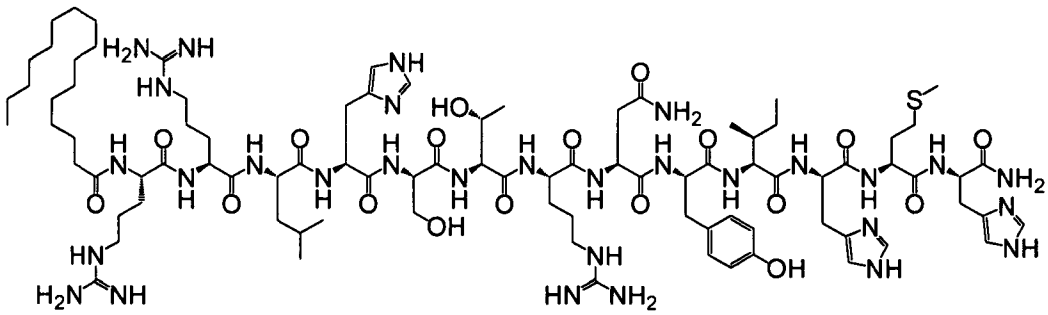
The teachings of all patents, published applications and references cited herein are incorporated by reference in their entirety.

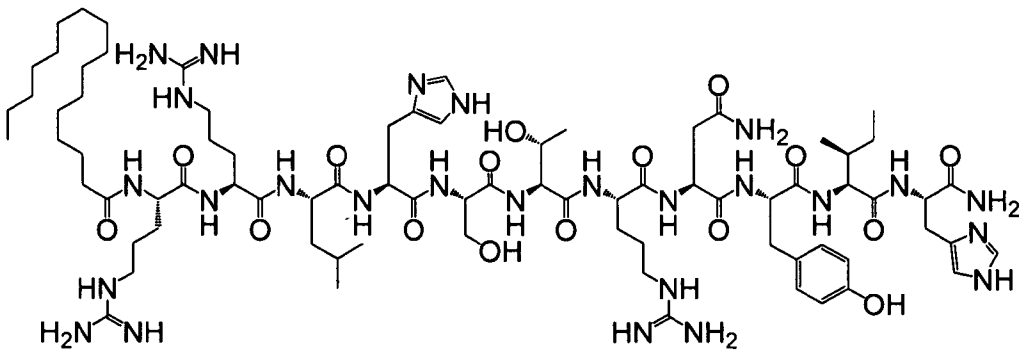
While this invention has been particularly shown and described with references to example embodiments thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the scope of the invention encompassed by the appended claims.

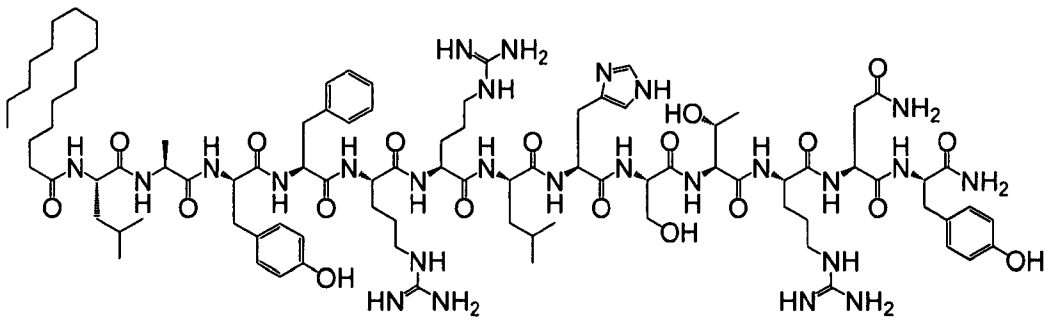
CLAIMS

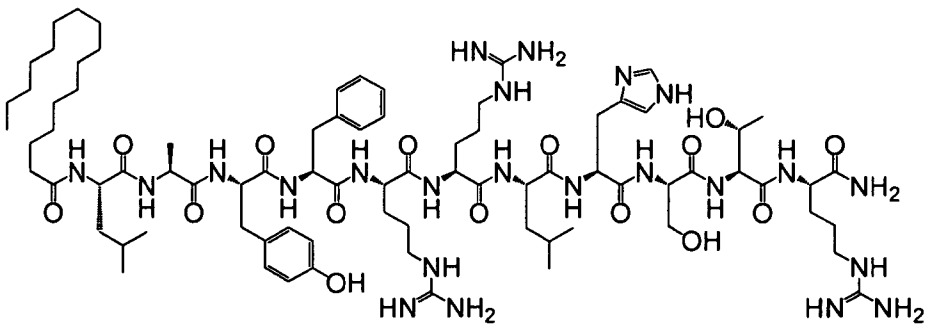
What is claimed is:

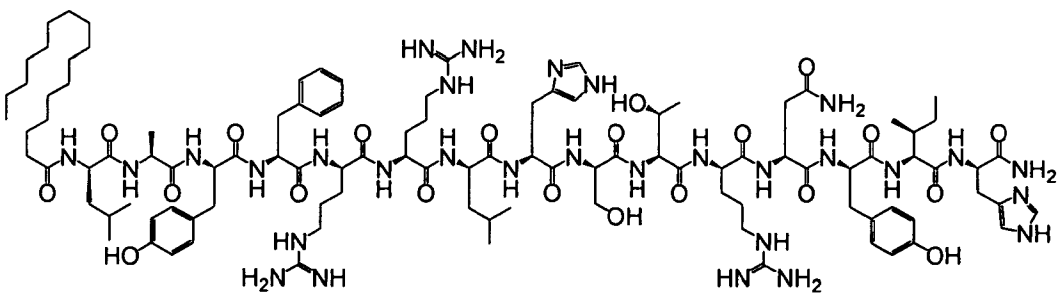
1. A compound selected from the following group:

