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(54) Title: ASSESSMENT OF NEURONS IN THE ARCUATE NUCLEUS TO SCREEN FOR AGENTS THAT MODIFY FEEDING BEHAVIOR

(57) Abstract: Screening methods of use in identifying agents that affect caloric intake, food intake, appetite, and energy expenditure are disclosed herein. These methods are used to identify agents of use in treating obesity, or that can be used to decrease the weight of a subject. These methods can also be used to identify agents of use in treating anorexia or cachexia and can be used to increase appetite and to increase the weight and lean body mass of a subject.

ASSESSMENT OF NEURONS IN THE ARCUATE NUCLEUS TO SCREEN FOR AGENTS THAT MODIFY FEEDING BEHAVIOR

PRIORITY CLAIM

This application claims the benefit of U.S. Provisional Application No. 60/324,406, filed September 24, 2001, and U.S. Provisional Application No. 60/392,109, filed June 28, 2002, which are both incorporated by reference in their entirety herein.

STATEMENT OF GOVERNMENT SUPPORT

This invention was made with United States government support pursuant to grants TW001233, RR00163, DK51730 and DK55819, from the National Institutes of Health. The United States government has certain rights in the invention.

FIELD

This application relates to the field of weight gain and reduction, specifically to screening for agents that can be used to control appetite, food intake, and calorie intake. Methods for screening for agents that can be used to treat obesity, and related disorders, are disclosed. Methods are also disclosed for screening for agents that can be used to treat cachexia, anorexia, and other disorders of energy homeostasis.

BACKGROUND

All references, including any patents or patent applications, cited in this specification are hereby incorporated by reference. No admission is made that any reference constitutes prior art. The discussion of the references states what their authors assert, and the applicants reserve the right to challenge the accuracy and pertinency of the cited documents. It will be clearly understood that, although a number of prior art

publications are referred to herein, this reference does not constitute an admission that any of these documents forms part of the common general knowledge in the art, in Australia or in any other country.

According to the National Health and Nutrition Examination Survey (NHANES III, 1988 to 1994), between one third and one half of men and women in the United States are overweight. In the United States, sixty percent of men and fifty-one percent of women, of the age of 20 or older, are either overweight or obese. In addition, a large percentage of children in the United States are overweight or obese.

The cause of obesity is complex and multi-factorial. Increasing evidence suggests that obesity is not a simple problem of self-control but is a complex disorder involving appetite regulation and energy metabolism. In addition, obesity

is associated with a variety of conditions associated with increased morbidity and mortality in a population. Although the etiology of obesity is not definitively established, genetic, metabolic, biochemical, cultural and psychosocial factors are believed to contribute. In general, obesity has been described as a condition in which excess body fat puts an individual at a health risk.

There is strong evidence that obesity is associated with increased morbidity and mortality. Disease risk, such as cardiovascular disease risk and type 2 diabetes disease risk, increases independently with increased body mass index. Indeed, this risk has been quantified as a five percent increase in the risk of cardiac disease for males, and a seven percent increase in the risk of cardiac disease for females, for each point of a BMI greater than 24.9 (see Kenchaiah et al., *N. Engl. J. Med.* 347:305, 2002; Massie, *N. Engl. J. Med.* 347:358, 2002). In addition, there is substantial evidence that weight loss in obese persons reduces important disease risk factors. Even a small weight loss, such as 10% of the initial body weight, in both overweight and obese adults has been associated with a decrease in risk factors such as hypertension, hyperlipidemia, and hyperglycemia.

Although diet and exercise provide a simple process to decrease weight gain, overweight and obese individuals often cannot sufficiently control these factors to effectively lose weight. Weight loss surgery is an option in carefully selected patients with clinically severe obesity. However, these treatments are high-risk, and a suitable for use in only a limited number of patients. Limited pharmacotherapy is available; several weight loss drugs have been approved by the Food and Drug Administration that can be used as part of a comprehensive weight loss program. However, there remains a need for agents that can be used to effect weight loss in overweight and obese subjects.

Under normal circumstances, animals and humans respond to starvation with a complex neuroendocrine response that ultimately leads to an increase in appetite, a relative sparing of lean body mass and burning of fat stores, and an overall decrease in basal metabolic rate (Webber & Macdonald, 1994, *Brit. J. Nutr.* 71:437-447; Ahima et al., 1996, *Nature* 382:250-252). In contrast, in some diseases a devastating pathological state of malnutrition known as cachexia arises, brought about by a synergistic combination of a dramatic decrease in appetite and an

increase in metabolic rate and metabolism of both fat and lean body mass, producing a relative wasting of lean body mass (Tisdale, 1997, *J. Natl. Cancer Inst.* 89:1763-1773; Inui, 1999, *Cancer Res.* 59:4493-4501; Fong et al., 1989, *Amer. J. Phys.* 256:R659-R665; Bruera, 1997, *Brit. Med. J.* 315:1219-1222; Emery, 1999, *Nutrition* 15:600-603). This combination is found in a number of disorders including cancer, cystic fibrosis, AIDS, rheumatoid arthritis, and renal failure (Tisdale, 1997, *ibid.*).

The severity of cachexia in many illnesses may be the primary determining factor in both quality of life, and in eventual mortality (Tisdale, 1997, *ibid.*; Larkin, 1998, *Lancet* 351:1336). Indeed, body mass retention in AIDS patients has a stronger correlation with survival than any other current measure of the disease (Kotler et al., 1989, *Amer. J. Clin. Nutr.* 50:444-447). Many different tumor types have been studied and it is a common finding that tumor-bearing animals die from cachexia and exhaustion of metabolic fuels, rather than from metastasis or infection (Svaninger et al., 1987, *J. Natl. Cancer Inst.* 78:943-950; Emery, 1999, *Nutrition* 15:600-603; Svaninger *et al.*, 1989, *Eur. J. Cancer Clin. Oncol.* 25:1295-1302; Emery *et al.*, 1984, *Cancer Res.* 44:2779-2784). Cachexia is commonly observed in patients with cancer, particularly in children and elderly individuals (Bruera, 1997, *ibid.*). The resulting malnutrition and loss of lean body mass reduces the quality of life for the affected individual and compromises recovery by decreasing tolerance to therapy and increasing post-surgical complications (Larkin, 1998, *ibid.*; Inui, 1999, *ibid.*).

Attempts at drug therapy for cachexia with a variety of agents has met with limited success (DeConno et al., 1998, *Eur. J Cancer* 34:1705-1709; Windisch et al., 1998, *Ann. Pharmacother.* 32:437-445; Rivandeneria et al., 1999, *Nutr. Cancer* 35:202-206; McCarthy, 1999, *Res. Nurs. Health* 22:380-387). The most widely utilized agent, megestrol acetate, has shown some promise in reversing weight loss, but this is primarily due to increases in fat mass and water retention, rather than preservation of lean body mass (Strang, 1997, *Anticancer Res.* 17:657-662). Thus, there is clearly a need to identify new agents that can be used in the treatment of cachexia and that may of use in treating other disorders, such as anorexia.

SUMMARY

Screening methods of use in identifying agents that affect caloric intake, food intake, appetite, and energy expenditure are disclosed herein. These methods are used to identify agents of use in treating obesity, or that can be used to decrease the weight of a subject. These methods are also of use to identify agents of use in treating cachexia or anorexia, or that can be used to increase the weight of a subject.

A method is disclosed herein for screening for an agent that affects caloric intake. The method includes contacting a histological section of an arcuate nucleus from a mouse expressing a marker in proopiomelanocortin neurons with an agent to be tested. The mouse comprises a transgene comprising a nucleic acid encoding the marker operably linked to a proopiomelanocortin nucleic acid sequence, wherein the proopiomelanocortin nucleic acid sequence directs expression of the marker in proopiomelanocortin neurons in the arcuate nucleus of the mouse. An electrophysiological response of a proopiomelanocortin neuron in the histological section is assessed, thereby determining if the agent affects caloric intake.

A method is disclosed herein for screening for an agent that affects food intake. The method includes contacting a histological section of an arcuate nucleus from a mouse expressing a marker in proopiomelanocortin neurons with an agent to be tested. The mouse comprises a transgene comprising a nucleic acid encoding the marker operably linked to a proopiomelanocortin nucleic acid sequence, wherein the proopiomelanocortin nucleic acid sequence directs expression of the marker in proopiomelanocortin neurons in the arcuate nucleus of the mouse. An electrophysiological response of a proopiomelanocortin neuron in the histological section is assessed, thereby determining if the agent affects food intake.

A method is disclosed herein for screening for an agent that affects appetite. The method includes contacting a histological section of an arcuate nucleus from a mouse expressing a marker in proopiomelanocortin neurons with an agent to be tested. The mouse comprises a transgene comprising a nucleic acid encoding the marker operably linked to a proopiomelanocortin nucleic acid sequence, wherein the proopiomelanocortin nucleic acid sequence directs expression of the marker in proopiomelanocortin neurons in the arcuate nucleus of the mouse. An

electrophysiological response of a proopiomelanocortin neuron in the histological section is assessed, thereby determining if the agent affects appetite.

A method is disclosed herein for screening for an agent that affects caloric intake, appetite, energy expenditure or food intake. The method includes contacting a histological section of an arcuate nucleus, with an agent to be tested. Proopiomelanocortin neurons in the histological section express a heterologous marker that distinguishes the proopiomelanocortin neurons from other cells in the histological section. An electrophysiological response of a proopiomelanocortin neuron in the histological section is assessed, thereby determining if the agent affects caloric intake, appetite, energy expenditure, or food intake.

The foregoing and other features and advantages will become more apparent from the following detailed description of several embodiments, which proceeds with reference to the accompanying figures.

BRIEF DESCRIPTION OF THE FIGURES

Fig. 1 is a set of diagrams and digital images showing the generation of transgenic mice expressing EGFP in ARC POMC neurons. Fig. 1a is a schematic diagram of the structure of the POMC-EGFP transgene. Fig. 1b is a digital image showing the identification of a single POMC neuron (arrowhead on recording electrode tip) by EGFP fluorescence (upper) and IR-DIC microscopy (lower) in a living ARC slice prior to electrophysiological recordings. Fig. 1c is a set of digital images showing the co-localization (bright, on right) of EGFP (left) and ß-endorphin immunoreactivity (middle) in ARC POMC neurons. Scale bars: b & c, 50 μm. Fig. 1d is a set of diagrams showing the distribution of EGFP-positive neuronal soma throughout the ARC nucleus. o = 5 cells, • = 10 cells.

Fig. 2 is a tracing and graphs showing activation of MOP-Rs hyperpolarizes the EGFP-labeled POMC neurons by opening G protein-coupled inwardly-rectifying potassium channels. Fig. 2a is a tracing showing met-enkephalin hyperpolarizes POMC neurons and inhibits all action potentials. The horizontal bar indicates the time when 30 μM Met-Enk was bath-applied to the slice. Fig. 2b is a graph showing

met-enkephalin current and reversal potential is shifted by extracellular K^+ concentration. Fig. 2c is a graph showing met-enkephalin activates MOP-Rs on POMC neurons. A Met-Enk (30 μM) current was observed and the MOP-R specific antagonist CTAP (1 μM) was applied for 1 minute. Following CTAP Met-Enk elicited no current. The figure is representative of three experiments.

Fig. 3 are tracings and graphs demonstrating that leptin depolarizes POMC neurons via a non-specific cation channel, and decreases GABAergic tone onto POMC cells. Fig. 3a is a tracing demonstrating that leptin depolarizes POMC neurons and increases the frequency of action potentials within 1 to 10 minutes of addition. The figure is a representative example of recordings made from 77 POMC neurons. Fig. 3b is a graph showing that leptin causes a concentration dependent depolarization of POMC cells. The depolarization caused by leptin was determined at 0.1, 1, 10, 50, and 100 nM (EC_{50} =5.9 nM) in (8, 7, 9, 3, 45) cells respectively. Fig. 3c is a graph showing that leptin depolarizes POMC cells by activating a nonspecific cation current. The figure is representative of the response in 10 cells. Fig. 3d is a graph showing that leptin decreases the frequency of IPSCs in POMC cells. The figure is an example of 5 cells in which leptin (100 nM) decreased the frequency of IPSCs. Fig. 3e is a tracing demonstrating that leptin had no effect on 5 adjacent non-fluorescent ARC neurons. Fig. 3f is a tracing showing that leptin hyperpolarized 5 non-fluorescent ARC neurons.

Fig. 4 is a set of images showing that the GABAergic inputs to POMC cells are from NPY neurons that co-express GABA. Fig. 4a is a graph showing that NPY decreases the frequency of mini IPSCs in POMC neurons. Fig. 4b is a graph demonstrating that D-Trp^8-γMSH (7nM), a dose that selectively activates MC3-R, increases the frequency of GABAergic IPSCs in POMC neurons. Fig. 4c is a tracing showing that D-Trp^8-γMSH hyperpolarizes POMC neurons. Figs. 4a, 4b and 4c are representative. Fig. 4d is a set of digital images demonstrating that expression of NPY in nerve terminals adjacent to POMC neurons in the ARC. NPY nerve terminals (black, arrowheads); POMC neuronal soma (grey). Scale bar, 10 μm. Fig. 4e is a digital image showing expression of GABA and NPY in nerve terminals

synapsing onto POMC neurons in the ARC. GABA immunoreactivity (10 nm gold particles, arrowheads without tail) and NPY immunoreactivity (25 nm gold particles, arrows with tail) are in separate vesicle populations co-localized within synaptic boutons that make direct contact with the soma of POMC neurons (DAB contrasted with uranyl acetate and lead citrate, diffuse black in cytoplasm). Scale bar, 1 μm. Fig. 4f is a diagram of the model of NPY/GABA and POMC neurons in the ARC.