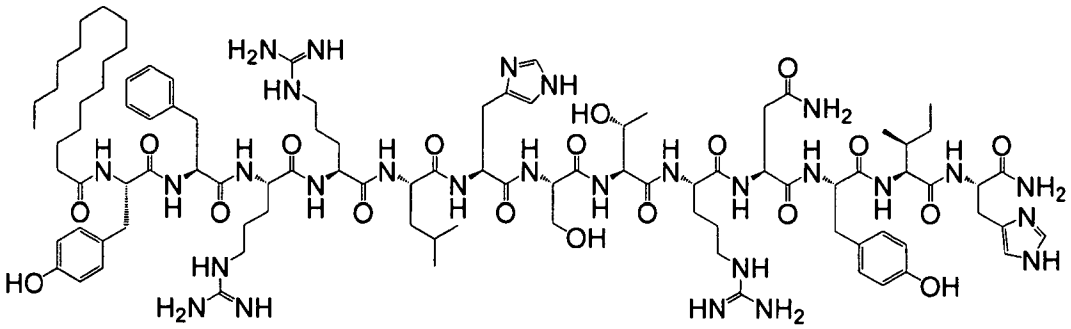


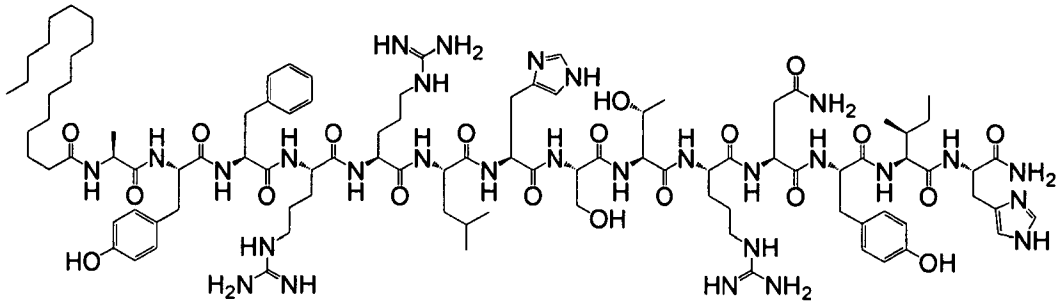


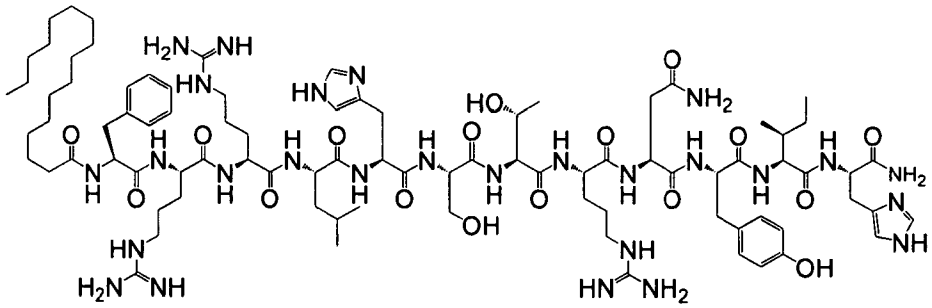


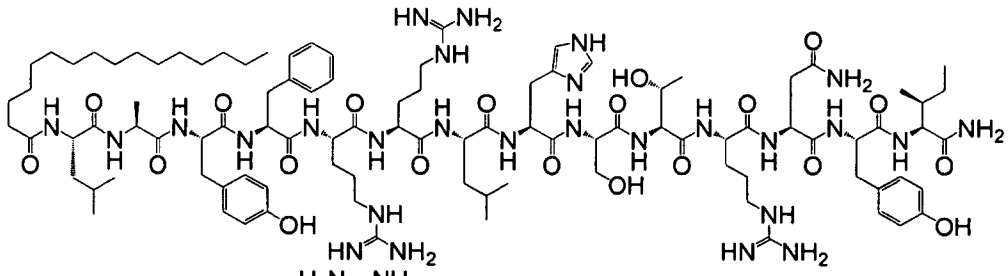


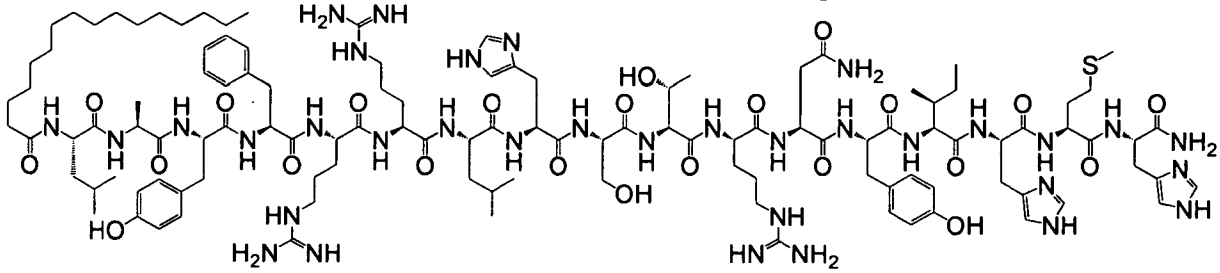


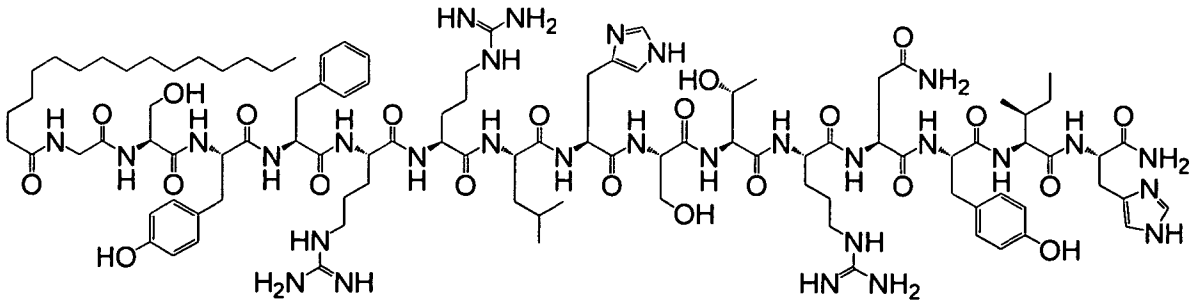


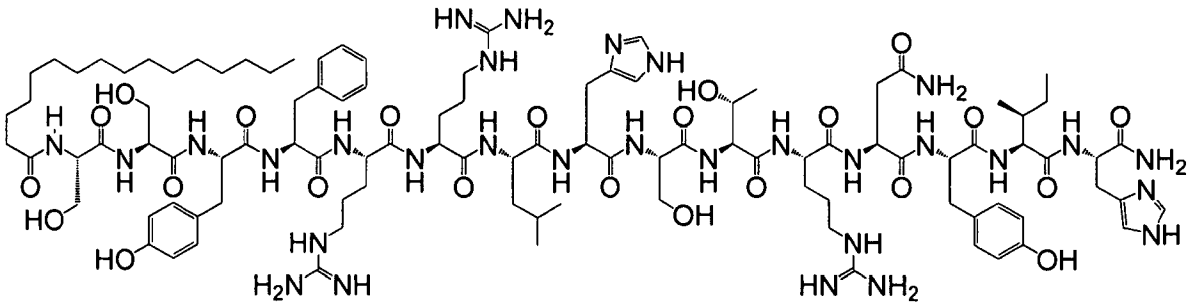


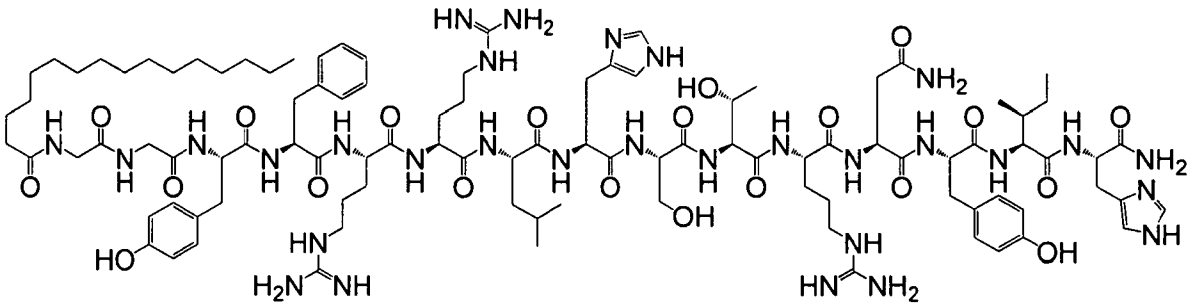


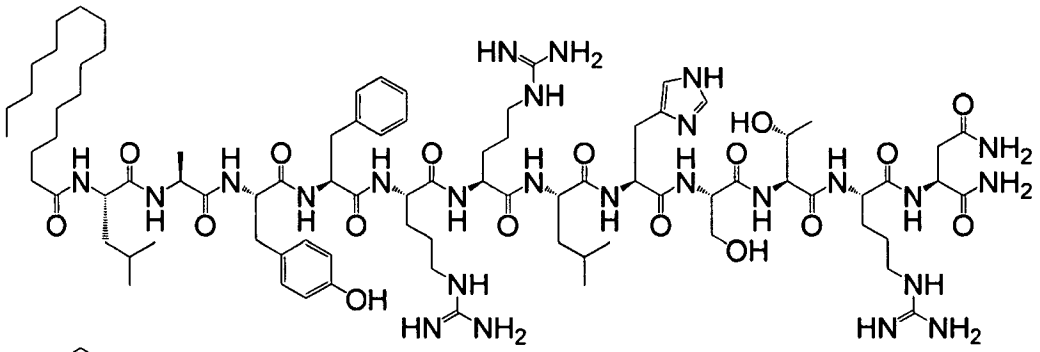


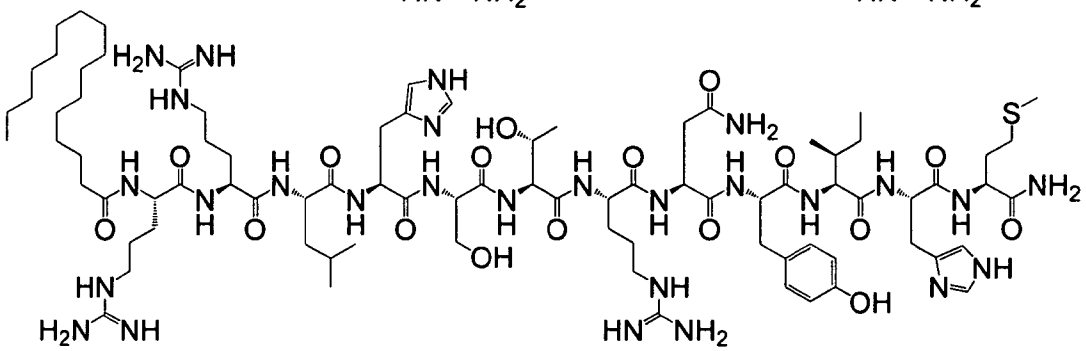


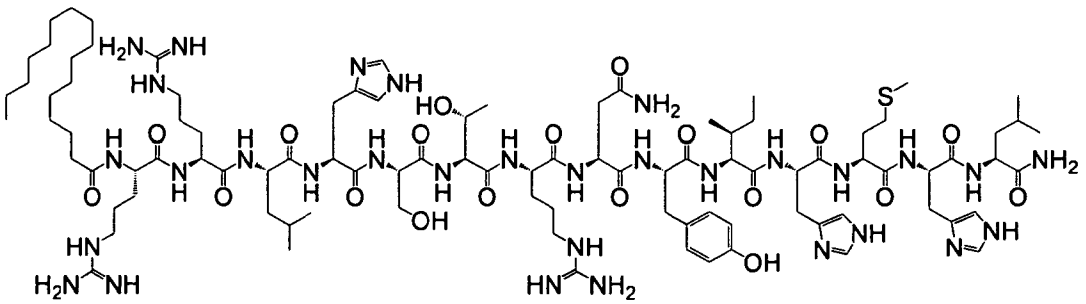


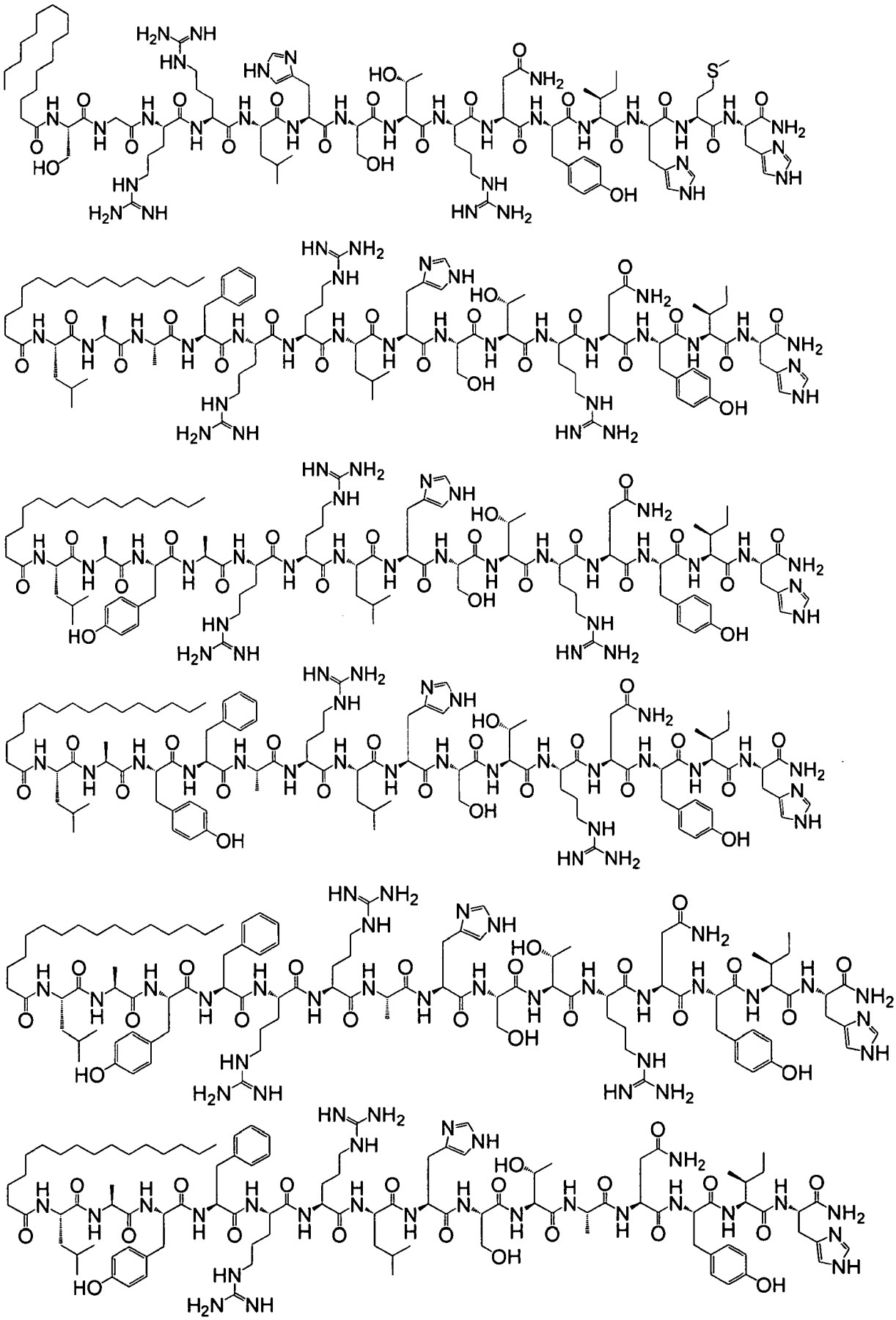


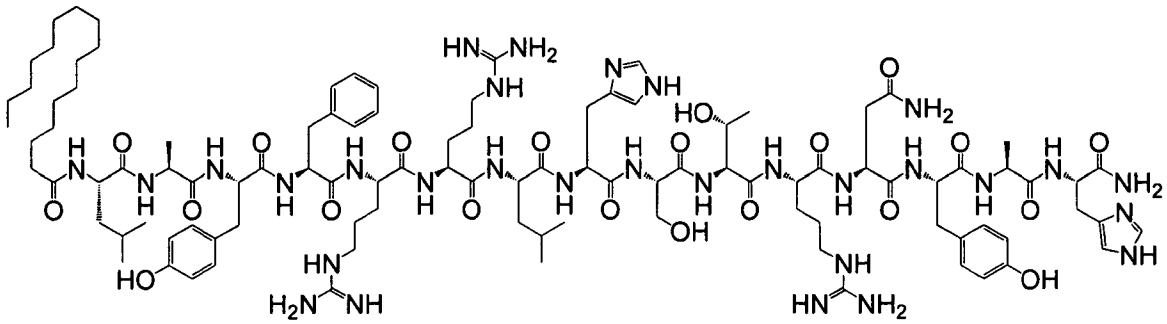