Fig. 5 is a set of digital images of c-fos expression in *Pomc-EGFP* mice. Figs. 5a and 5b are digital images of representative sections (bregma –1.4 mm) of c-fos expression in the arcuate nucleus of *Pomc-EGFP* mice response to intraperitoneal saline (Fig. 5a) or PYY_{3-36} (5μg/100g) (Fig. 5b). Scale bar 100 μm. 3V, third ventricle; Arc, arcuate nucleus. Figs. 5c and 5d are digital images of representative sections showing POMC-EGFP neurones (Fig. 5c) and c-fos immunoreactivity (Fig. 5d) either co-localising (bright arrows) or alone (single darker arrow). Scale bar 25 μm.

Fig. 6 is a set of images relating to the electrophysiological responses to PYY_{3-36} and Y2A. Fig. 6a is a tracing and bar graph showing the effect of PYY_{3-36} (10 nM) on the frequency of action potentials in POMC neurons (whole-cell configuration recordings; n = 22) * $p< 0.05$. PYY_{3-38} was administered at time D for 3 minutes; baseline, -3 to 0 minute; PYY_{3-36}, 2-5 minutes; and wash-out, 8-11 minutes. Inset shows a representative recording of membrane potential and action potential frequency. Fig. 6b is a graph of the effect if PYY_{3-38} (10nM) on the frequency of action potentials in loose cell-attached patch recordings (n=8). Data from individual cells were normalized to the firing rate for the 200s before PYY_{3-38} addition. Fig. 6c is a tracing and a graph of the effect of PYY_{3-38} (50nM) on spontaneous IPSCs onto POMC neurons (n=13). Inset shows a representative recording of IPSCs before and after PYY_{3-36} (50nM), respectively, results in Fig. 6a-6c are expressed as mean ± s.e.m.

Fig. 7 is a schematic diagram of transgenes carrying variable lengths or deletions of 5' flanking sequences of the mouse POMC gene. The EGFP gene was

inserted into the second exon immediately before the site of translational initiation. A polyadenylation signal from the large T antigen of the SV40 virus was included immediately adjacent and 3' to the EGFP gene. Black boxes are mouse POMC exons. Open boxes, EGFP. Striped boxes are nPOMC1 and nPOMC2 sites. The white box is the TK minimal promoter in front of the hGH structural gene. Right: Plus signs indicate that the transgene is expressed correctly in POMC pituitary cells or in POMC hypothalamic neurons and minus signs indicate the absence of expression.

Fig. 8 is a set of digital images of sections of the arcuate nucleus. Fig. 8a is a digital image showing fluorescence in POMC neurons of the arcuate nucleus of the hypothalamus in a –13/+8 POMC-EGFP (delta –6.5/0.8) transgenic mouse. Fig. 8b is a digital image showing immunofluorescence histochemistry using an antisera specific for human growth hormone. POMC neurons in the arcuate nucleus of a –13/-9 POMC-TKhGH transgenic mouse express the hGH marker. Fig. 8c is a digital image of higher power magnification of neuronal cell bodies and processes from Fig. 8b.

Fig. 9 is a set of diagrams showing sequence alignments. Fig. 9a is a PIP Maker multiple sequence alignment between 24 kb containing the human POMC gene, 4 kb of the mouse 5' flanking region located between 9 and 13 kb from the TATA box, and the three exons with short flanking sequences obtained from Genebank (J00610, J00611, and J00612). Conserved regions are indicated with horizontal black lines on gray shaded background. Exons 1, 2, and 3 are indicated; repetitive intergenic regions are present at -5kb and -6kb; two highly conserved intergenic regions longer than 100 bp are identified as nPOMC1 and nPOMC2. The gray and white horizontal boxes indicated GC-rich regions. Fig. 9b is a similar analysis performed with the Dotter program using the 4kb between -13 and -9 of the mouse POMC gene and 15 kb of the human 5' flanking region. Diagonal lines inside the gray-shaded areas indicate the conserved sites nPOMC1 and nPOMC2.

Fig. 10 is the sequence alignments of nPOMC1 (5' half, human (SEQ ID NO: 336, cow (SEQ ID NO: 337), hamster (SEQ ID NO: 338), mouse (SEQ ID NO: 339), and rat (SEQ ID NO: 340)), nPOMC1 (3' half, human (SEQ ID NO: 341), mouse (SEQ ID NO: 342), and rat (SEQ ID NO: 343)) and nPOMC2 (human (SEQ ID NO: 344), Cow (SEQ ID NO: 345), mouse (SEQ ID NO: 346) and rabbit (SEQ ID NO: 347).

Fig. 11 is the nucleotide sequences of mouse and human nPOMC1 and nPOMC2 elements. NPOMC1 element from mouse chromosome 12 nucleotides 3,808,013-3,808,447 (SEQ ID NO: 348), nPOMC1 element from human chromosome 2 nucleotides 2,324,416-2,323,942 (SEQ ID NO: 349), nPOMC2 element from mouse chromosome 12 (SEQ ID NO: 350) and the nPOMC2 element from human chromosome 2 (SEQ ID NO: 351) are shown.

Fig. 12 is a set of graphs demonstrating that Ghrelin increases the secretory activity of NPY neurons onto POMC neurons, hyperpolarizes POMC neurons, and decreases the frequency of action potentials in POMC neurons.
Fig. 12a is a graph demonstrating that Ghrelin increases the frequency of spontaneous synaptic GABA release onto POMC neurons. Results shown in the figure are representative of 18 experiments. Increased release of GABA from NPY neurons is shown, thus Ghrelin is increasing the activity of NPY neurons. Fig. 12b is a graph demonstrating that Ghrelin mildly hyperpolarizes POMC neurons and decreases the spontaneous activity of POMC neurons. Results shown in the figure are representative of 34 experiments. Figs. 12a and 12b were recorded in conventional whole cell mode. Fig. 12c is a graph demonstrating that Ghrelin decreases the frequency of action potentials in POMC neurons, an effect that reverses with washout of the drug. Ghrelin induced a 50% decrease of the normalized mean (+/- s.e.m.) POMC neuron firing rate. These recordings were made in loose-cell-attached mode.

Fig. 13 is a set of graphs demonstrating that fenfluramine (d-FEN) increases the frequency of action potentials and depolarizes POMC neurons. Fig. 13a shows

the results obtained using a loose-cell-attached mode. d-FEN (20 microM) induced a doubling of the mean (+/- s.e.m.) POMC-neuron firing rate (n=3). This effect was reversed with drug washout. Fig. 13b is a graph of the mean (+/- s.e.m.) peak depolarization of POMC neurons (n= 4-8 per dose) bathed with d-FEN, 5-HT mCPP or MK 212 using conventional whole cell recordings.

SEQUENCE LISTING

The nucleic and amino acid sequences listed in the accompanying sequence listing are shown using standard letter abbreviations for nucleotide bases, and three letter code for amino acids, as defined in 37 C.F.R. 1.822. Only one strand of each nucleic acid sequence is shown, but the complementary strand is understood as included by any reference to the displayed strand.

DETAILED DESCRIPTION

I. Abbreviations

α-MSH: alpha melanocortin stimulating hormone

Arc: arcuate nucleus of the hypothalamus

CPP: *m*-CPP hydrochloride, 1-(3-Chlorophenyl)piperazine 5-$HT_{2B/2C}$ receptor agonist

d-FEN: fenfluarmine

EPSP: excitatory postsynaptic potential

GABA: γaminobutyric acid

GFP, EGFP: green fluorescent protein, enhanced green fluorescent protein

IPSC: inhibitory postsynaptic current

kb: kilobase

kg: kilogram

MOP-R: μ-opiod receptor

MK: MK212 hydrochloride, or 6-Chloro-2-(1-piperazinyl)pyrazine 5-HT_{2C} serotonin receptor agonist.

MV: millivolts

nPOMC1: neural POMC regulatory element 1

nPOMC2: neural POMC regulatory element 2

NPY: neuropeptide Y

pmol: picomole

POMC: proopiomelanocortin

RIA: radioimmunoassay

RPA: RNase protection assay

s.e.m.: standard error of the mean

TH: tyrosine hydroxylase

µM: micromolar

V: volts

Y2A: N-acetyl (Leu ^{28}, Leu ^{31}) NPY (24-36)

II. Terms

Unless otherwise noted, technical terms are used according to conventional usage. Definitions of common terms in molecular biology may be found in Benjamin Lewin, *Genes V*, published by Oxford University Press, 1994 (ISBN 0-19-854287-9); Kendrew et al. (eds.), *The Encyclopedia of Molecular Biology*, published by Blackwell Science Ltd., 1994 (ISBN 0-632-02182-9); and Robert A. Meyers (ed.), *Molecular Biology and Biotechnology: a Comprehensive Desk Reference*, published by VCH Publishers, Inc., 1995 (ISBN 1-56081-569-8).

In order to facilitate review of the various embodiments of this disclosure, the following explanations of specific terms are provided:

Action potential: A rapidly propagated electrical message that speeds along an axon of a neuron and over the surface membrane of many muscle and glandular cells. In axons they are brief, travel at constant velocity, and maintain a constant amplitude. Like all electrical messages of the central nervous system, the action potential is a membrane potential change caused by the flow of ions through ion channels in the membrane. In one embodiment, an action potential is a regenerative wave of sodium permeability.

Agent: Any polypeptide, compound, small molecule, organic compound, salt, polynucleotide, or other molecule of interest.

Animal: Living multi-cellular vertebrate organisms, a category that includes, for example, mammals and birds. The term mammal includes both human and non-human mammals. Similarly, the term "subject" includes both human and veterinary subjects.

Anorexia: A lack or loss of the appetite for food. In one embodiment, anorexia is a result of "anorexia nervosa." This is an eating disorder primarily affecting females, usually with onset in adolescence, characterized by refusal to maintain a normal minimal body weight, intense fear of gaining weight or becoming obese, and a disturbance of body image resulting in a feeling of being fat or having fat in certain areas even when extremely emaciated, undue reliance on body weight or shape for self-evaluation, and amenorrhea. Associated features often include denial of the illness and resistance to psychotherapy, depressive symptoms, markedly decreased libido, and obsessions or peculiar behavior regarding food, such as hoarding. The disorder is divided into two subtypes, a *restricting* type, in which weight loss is achieved primarily through diet or exercise, and a *binge-eating/purging* type, in which binge eating or purging behavior also occur regularly.

Antagonist: A substance that tends to nullify the action of another, as an agent that binds to a cell receptor without eliciting a biological response, blocking binding of substances that could elicit such responses.

Appetite: A natural desire, or longing for food. In one embodiment, appetite is measured by a survey to assess the desire for food. Increased appetite generally leads to increased feeding behavior.

Appetite Suppressants: Compounds that decrease the desire for food. Commercially available appetite suppressants include, but are not limited to, amfepramone (diethylpropion), phentermine, mazindol and phenylpropanolamine fenfluramine, dexfenfluramine, and fluoxetine.

Binding: A specific interaction between two molecules, such that the two molecules interact. Binding can be specific and selective, so that one molecule is bound preferentially when compared to another molecule. In one embodiment, specific binding is identified by a disassociation constant (K_d).

Body Mass Index (BMI): A mathematical formula for measuring body mass, also sometimes called Quetelet's Index. BMI is calculated by dividing weight

(in kg) by height^2 (in meters^2). The current standards for both men and women accepted as "normal" are a BMI of 20-24.9 kg/m^2. In one embodiment, a BMI of greater than 25 kg/m^2 can be used to identify an obese subject. Grade I obesity corresponds to a BMI of 25-29.9 kg/m^2. Grade II obesity corresponds to a BMI of 30-40 kg/m^2; and Grade III obesity corresponds to a BMI greater than 40 kg/m^2 (Jequier, *Am. J Clin. Nutr.*, 45:1035-47, 1987). Ideal body weight will vary among species and individuals based on height, body build, bone structure, and sex.

c-fos: The cellular homologue of the viral v-fos oncogene found in FBJ (Finkel-Biskis-Jinkins) and FBR murine osteosarcoma viruses (MSV). The human fos gene maps to chromosome 14q21-q31. Human fos has been identified as TIS-28.

C-fos is thought to have an important role in signal transduction, cell proliferation, and differentiation. It is a nuclear protein which, in combination with other transcription factors (for example, jun) acts as a trans-activating regulator of gene expression. C-fos is an immediate early response gene, which are believed to play a key role in the early response of cells to growth factors. C-fos is involved also in the control of cell growth and differentiation of embryonic hematopoietic cells and neuronal cells. The human c-fos coding amino acid and nucleic acid sequences are known (e.g. see Verma et al., Cold Spring Harb. Symp. Quant. Biol. 51, 949, 1986; GenBank Accession Nos. K00650 and M16287, and are available on the internet).

Cachexia: General physical wasting and malnutrition that is often associated with a chronic disease process. Cahexia is frequently seen in patients with cancer, AIDS, or other diseases. Cachexia includes, but is not limited to 1) cancerous cachexia, seen in cases of malignant tumor; 2) cardiac cachexia, an emaciation due to heart disease, usually caused by a combination of increased caloric expenditure and decreased caloric intake or utilization; 3) fluoric cachexia, seen in fluorosis; 4) hypophysial cachexia; 5) cachexia hypophysiopriva, a cluster of symptoms resulting from total deprivation of function of the pituitary gland, including phthisis, loss of sexual function, atrophy of the pituitary target glands, bradycardia, hypothermia, apathy, and coma; 6) malarial cachexia, a group of physical signs of a chronic nature that result from antecedent attacks of severe malaria; 7) cachexia mercurialis, seen in chronic mercury poisioning; 8) pituitary

cachexia; 9) saturnine cachexia, seen in chronic lead poisioning; 10) cachexia suprarenalis, associated with Addison's disease; and 11) uremic cachexia, associated with other systemic symptoms of advanced renal failure.

Caloric intake or calorie intake: The number of calories (energy) consumed by an individual.