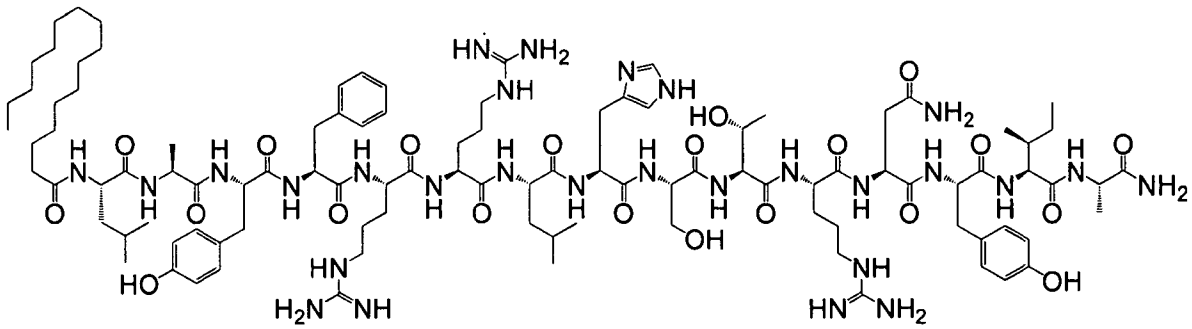


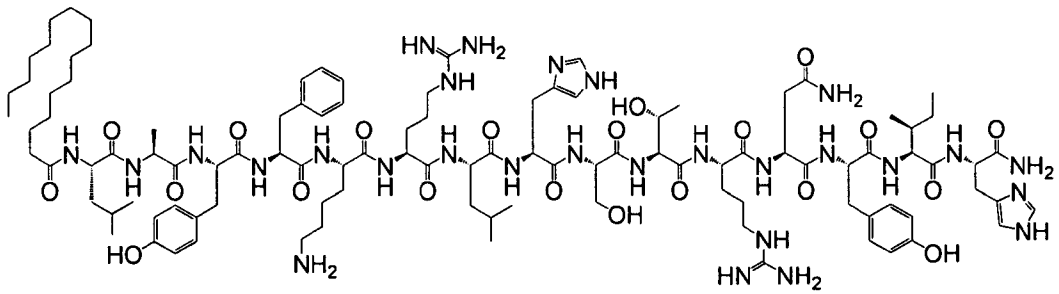


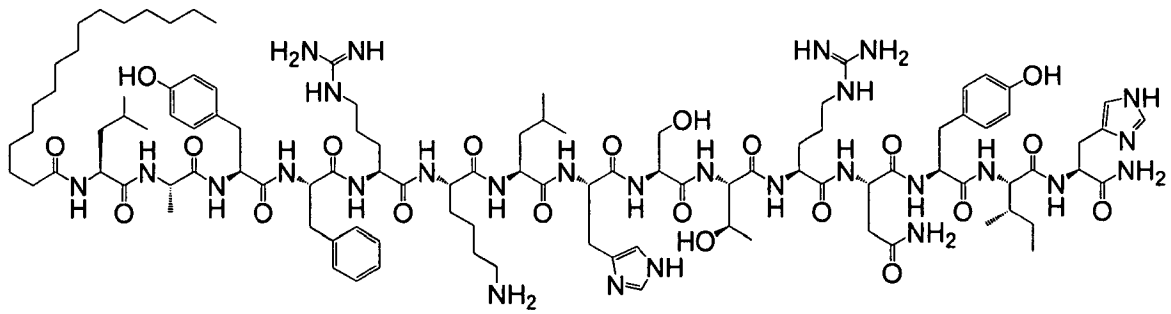


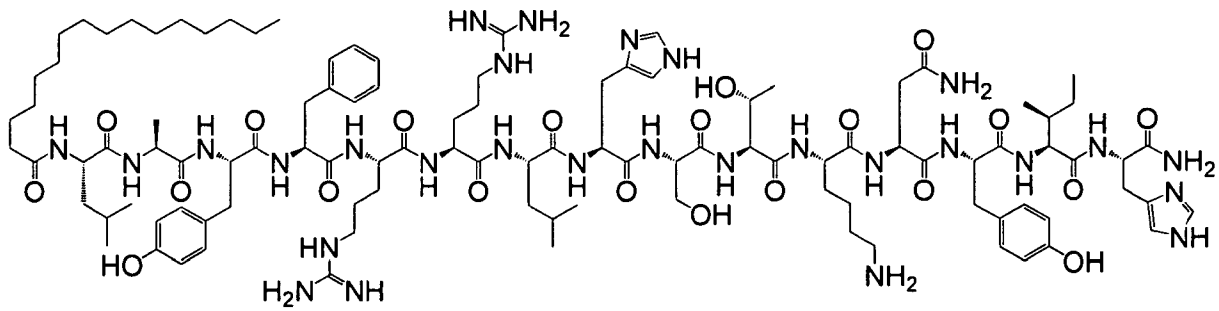


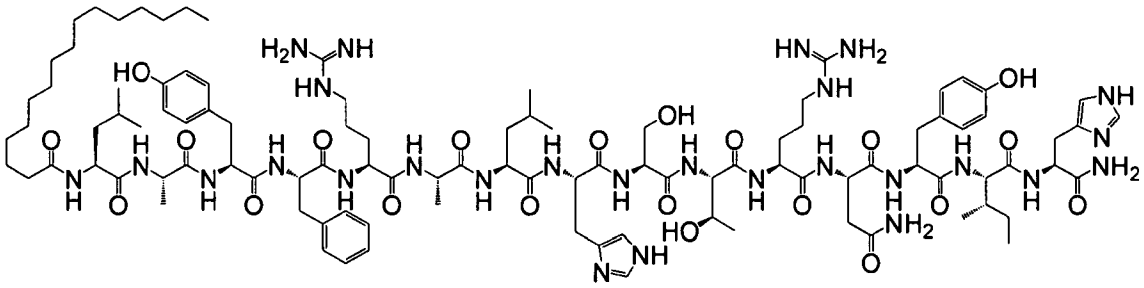


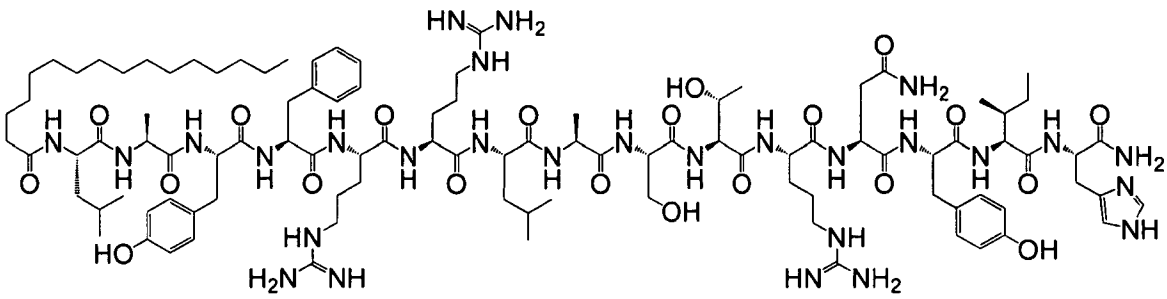


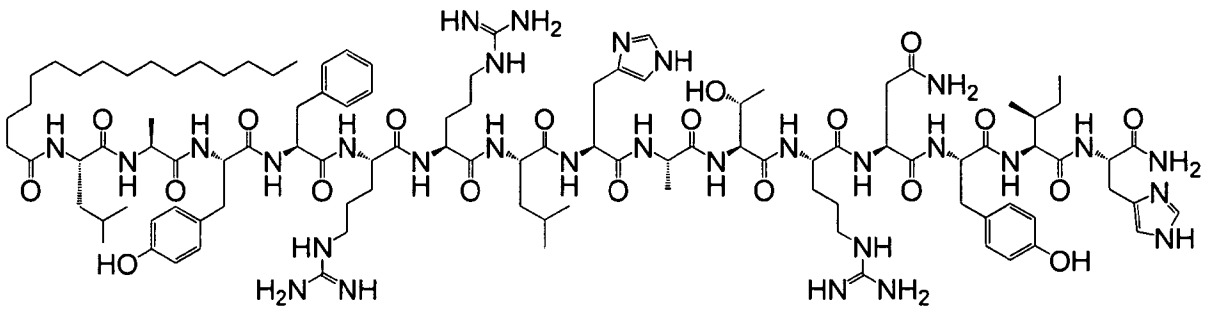


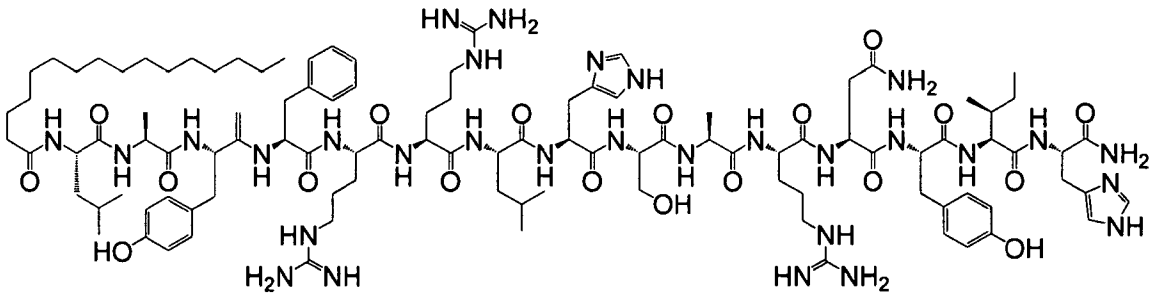












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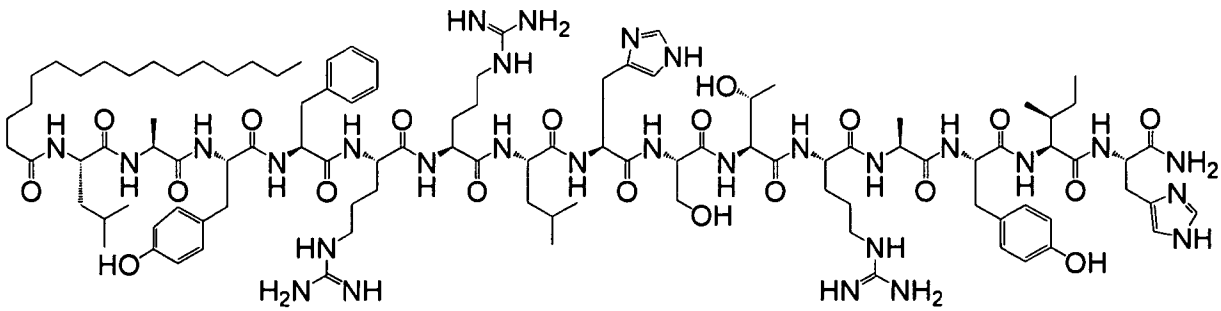
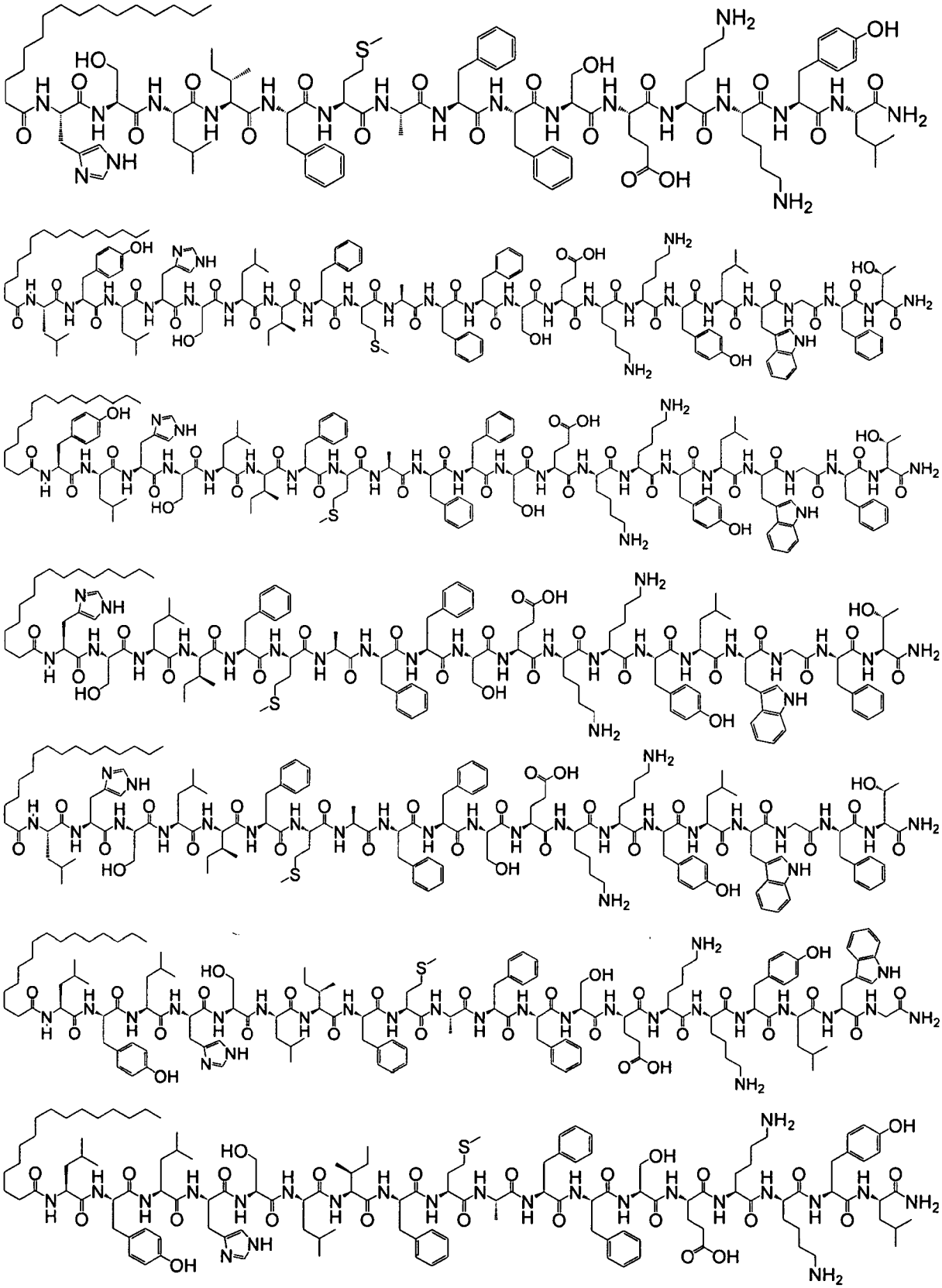
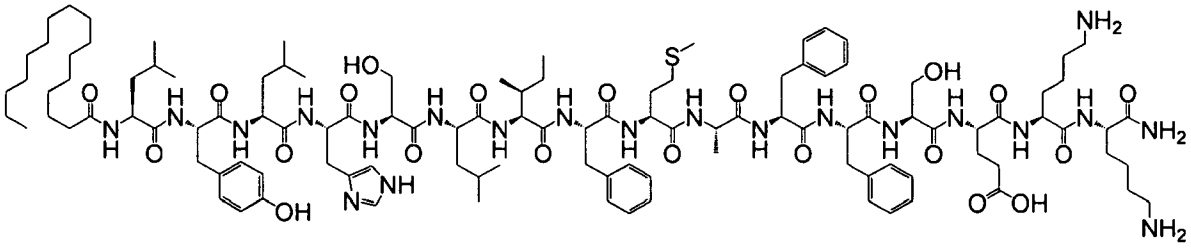
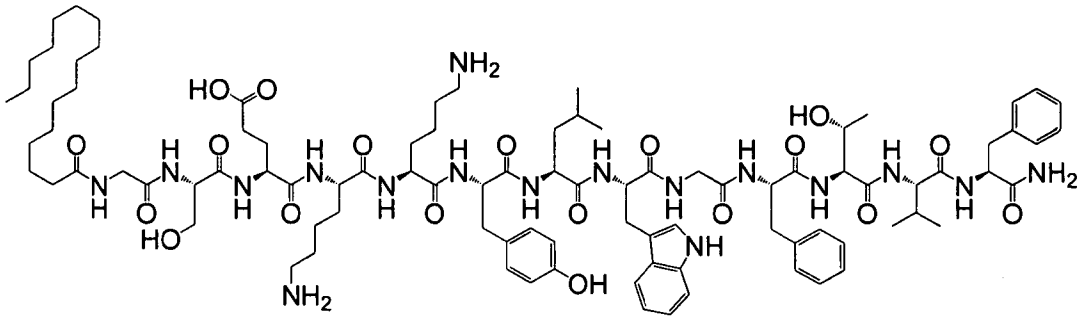


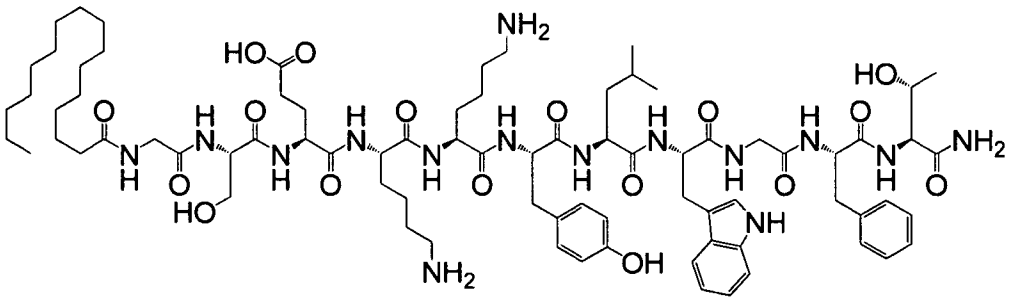
or a pharmaceutically acceptable salt thereof.

2. A compound selected from the following group:









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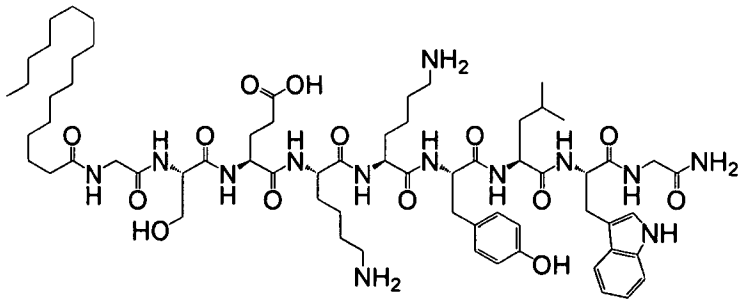
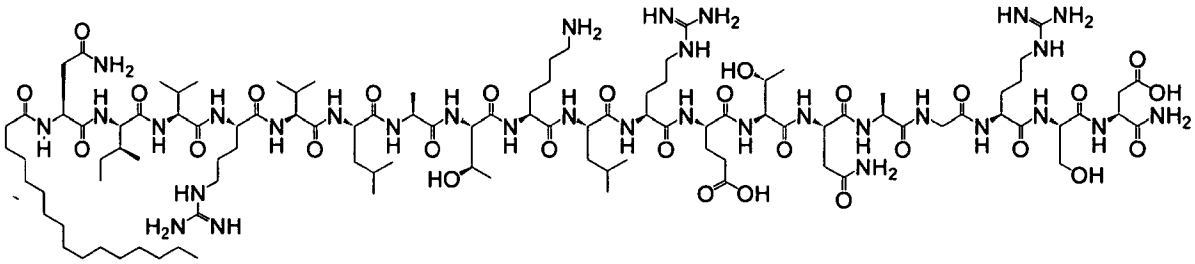
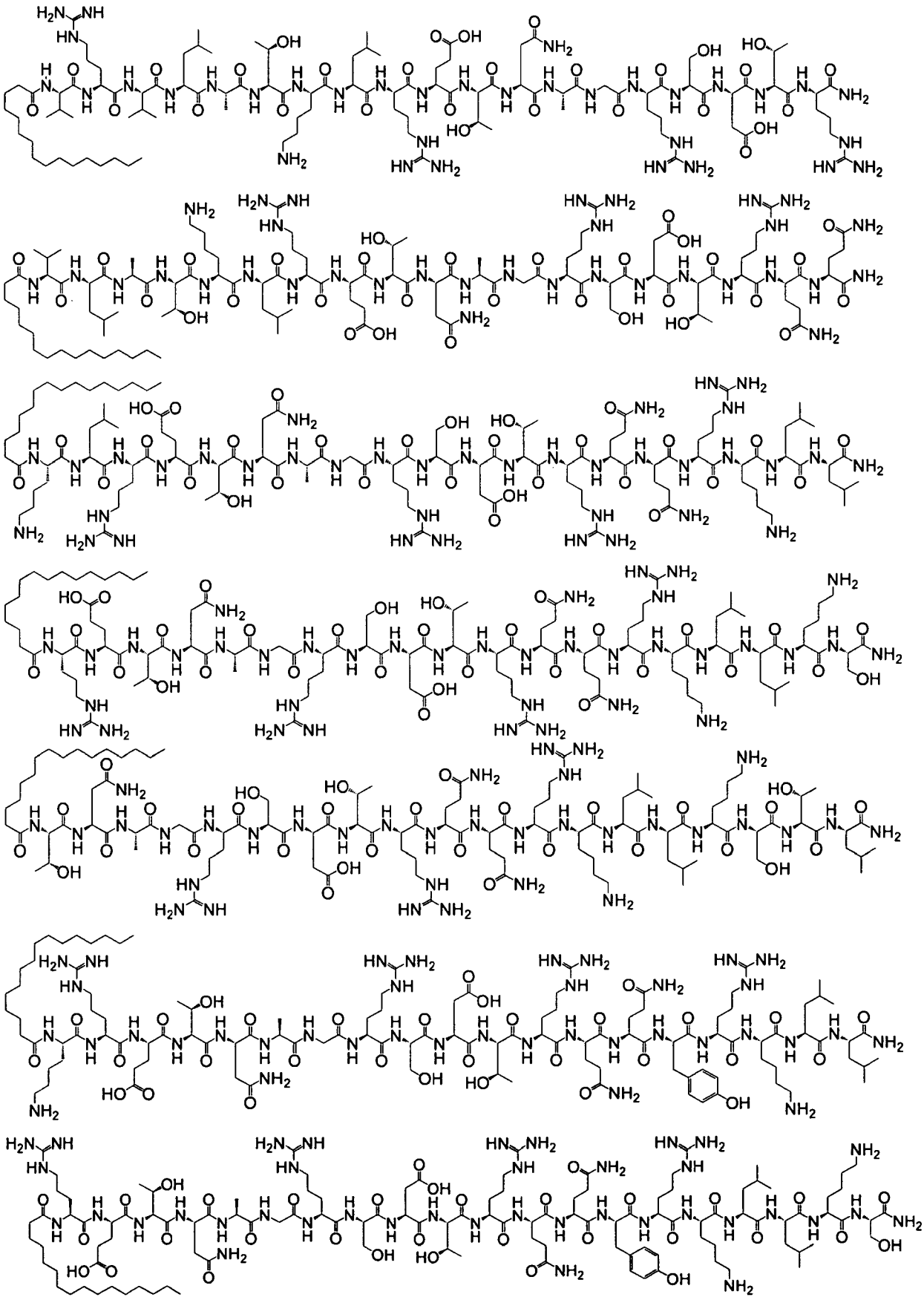
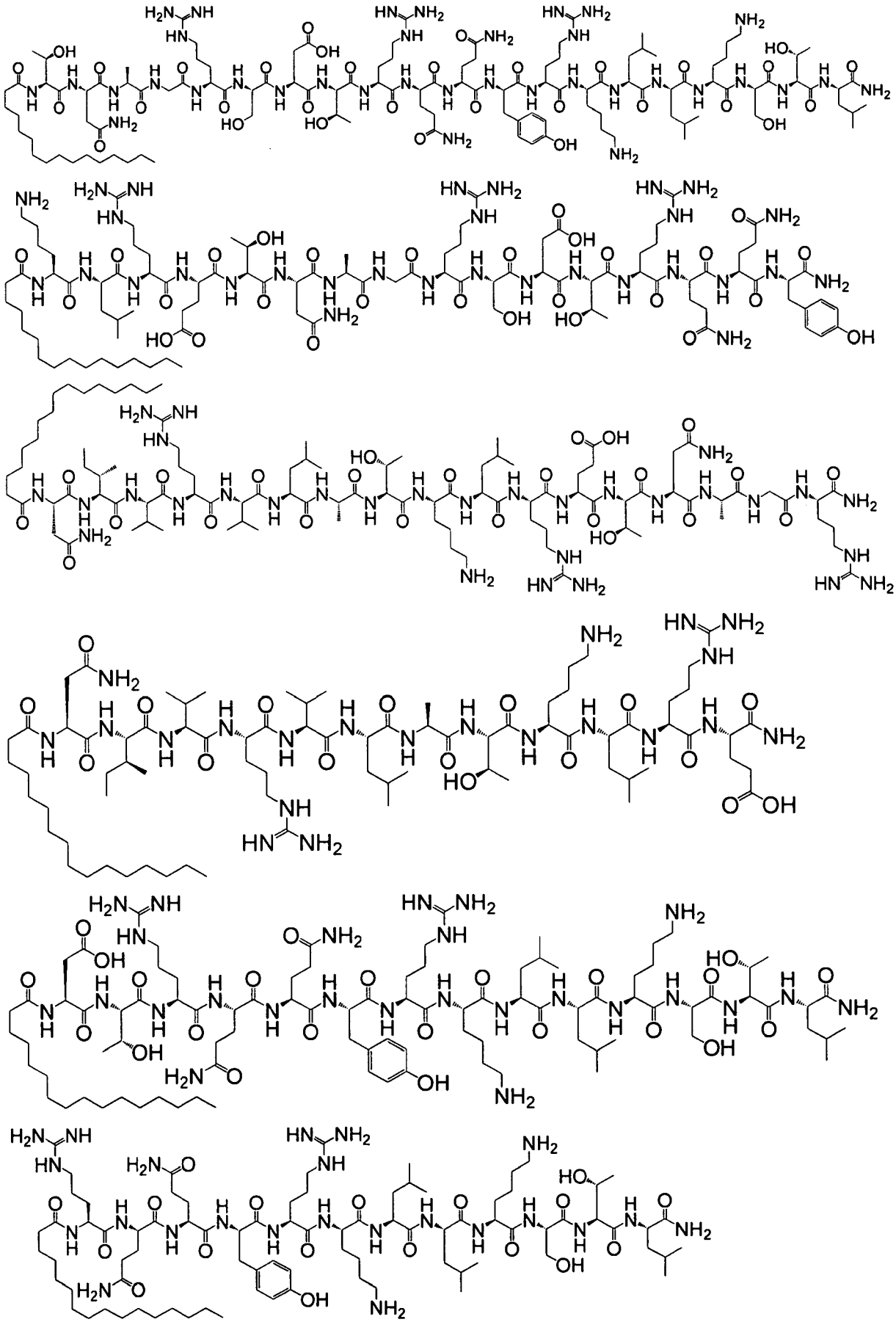