Calorie: A unit of measurement in food. A standard calorie is defined as 4.184 absolute joules, or the amount of energy it takes to raise the temperature of one gram of water from 15 to 16° C (or1/100th the amount of energy needed to raise the temperature of one gram of water at one atmosphere pressure from 0° C to 100° C), food calories are actually equal to 1,000 standard calories (1 food calorie = 1 kilocalorie).

Conservative variation: The replacement of an amino acid residue by another, biologically similar residue. Examples of conservative variations include the substitution of one hydrophobic residue such as isoleucine, valine, leucine or methionine for another, or the substitution of one polar residue for another, such as the substitution of arginine for lysine, glutamic for aspartic acid, or glutamine for asparagine, and the like. The term "conservative variation" also includes the use of a substituted amino acid in place of an unsubstituted parent amino acid provided that antibodies raised to the substituted polypeptide also immunoreact with the unsubstituted polypeptide.

Non-limiting examples of conservative amino acid substitutions include those listed below:

Original Residue	Conservative Substitutions
Ala	Ser
Arg	Lys
Asn	Gln, His
Asp	Glu
Cys	Ser
Gln	Asn
Glu	Asp
His	Asn; Gln
Ile	Leu, Val
Leu	Ile; Val
Lys	Arg; Gln; Glu
Met	Leu; Ile

Phe	Met; Leu; Tyr
Ser	Thr
Thr	Ser
Trp	Tyr
Tyr	Trp; Phe
Val	Ile; Leu

Current: The amount of charge per unit time. Current is generated in a cell membrane of a neuron by an action potential or by opening of ion channels in the cell membrane and serves to depolarize or hyperpolarize adjacent membrane areas.

Deletion: The removal of a sequence of nucleic acid, such as DNA, the regions on either side being joined together.

DNA (deoxyribonucleic acid): DNA is a long chain polymer which comprises the genetic material of most living organisms (some viruses have genes comprising ribonucleic acid, RNA). The repeating units in DNA polymers are four different nucleotides, each of which comprises one of the four bases, adenine, guanine, cytosine, and thymine bound to a deoxyribose sugar to which a phosphate group is attached. Triplets of nucleotides, referred to as codons, in DNA molecules code for amino acid in a polypeptide. The term codon is also used for the corresponding (and complementary) sequences of three nucleotides in the mRNA into which the DNA sequence is transcribed.

Depolarization: An increase in the membrane potential of a cell. Certain stimuli reduce the charge across the plasma membrane. These can be electrical stimuli (which open or close voltage-gated channels), mechanical stimuli (which activate mechanically-gated channels) or certain neurotransmitters (which open ligand-gated channels). In each case, the facilitated diffusion of sodium into the cell increases the resting potential at that spot on the cell creating an excitatory postsynaptic potential (EPSP). Depolarizations can also be generated by decreasing the frequency of inhibitory postsynaptic currents (IPSCs), these are due to inhibitory neurotransmitters facilitating the influx of chloride ions into the cell, creating an IPSC. Depolarizations can also be induced by closing some ion channels. If the potential is increased to the threshold voltage (about -50 mV in mammalian neurons), an action potential is usually generated in the cell.

Diabetes: A failure of cells to transport endogenous glucose across their membranes either because of an endogenous deficiency of insulin and/or a defect in insulin sensitivity. Diabetes is a chronic syndrome of impaired carbohydrate, protein, and fat metabolism owing to insufficient secretion of insulin or to target tissue insulin resistance. It occurs in two major forms: insulin-dependent diabetes mellitus (IDDM, type I) and non-insulin dependent diabetes mellitus (NIDDM, type II) which differ in etiology, pathology, genetics, age of onset, and treatment.

The two major forms of diabetes are both characterized by an inability to deliver insulin in an amount and with the precise timing that is needed for control of glucose homeostasis. Diabetes type I, or insulin dependent diabetes mellitus (IDDM) is caused by the destruction of β cells, which results in insufficient levels of endogenous insulin. Diabetes type II, or non-insulin dependent diabetes, results from a defect in both the body's sensitivity to insulin, and a relative deficiency in insulin production.

Electroporation: A method of inducing or allowing a cell to take up macromolecules by applying electric fields to reversibly permeabilize the cell walls. Various methods and apparatuses used are further defined and described in: U.S. Patent No. 4,695,547; U.S. Patent No. 4,764,473; U.S. Patent No. 4,882,28; and U.S. Patent No. 4,946,793; U.S. Patent No. 4,906,576; U.S. Patent No. 4,923,814; and U.S. Patent No. 4,849,089.

Eukaryotic cell: A cell having an organized nucleus bounded by a nuclear membrane. These include simpler organisms such as yeasts, slime molds, and the like, as well as cells from multicellular organisms such as invertebrates, vertebrates, and mammals. Multicellular organisms include a variety of cell types, such as: endothelial cell, smooth muscle cell, epithelial cell, hepatocyte, cells of neural crest origin, tumor cell, hematopoietic cell, immunologic cell, T cell, B cell, monocyte, macrophage, dendritic cell, fibroblast, keratinocyte, neuronal cell, glial cell, adipocyte, myoblast, myocyte, chondroblast, chondrocyte, osteoblast, osteocyte, osteoclast, secretory cell, endocrine cell, oocyte, and spermatocyte. These cell types are described in standard histology texts, such as McCormack, Introduction to Histology, (c) 1984 by J.P. Lippincott Co.; Wheater *et al.*, eds., Functional

Histology, 2nd Ed., (c) 1987 by Churchill Livingstone; Fawcett *et al.*, eds., Bloom and Fawcett: A Textbook of Histology, (c) 1984 by William and Wilkins.

Gene: A DNA sequence that comprises control and coding sequences necessary for the production of a polypeptide or protein. The polypeptide can be encoded by a full-length coding sequence or by any portion of the coding sequence in some embodiments, so long as at least a portion of the desired activity of the polypeptide is retained. A "foreign gene" is any nucleic acid (e.g., gene sequence) that is introduced into the genome of an animal by experimental manipulations and can include gene sequences found in that animal so long as the introduced gene contains some modification (e.g., a point mutation, the presence of a selectable marker gene, a non-native regulatory sequence, or a native sequence integrated into the genome at a non-native location, etc.) relative to the naturally-occurring gene.

Food intake: The amount of food consumed by an individual. Food intake can be measured by volume or by weight. In one embodiment, food intake is the total amount of food consumed by an individual. In another embodiment, food intake is the amount of proteins, fat, carbohydrates, cholesterol, vitamins, minerals, or any other food component, of the individual. "Protein intake" refers to the amount of protein consumed by an individual. Similarly, "fat intake," "carbohydrate intake," "cholesterol intake," "vitamin intake," and "mineral intake" refer to the amount of proteins, fat, carbohydrates, cholesterol, vitamins, or minerals consumed by an individual.

Hyperpolarization: A decrease in the membrane potential of a cell. Inhibitory neurotransmitters inhibit the transmission of nerve impulses via hyperpolarization. This hyperpolarization is called an inhibitory postsynaptic potential (IPSP). Hyperpolarization may also be caused by opening or closing of ion channels. Although the threshold voltage of the cell is unchanged, a hyperpolarized cell requires a stronger excitatory stimulus to reach threshold.