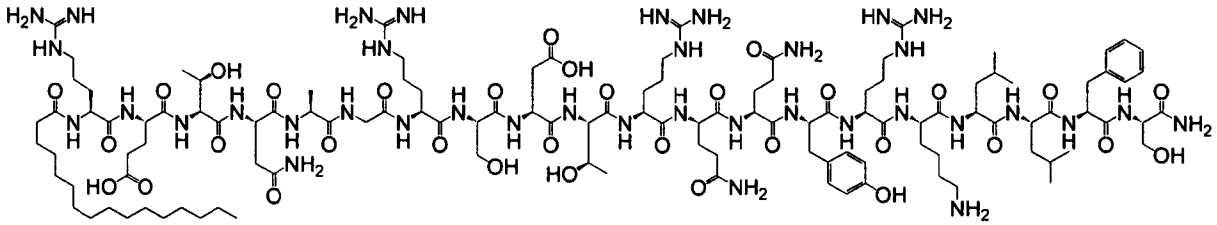
or pharmaceutically acceptable salt thereof.

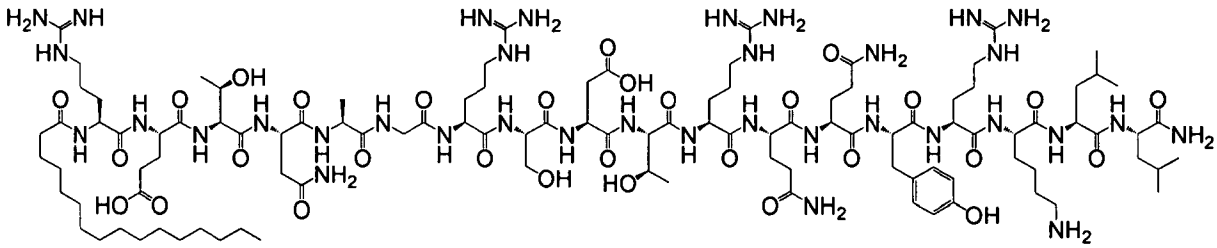
3. A compound selected from the following group:

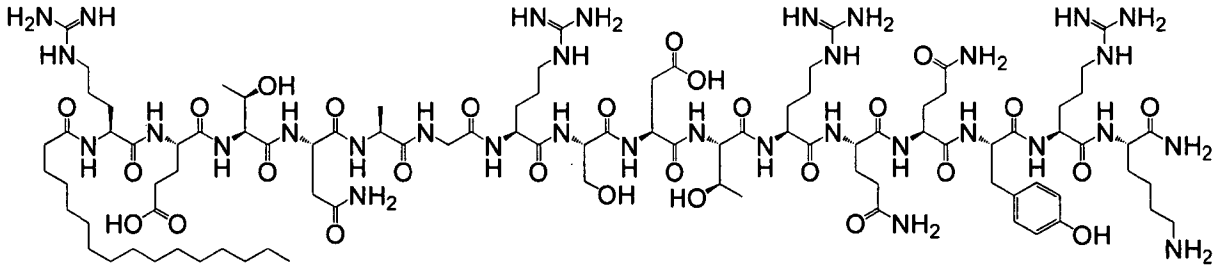


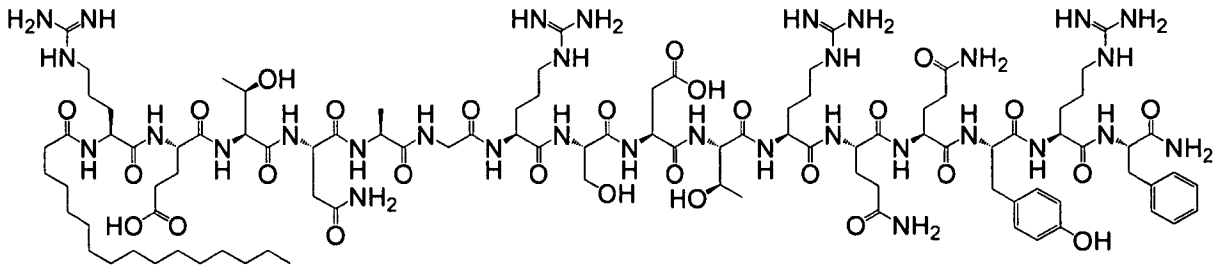


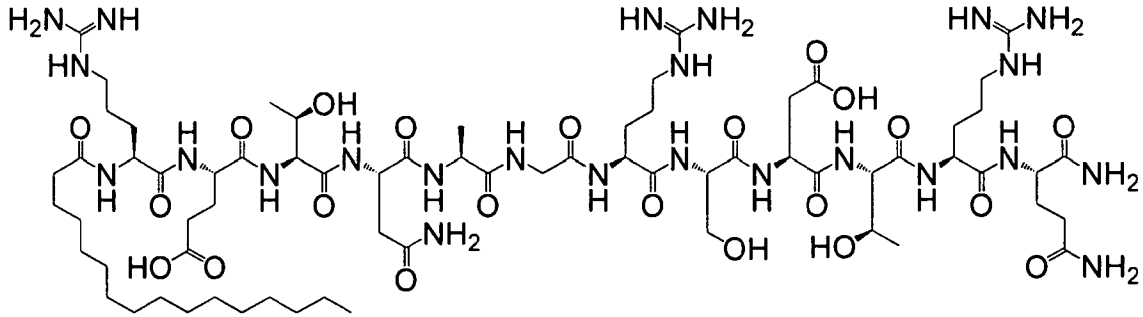


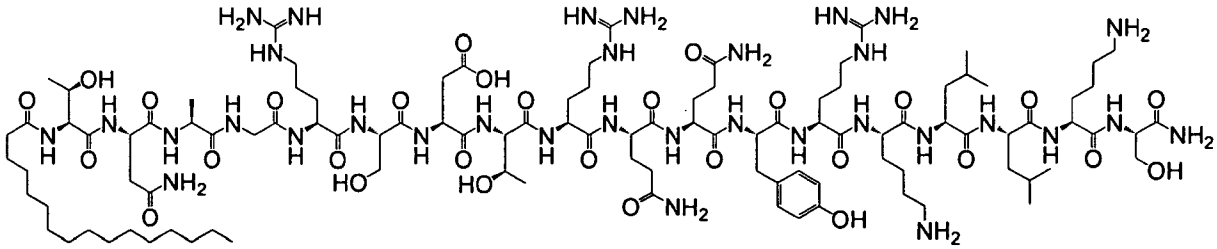


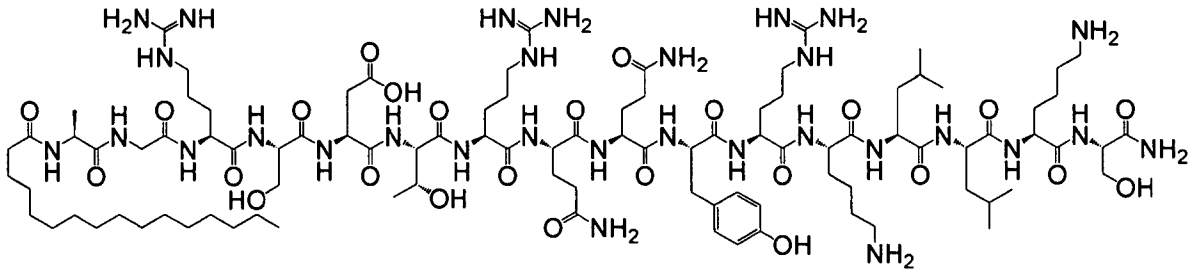


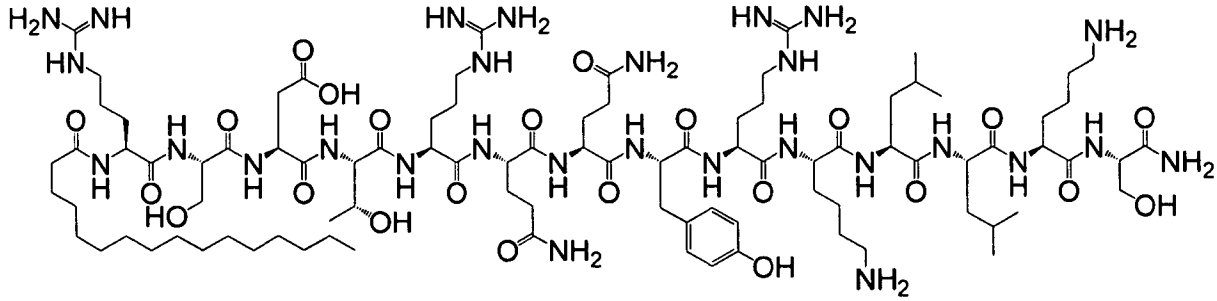


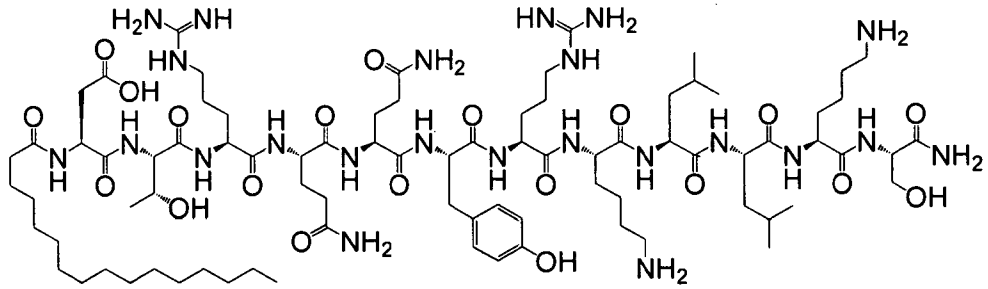


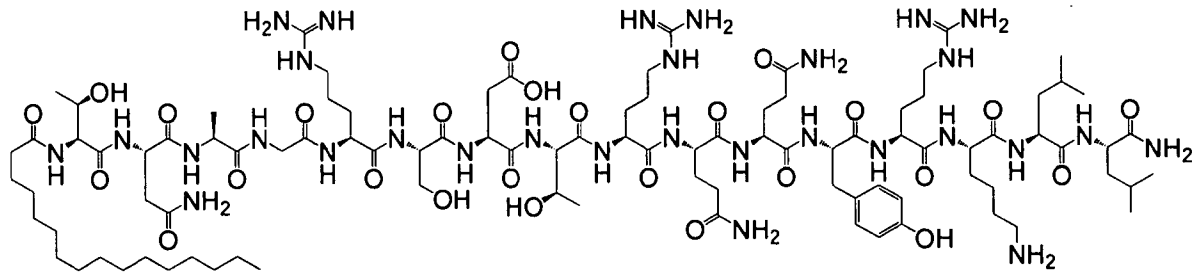


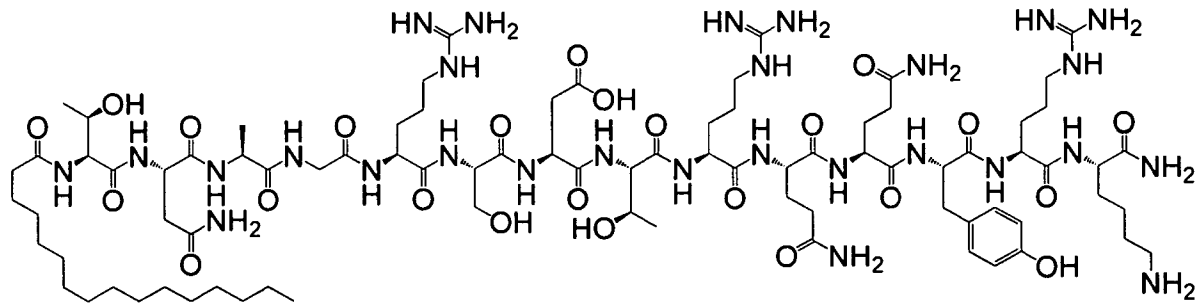


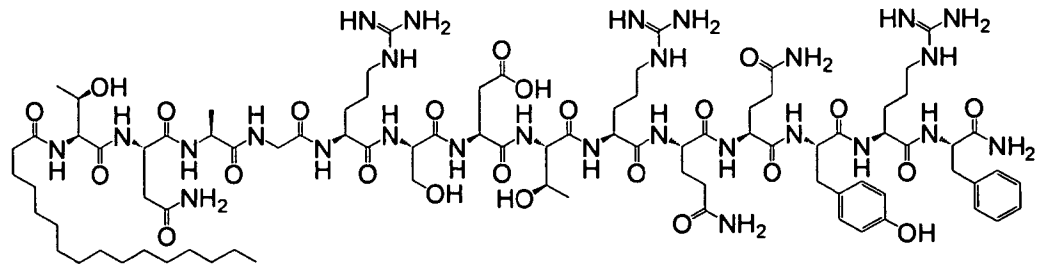


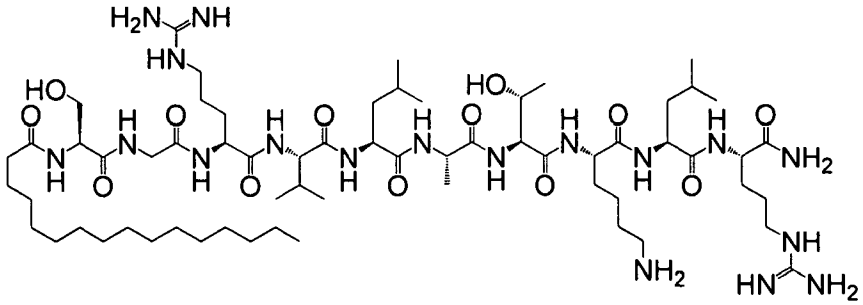


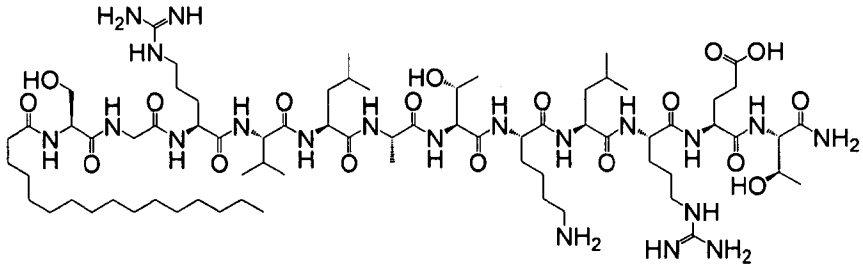


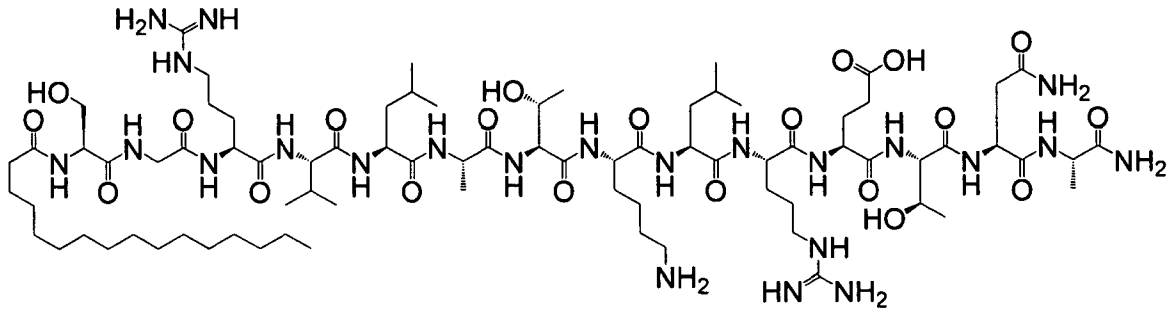


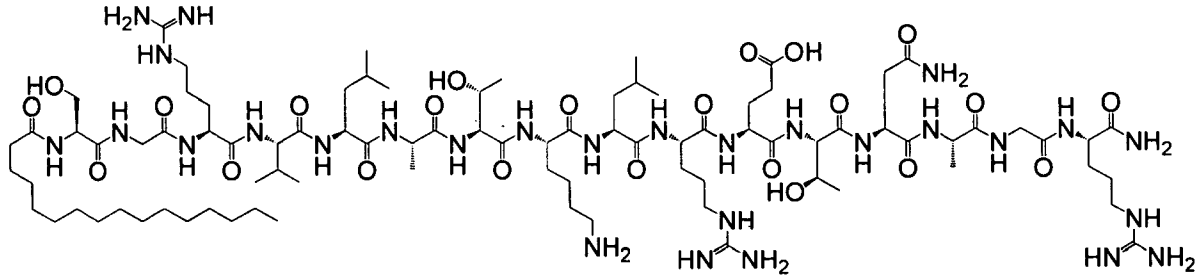


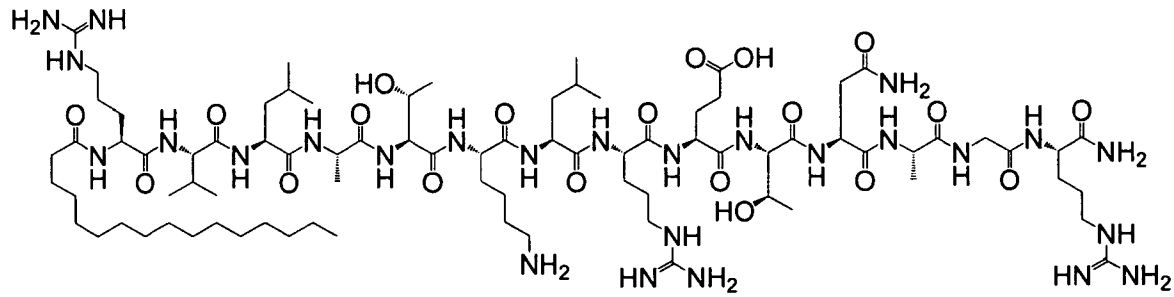


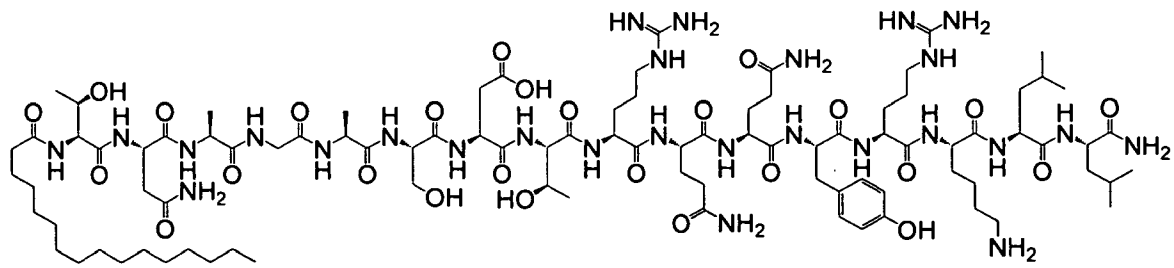


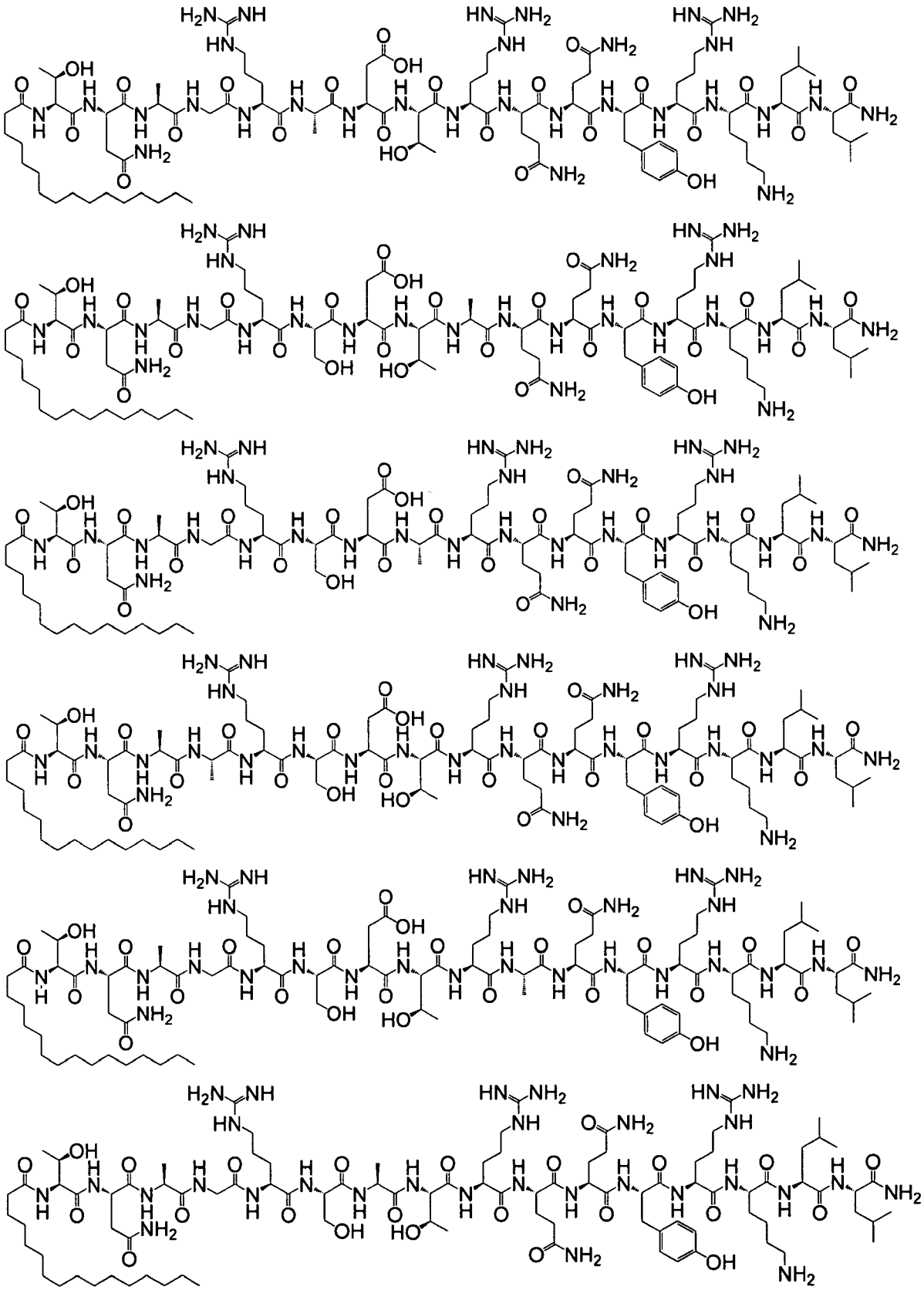


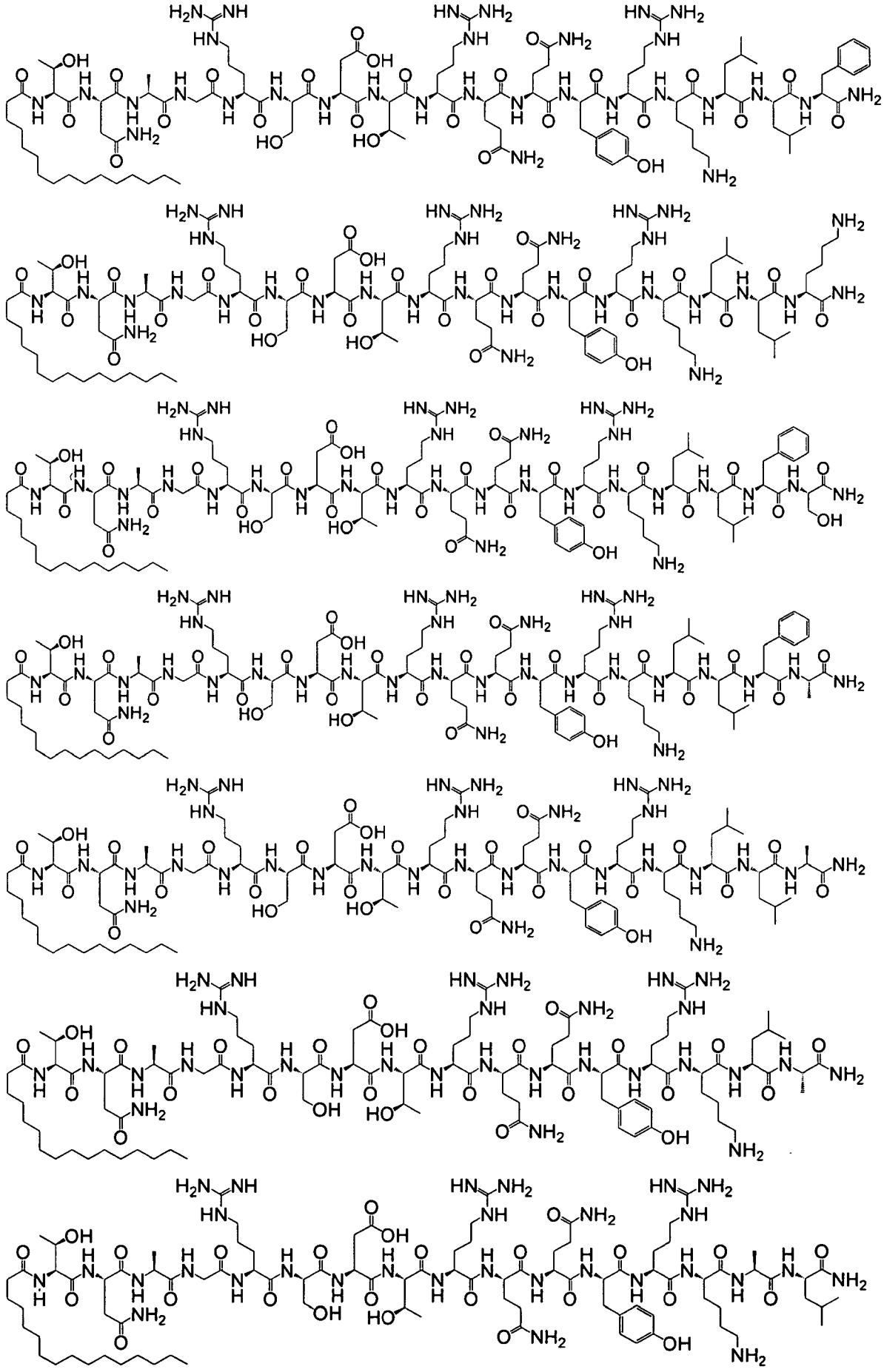


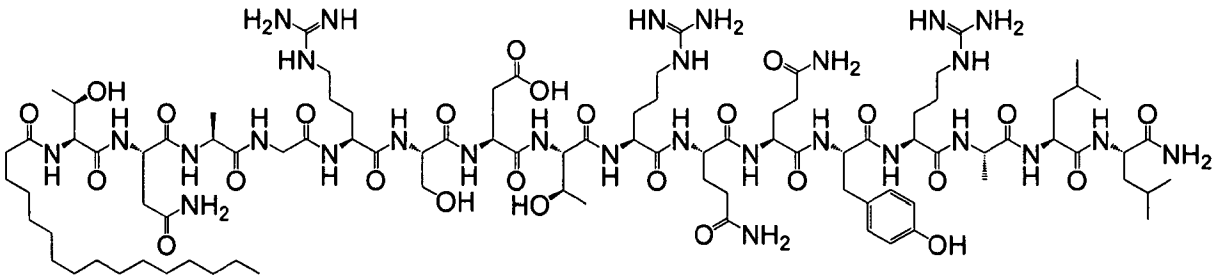


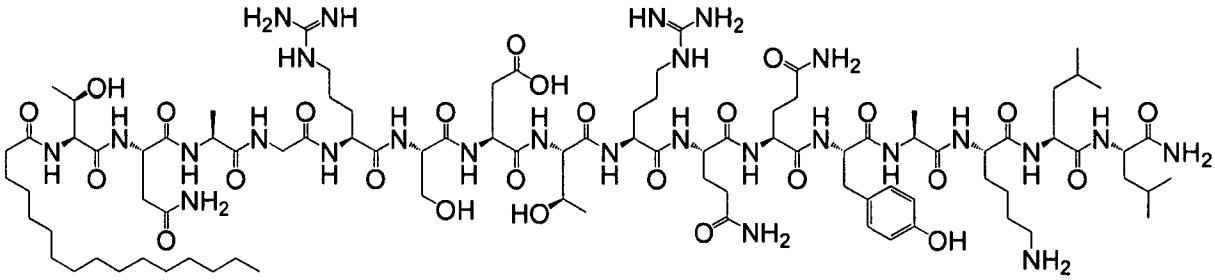


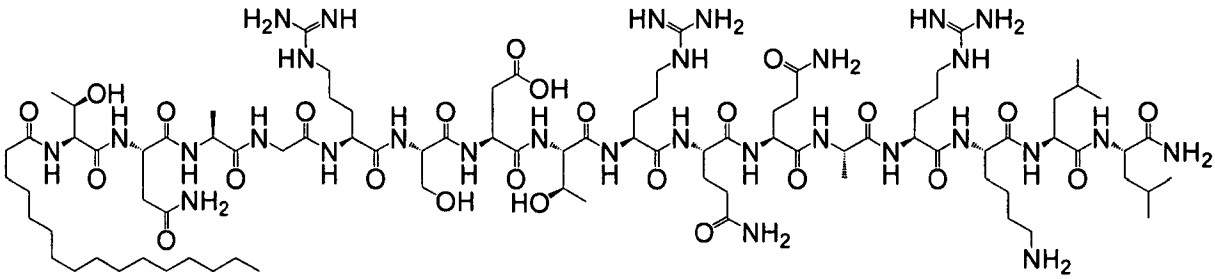


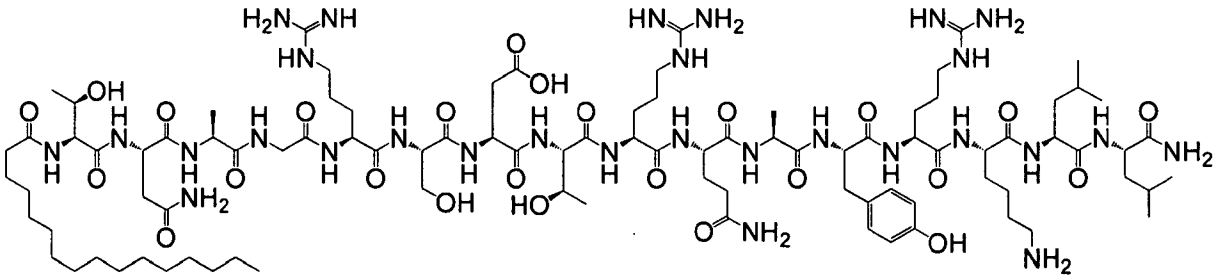


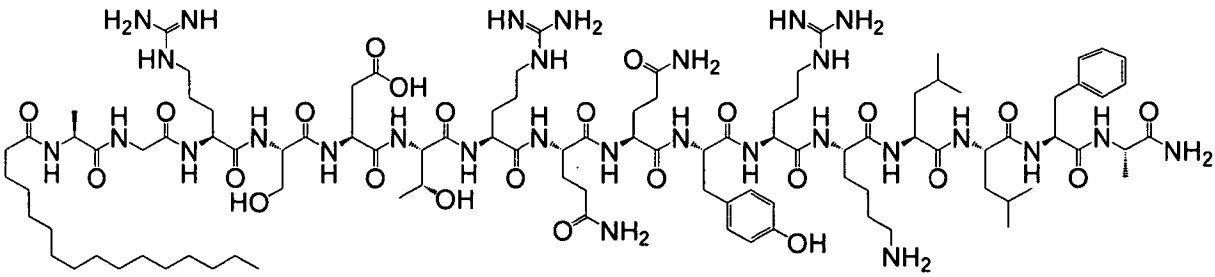












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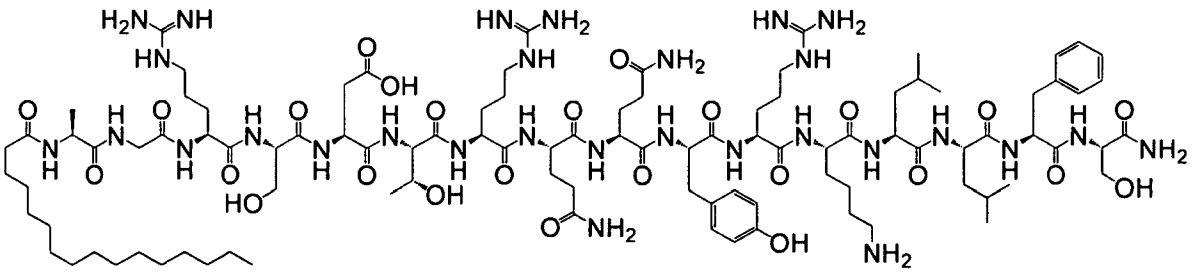
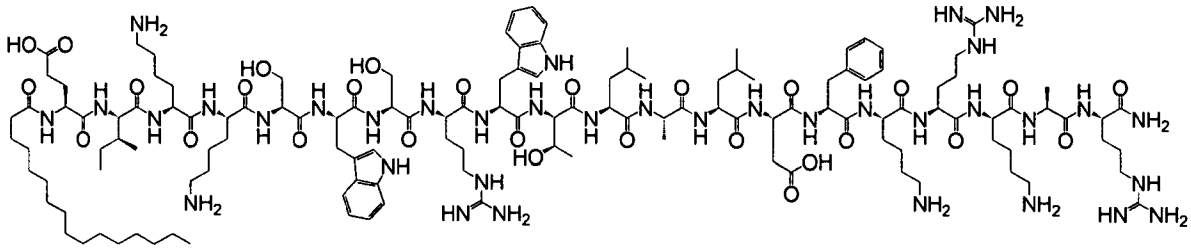
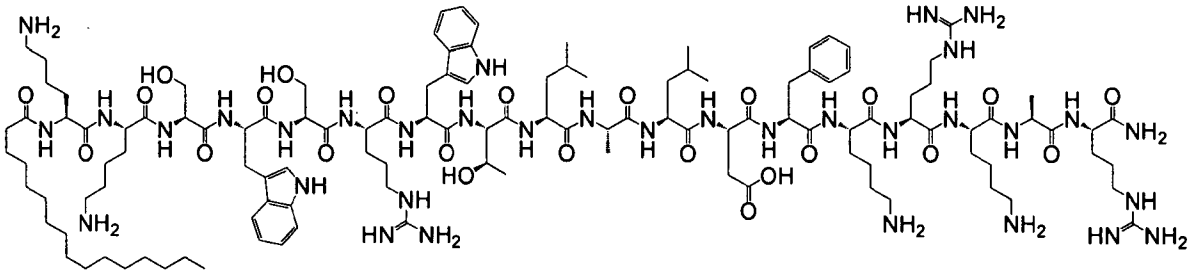
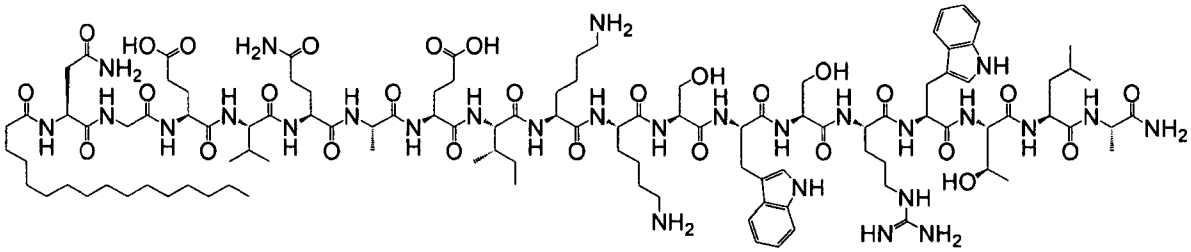


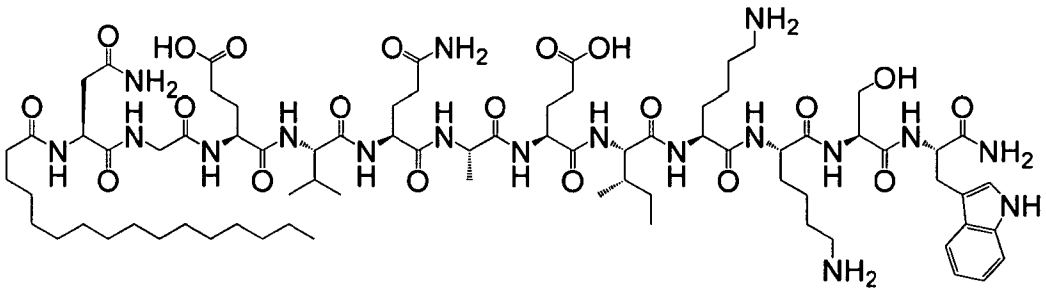
or a pharmaceutically acceptable salt thereof.