Inhibitory Postsynaptic Current: A current that inhibits an electrophysiological parameter of a postsynaptic cell. The potential of a postsynaptic cell can be analyzed to determine an effect on a presynaptic cell. In one embodiment, the postsynaptic cell is held in voltage clamp mode, and postsynaptic currents are recorded. If necessary, antagonists of other classes of

current can be added. In one specific, non-limiting example, to record GABAergic IPSCs, blockers of excitatory channels or receptors can be added. The instantaneous frequency over time is then determined.

In one embodiment, IPSCs give a measure of the frequency of GABA release from an NPY neuron. Thus, as NPY neurons release GABA onto POMC neurons, measurement of IPSC frequency is a gauge of the inhibitory tone that POMC neurons are receiving, and can be used to assess the effect of an agent that affects an NPY neuron, such as an antagonist or agonist of PYY.

Intron: An intragenic nucleic acid sequence in eukaryotes that is not expressed in a mature RNA molecule. Introns of the present disclosure include full-length intron sequences, or a portion thereof, such as a part of a full-length intron sequence.

***In vitro* amplification:** Techniques that increases the number of copies of a nucleic acid molecule in a sample or specimen. An example of amplification is the polymerase chain reaction, in which a biological sample collected from a subject is contacted with a pair of oligonucleotide primers, under conditions that allow for the hybridization of the primers to nucleic acid template in the sample. The primers are extended under suitable conditions, dissociated from the template, and then re-annealed, extended, and dissociated to amplify the number of copies of the nucleic acid. The product of *in vitro* amplification may be characterized by electrophoresis, restriction endonuclease cleavage patterns, oligonucleotide hybridization or ligation, and/or nucleic acid sequencing, using standard techniques. Other examples of *in vitro* amplification techniques include strand displacement amplification (see U.S. Patent No. 5,744,311); transcription-free isothermal amplification (see U.S. Patent No. 6,033,881); repair chain reaction amplification (see WO 90/01069); ligase chain reaction amplification (see EP-A-320 308); gap filling ligase chain reaction amplification (see U.S. Patent No. 5,427,930); coupled ligase detection and PCR (see U.S. Patent No. 6,027,889); and NASBA™ RNA transcription-free amplification (see U.S. Patent No. 6,025,134).

Isolated: An isolated biological component (such as a nucleic acid, peptide or protein) has been substantially separated, produced apart from, or purified away from other biological components in the cell of the organism in which the

component naturally occurs, *i.e.* other chromosomal and extrachromosomal DNA and RNA, and proteins. Nucleic acids, peptides and proteins that have been isolated include nucleic acids and proteins purified by standard purification methods. The term also embraces nucleic acids, peptides, and proteins prepared by recombinant expression in a host cell as well as chemically synthesized nucleic acids.

Marker: A protein, or a gene encoding a protein, for which a system is available to identify cells that produce the protein. Specific non-limiting examples of a marker include drug resistance markers, such as G148 or hygromycin. Additionally, a marker can be a protein or a gene encoding a protein for which negative selection can be used to identify the cell expressing the marker. A specific, non-limiting examples of a negative selection marker includes, but is not limited to, the HSV-tk gene. This gene will make the cells sensitive to agents such as acyclovir and gancyclovir. Another specific, non-limiting example of a selectable marker is a protein, or a gene encoding a protein, wherein selection can be made by using a cell surface marker, for example, to select overexpression of the marker by fluorescence activated cell sorting (FACS). In another specific, non-limiting example of a selectable marker is a protein, or a gene encoding a protein, that can be identified in a cell based on its fluorescent or enzymatic properties. Specific, non-limiting examples include, but are not limited to, enhanced fluorescent green protein (EGFP), alkaline phosphatase, or horseradish peroxidase. A marker can also be a polypeptide or antigenic epitope thereof, wherein an antibody that specifically binds the polypeptide can be used to identify cells that express the polypeptide or antigenic epitope. One specific, non-limiting example of a polypeptide of use is human growth Hormone (hGH).

Membrane potential: The electrical potential of the interior of the cell with respect to the environment, such as an external bath solution. One of skill in the art can readily assess the membrane potential of a cell, such as by using conventional whole cell techniques. Activation of a cell is associated with less negative membrane potentials (for example shifts from about –50 mV to about –40 mV). These changes in potential increase the likelihood of action potentials, and thus lead to an increase in the rate of action potentials.

The rate of action potentials can be assessed using many approaches, such as using conventional whole cell access, or using, for example, perforated-patch whole-cell and cell-attached configurations. In each event the absolute voltage or current is not assessed, rather the frequency of rapid deflections characteristic of action potentials is assessed, as a function of time (therefore this frequency is an instantaneous frequency, reported in "bins"). This time component can be related to the time at which a compound, such as a PYY agonist, is applied to the bath to analyze the effect of the compound, such as the PYY agonist, on action potential firing rate.

Neuropeptide Y (NPY): A 36-amino acid peptide that is a neuropeptide identified in the mammalian brain. NPY is believed to be an important regulator in both the central and peripheral nervous systems and influences a diverse range of physiological parameters, including effects on psychomotor activity, food intake, central endocrine secretion, and vasoactivity in the cardiovascular system. High concentrations of NPY are found in the sympathetic nerves supplying the coronary, cerebral, and renal vasculature and have contributed to vasoconstriction. NPY binding sites have been identified in a variety of tissues, including spleen, intestinal membranes, brain, aortic smooth muscle, kidney, testis, and placenta. In addition, binding sites have been reported in a number of rat and human cell lines.

Neuropeptide Y (NPY) receptor has structure/activity relationships within the pancreatic polypeptide family. This family includes NPY, which is synthesized primarily in neurons; peptide YY (PYY), which is synthesized primarily by endocrine cells in the gut; and pancreatic polypeptide (PP), which is synthesized primarily by endocrine cells in the pancreas. These 36 amino acid peptides have a compact helical structure involving an amino acid structure, termed a "PP-fold" in the middle of the peptide.

NPY binds to several receptors, including the Y1, Y2, Y3, Y4 (PP), Y5, Y6, and Y7 receptors. These receptors are recognized based on binding affinities, pharmacology, and sequence (if known). Most, if not all of these receptors are G protein coupled receptors. The Y1 receptor is generally considered to be postsynaptic and mediates many of the known actions of neuropeptide Y in the periphery. Originally, this receptor was described as having poor affinity for C-

terminal fragments of neuropeptide Y, such as the 13-36 fragment, but interacts with the full length neuropeptide Y and peptide YY with equal affinity (e.g. see Patent Cooperation Treaty publication WO 93/09227).