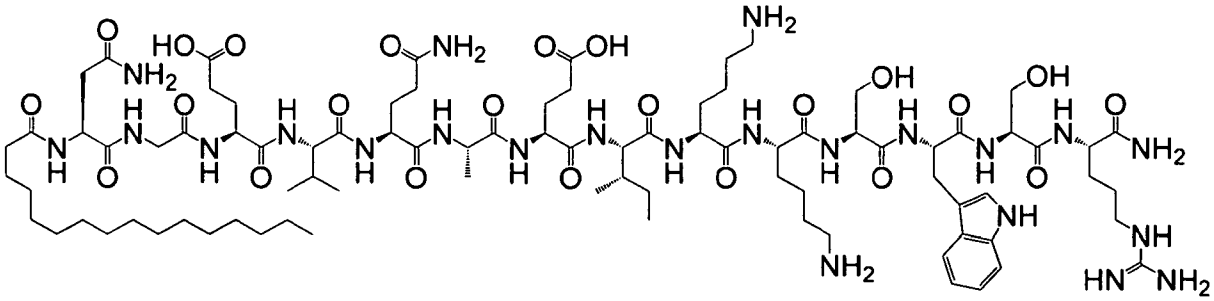
4. A compound selected from the following group:

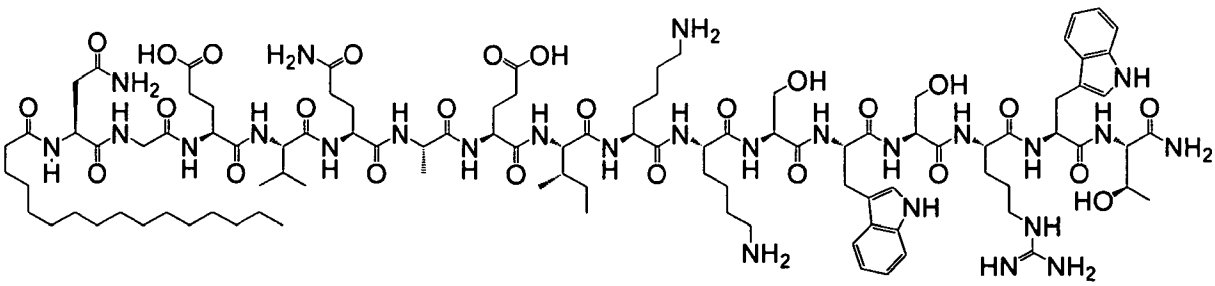


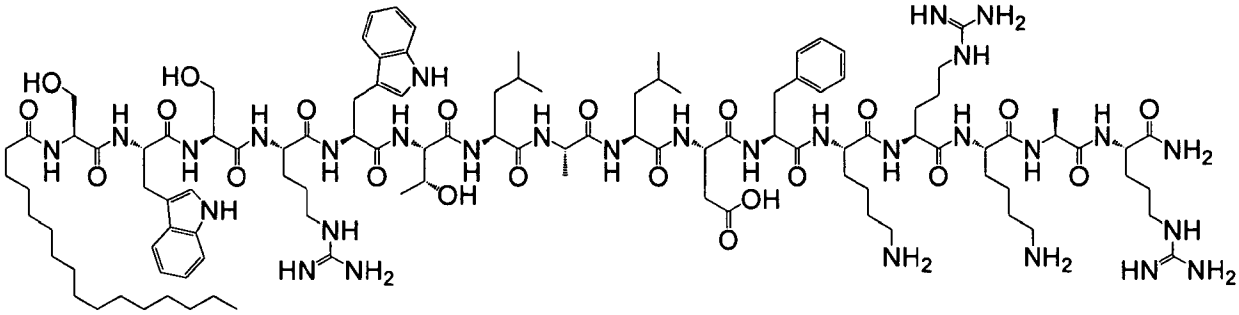


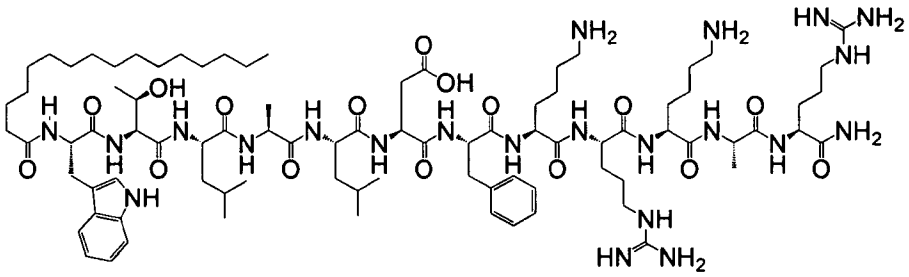


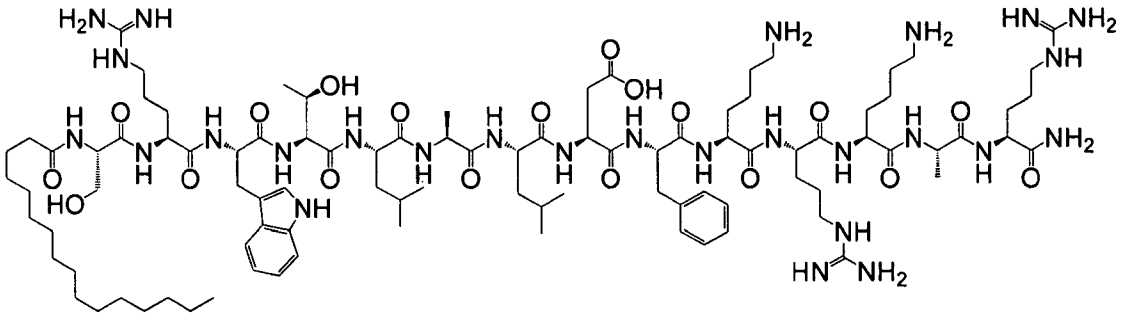












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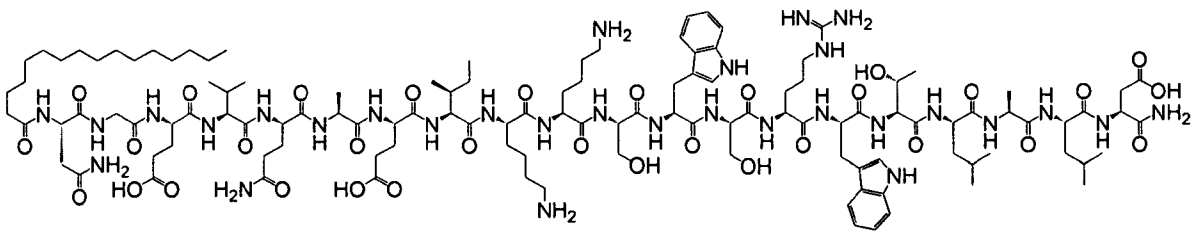


or pharmaceutically acceptable salt thereof.

5. A compound represented by Formula I:

T-L-P,

or pharmaceutically acceptable salts thereof, wherein:

P is a peptide sequence selected from: SEQ ID NOS: 2-33; SEQ ID NOS: 35-44; SEQ ID NOS: 46-99; and SEQ ID NOS: 101-110;

L is a linking moiety represented by C(O) and bonded to P at an N terminal nitrogen of an N-terminal amino-acid residue;

and T is a lipophilic tether moiety bonded to L, where the C-terminal amino acid residue of P is optionally functionalized.

6. The compound of Claim 5, wherein P is selected from SEQ ID NOS: 2-33.

7. The compound of Claim 5, wherein P is selected from SEQ ID NOS: 35-44.

8. The compound of Claim 5, wherein P is selected from SEQ ID NOS: 46-99.

9. The compound of Claim 1, wherein P is selected from SEQ ID NOS: 101-110.

10. The compound of any one of Claims 5-9, wherein T is an optionally substituted (C_6-C_{30})alkyl, (C_6-C_{30})alkenyl, (C_6-C_{30})alkynyl, wherein 0-3 carbon atoms are replaced with oxygen, sulfur, nitrogen or a combination thereof.

11. The compound of Claim 10, wherein T is selected from the group consisting of: $CH_3(CH_2)_{16}$, $CH_3(CH_2)_{15}$, $CH_3(CH_2)_{14}$, $CH_3(CH_2)_{13}$,$CH_3(CH_2)_{12}$, $CH_3(CH_2)_{11}$,

$CH_3(CH_2)_{10}$, $CH_3(CH_2)_9$, $CH_3(CH_2)_8$, $CH_3(CH_2)_9OPh$-, $CH_3(CH_2)_6C=C(CH_2)_6$, $CH_3(CH_2)_{11}O(CH_2)_3$, and $CH_3(CH_2)_9O(CH_2)_2$.

12. The compound of any one of Claims 5-9, wherein T is a fatty acid derivative.

13. The compound of Claim 12, wherein the fatty acid is selected from the group consisting of: butyric acid, caproic acid, caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, arachidic acid, behenic acid, lignoceric acid, myristoleic acid, palmitoleic acid, oleic acid, linoleic acid, α-linolenic acid, arachidonic acid, eicosapentaenoic acid, erucic acid, docosahexaenoic acid.

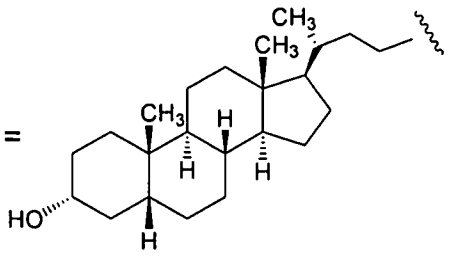
14. The compound of any one of Claims 5-9, wherein T is a bile acid derivative.

15. The compound of Claim 14, wherein the bile acid is selected from the group consisting of: lithocholic acid, chenodeoxycholic acid, deoxycholic acid, cholanic acid, cholic acid, ursocholic acid, ursodeoxycholic acid, isoursodeoxycholic acid, lagodeoxycholic acid, dehydrocholic acid, hyocholic acid, and hyodeoxycholic acid.

16. The compound of any one of Claims 5-9, wherein T is selected from sterols; progestagens; glucocorticoids; mineralcorticoids; androgens; and estrogens.

17. The compound of any one of Claims 5-9, wherein TL is selected from:
$CH_3(CH_2)_{15}$–C(O);
$CH_3(CH_2)_{13}$– C(O);
$CH_3(CH_2)_9O(CH_2)_2C(O)$;
$CH_3(CH_2)_{10}O(CH_2)_2C(O)$;
$CH_3(CH_2)_6C=C(CH_2)_6$-C(O);
LCA-C(O); and
$CH_3(CH_2)_9OPh$-C(O) wherein

LCA =



18. A method of treating diseases and conditions associated with PTHR1 modulation in a patient in need thereof comprising administering to said patient and effective amount of a compound of Claim 1-17.

19. The method of Claim 18, wherein the disease or condition is selected from: osteoporosis; humoral hypercalcemia of malignancy; osteolytic and osteoblastic metastasis to bone; primary and secondary hyperparathyroidism associated increase in bone absorption; vascular calcification; psychiatric disorders and cognitive disorders associated with hyperparathyroidism; dermatological disorders; and excess hair growth.

20. A pharmaceutical composition comprising a compound of any one of Claims 1-17 and a pharmaceutically acceptable carrier.