Pharmacologically, the Y2 receptor is distinguished from Y1 by exhibiting affinity for C-terminal fragments of neuropeptide Y. The Y2 receptor is most often differentiated by the affinity of neuropeptide Y(13-36), although the 3-36 fragment of neuropeptide Y and peptide YY provides improved affinity and selectivity (see Dumont et al., *Society for Neuroscience Abstracts*, 19:726, 1993). Signal transmission through both the Y1 and the Y2 receptors are coupled to the inhibition of adenylate cyclase. Binding to the Y-2 receptor was also found to reduce the intracellular levels of calcium in the synapse by selective inhibition of N-type calcium channels. In addition, the Y-2 receptor, like the Y1 receptors exhibits differential coupling to second messengers (see U.S. Patent No. 6,355,478). Y2 receptors are found in a variety of brain regions, including the hippocampus, substantia nigra-lateralis, thalamus, hypothalamus, and brainstem. The human, murine, monkey and rat Y2 receptors have been cloned (e.g. see U.S. Patent No. 6,420,352 and U.S. Patent No. 6,355,478).

A Y2 receptor agonist is a peptide, small molecule, or chemical compound that preferentially binds to the Y2 receptor and stimulates intracellular signaling. In one embodiment, an agonist for the Y2 receptor binds to the receptor with an equal or greater affinity than NPY. In another embodiment, an agonist selectively binds the Y2 receptor, as compared to binding to another receptor.

One of skill in the art can readily determine the dissociation constant (K_d) value of a given compound. This value is dependent on the selectivity of the compound tested. For example, a compound with a K_d which is less than 10 nM is generally considered an excellent drug candidate. However, a compound that has a lower affinity, but is selective for the particular receptor, can also be a good drug candidate. In one specific, non-limiting example, an assay, such as a competition assays, is used to determine if a compound of interest is a Y2 receptor agonist. Assays useful for evaluating neuropeptide Y receptor antagonists are also well known in the art (see U.S. Patent No. 5,284,839, which is herein incorporated by reference, and Walker et al., *Journal of Neurosciences*, 8:2438-2446, 1988).

Normal Daily Diet: The average food intake for an individual of a given species. A normal daily diet can be expressed in terms of caloric intake, protein intake, carbohydrate intake, and/or fat intake. A normal daily diet in humans generally comprises the following: about 1,500, about 1,800, about 2,000, about 2,400, or about 2,800 to significantly more calories. In addition, a normal daily diet in humans generally includes about 12 g to about 45 g of protein, about 120 g to about 610 g of carbohydrate, and about 11 g to about 90 g of fat. A low calorie diet would be no more than about 85%, and preferably no more than about 70%, of the normal caloric intake of a human individual.

In animals, the caloric and nutrient requirements vary depending on the species and size of the animal. For example, in cats, the total caloric intake per pound, as well as the percent distribution of protein, carbohydrate and fat varies with the age of the cat and the reproductive state. A general guideline for cats, however, is 40 cal/lb/day (18.2 cal/kg/day). About 30% to about 40% should be protein, about 7% to about 10% should be from carbohydrate, and about 50% to about 62.5% should be derived from fat intake. One of skill in the art can readily identify the normal daily diet of an individual of any species.

Obesity: A condition in which excess body fat may put a person at health risk (see Barlow and Dietz, *Pediatrics* 102: E29, 1998; National Institutes of Health, National Heart, Lung, and Blood Institute (NHLBI), *Obes. Res.* 6 (suppl. 2):51S-209S, 1998). Excess body fat is a result of an imbalance of energy intake and energy expenditure. In one embodiment, the Body Mass Index (BMI) is used to assess obesity. In one embodiment, a BMI of 25.0 kg/m^2 to 29.9 kg/m^2 is overweight, while a BMI of 30 kg/m^2 is obese.

In another embodiment, waist circumference is used to assess obesity. In this embodiment, in men a waist circumference of 102 cm or more is considered obese, while in women a waist circumference of 89 cm or more is considered obese. Strong evidence shows that obesity affects both the morbidity and mortality of individuals. For example, an obese individual is at increased risk for heart disease, non-insulin dependent (type 2) diabetes, hypertension, stroke, cancer (e.g. endometrial, breast, prostate, and colon cancer), dyslipidemia, gall bladder disease,

sleep apnea, reduced fertility, and osteoarthritis, amongst others (see Lyznicki et al., *Am. Fam. Phys.* 63:2185, 2001).

Oligonucleotide: A linear polynucleotide sequence of up to about 200 nucleotide bases in length, for example a polynucleotide (such as DNA or RNA) which is at least 6 nucleotides, for example at least 15, 50, 100 or even 200 nucleotides long.

Operably linked: A first nucleic acid sequence is operably linked with a second nucleic acid sequence when the first nucleic acid sequence is placed in a functional relationship with the second nucleic acid sequence. For instance, a promoter is operably linked to a coding sequence if the promoter affects the transcription or expression of the coding sequence. Generally, operably linked DNA sequences are contiguous and, where necessary to join two protein coding regions, in the same reading frame.

Overweight: An individual who weighs more than their ideal body weight. An overweight individual can be obese, but is not necessarily obese. In one embodiment, an overweight individual is any individual who desires to decrease their weight. In another embodiment, an overweight individual is an individual with a BMI of 25.0 kg/m^2 to 29.9 kg/m^2

Pancreatic Polypeptide: A 36 amino acid peptide, produced by the pancreas that has homology to PYY and NPY.

Peripheral Administration: Administration outside of the central nervous system. Peripheral administration does not include direct administration to the brain. Peripheral administration includes, but is not limited to intravascular, intramuscular, subcutaneous, inhalation, oral, rectal, transdermal or intra-nasal administration

Polypeptide: A polymer in which the monomers are amino acid residues which are joined together through amide bonds. When the amino acids are alpha-amino acids, either the L-optical isomer or the D-optical isomer can be used, the L-isomers being preferred. The terms "polypeptide" or "protein" as used herein are intended to encompass any amino acid sequence and include modified sequences such as glycoproteins. The term "polypeptide" is specifically intended to cover naturally occurring proteins, as well as those which are recombinantly or

synthetically produced. The term "polypeptide fragment" refers to a portion of a polypeptide, for example such a fragment which exhibits at least one useful sequence in binding a receptor. The term "functional fragments of a polypeptide" refers to all fragments of a polypeptide that retain an activity of the polypeptide. Biologically functional peptides can also include fusion proteins, in which the peptide of interest has been fused to another peptide that does not decrease its desired activity.

Promoter: An array of nucleic acid control sequences which directs transcription of a nucleic acid. In one embodiment, a promoter includes necessary nucleic acid sequences near the start site of transcription, such as, in the case of a polymerase II type promoter, a TATA element. In another embodiment, a promoter includes an enhancer. In another embodiment, a promoter includes a repressor element. Enhancer and repressor elements can be located adjacent to, or distal to the sequences necessary for the start site of transcription, and can be located as much as several thousand base pairs from the start site of transcription.

A promoter can be a "strong" promoter, which promotes transcription of RNA at high levels, for example at levels such that the transcriptional activity of the promoter generally accounts for about 25% of transcriptional activity of all transcription within a cell. The strength of a promoter is often tissue-specific and thus can vary from one cell type to another. For example, CMV is a classic strong promoter because it generates high levels of transcriptional activity in many cell types.

In other embodiments, the promoter is a "tissue-specific promoter," which promotes transcription in a single cell type or narrow range of tissues. In one embodiment, a tissue specific promoter promotes expression in the pituitary, but not in other tissues. In a further embodiment, a tissue specific promoter promotes expression in the hypothalamus, but not in other tissues (e.g., heart, lung, pancreas, intestines, skin, etc.)

In other embodiments, the promoter is a "minimal" promoter, which has very low intrinsic transcriptional activity in the absence of operably linked enhancer sequences. A minimal promoter is one that does not have inherent cell-specific or tissue-specific activity, but may direct transcriptional initiation in multiple

eukaryotic cell types when operably linked to a cell- or tissue-specific enhancer sequence. One specific, non-limiting example of a minimal promoter is the minimal promoter sequences of the herpes simplex virus type 1thymidine kinase (HSV1-tk) gene.

Proopiomelanocortin (POMC): A glycosylated protein of a molecular weight of 31 kDa protein. POMC is synthesized mainly in the anterior pituitary but also found in the hypothalamus and brainstem. This protein is a precursor protein, post-translational processing of POMC yields several neuroactive peptides upon specific cleavage. The POMC coding sequence includes the amino acid sequences of adrenocoroticotropic (ACTH) hormone and beta-lipotropin. ACTH is processed to produce the proteins melanotropin (msh), corticotrophin-like intermediate lobe peptide. Beta-lipotropin is processed to produce the proteins alpha-lipotropin, beta-endorphins, beta-melanocyte stimulating hormone (MSH), and met-enkephalin. The amino-terminal fragment of POMC is processed to a family of gamma-MSH peptides and to a peptide with putative mitogenic stimulatory activity of the adrenal cortical cells. The biological activity of POMC-derived peptides is further regulated in a tissue-specific manner by acetylation of the amino-terminal amino acid residue and/or amidation of the carboxyterminal amino acid residue by the enzyme peptidyl-α-monooxygenase (PAM).

The POMC gene (human chromosome 2p23) contains three exons and two large introns: one, of about 3.5 kb, interrupts the N- terminal fragment of the common precursor mostly encoded in exon 3. Exon 2 contains the sequence for a portion of the 5' untranslated portion of the mRNA, all of the signal sequence which directs insertion of the precursor protein into the endoplasmic reticulum, and 8 amino acids of the N-terminal fragment. The overall arrangement of introns and exons in the POMC gene is almost identical in all mammalian species. Hormonal control of POMC gene transcription and release of peptide products derived from the POMC precursor is tissue-specific; for example, glucocorticoids specifically inhibit anterior but not intermediate pituitary POMC transcription.

Purified: The term purified does not require absolute purity; rather, it is intended as a relative term. Thus, for example, a purified protein preparation is one in which the protein is more pure than the protein in its natural environment within a

cell. Such proteins may be produced, for example, by standard purification techniques, or by recombinant expression. In some embodiments, a preparation of a protein is purified such that the protein represents at least 50%, for example at least 70%, of the total protein content of the preparation.

PYY: A peptide YY polypeptide obtained or derived from any species. Thus, PYY includes the human full length polypeptide (as set forth in SEQ ID NO: 1) and species variations of PYY, including e.g. murine, hamster, chicken, bovine, rat, and dog PYY. In one embodiment, PYY agonists do not include NPY. A "PYY agonist" is any compound which binds to a receptor that specifically binds PYY, and elicits an effect of PYY. In one embodiment, a PYY agonist is a compound that affects food intake, caloric intake, energy expenditure or appetite, and/or which binds specifically in a Y receptor assay or competes for binding with PYY, such as in a competitive binding assay with labeled PYY. PYY agonists include, but are not limited to, compounds that bind to the Y2 receptor.

Recombinant: A recombinant nucleic acid is one that has a sequence that is not naturally occurring or has a sequence that is made by an artificial combination of two otherwise separated segments of sequence. This artificial combination is often accomplished by chemical synthesis or, more commonly, by the artificial manipulation of isolated segments of nucleic acids, e.g. by genetic engineering techniques, such as those described in Sambrook et al. (in *Molecular Cloning: A Laboratory Manual*, Cold Spring Harbor, New York, 1989).

Sequence identity: The similarity between amino acid sequences is expressed in terms of the similarity between the sequences, otherwise referred to as sequence identity. Sequence identity is frequently measured in terms of percentage identity (or similarity or homology); the higher the percentage, the more similar the two sequences are. Homologues or variants of a POMC sequence will possess a relatively high degree of sequence identity when aligned using standard methods.

Methods of alignment of sequences for comparison are well known in the art. Various programs and alignment algorithms are described in: Smith and Waterman, *Adv. Appl. Math.* 2:482, 1981; Needleman and Wunsch, *J. Mol. Biol.* 48:443, 1970; Pearson and Lipman, *Proc. Natl. Acad. Sci. U.S.A.* 85:2444, 1988; Higgins and Sharp, *Gene* 73:237, 1988; Higgins and Sharp, *CABIOS* 5:151, 1989; Corpet et al., *Nucleic*

Acids Research 16:10881, 1988; and Pearson and Lipman, *Proc. Natl. Acad. Sci. U.S.A.* 85:2444, 1988. Altschul et al., *Nature Genet.*, 6:119, 1994 presents a detailed consideration of sequence alignment methods and homology calculations.

The NCBI Basic Local Alignment Search Tool (BLAST) (Altschul et al., *J. Mol. Biol.* 215:403, 1990) is available from several sources, including the National Center for Biotechnology Information (NCBI, Bethesda, MD) and on the Internet, for use in connection with the sequence analysis programs blastp, blastn, blastx, tblastn and tblastx. A description of how to determine sequence identity using this program is available on the NCBI website on the internet. Other specific, non-limiting examples of sequence alignment programs specifically designed to identify homologous regions of intragenic DNA of greater than or equal to 100 nucleotides are PIPMaker and DOTTER.

Homologues and variants of a POMC sequence are typically characterized by possession of at least 75%, for example at least 80%, 90%, 95%, 98%, or 99%, sequence identity counted over the full length alignment with the originating POMC sequence using the NCBI Blast 2.0, set to default parameters. Methods for determining sequence identity over such short windows are available at the NCBI website on the internet. One of skill in the art will appreciate that these sequence identity ranges are provided for guidance only; it is entirely possible that strongly significant homologues could be obtained that fall outside of the ranges provided.

Substantially purified: A polypeptide which is substantially free of other proteins, lipids, carbohydrates or other materials with which it is naturally associated. For example, the polypeptide may be at least 50%, 80% or 90% free of other proteins, lipids, carbohydrates or other materials with which it is naturally associated.

Therapeutically effective amount: A dose sufficient to prevent advancement, or to cause regression of a disorder, or which is capable of relieving a sign or symptom of a disorder. In several embodiments, a therapeutically effect of PYY or an agonist thereof is an amount sufficient to inhibit or halt weight gain, or an amount sufficient to decrease appetite, or an amount sufficient to reduce caloric intake or food intake or increase energy expenditure.

Transduced and Transfected: A virus or vector transduces or transfects a cell when it transfers nucleic acid into the cell. A cell is "transformed" by a nucleic acid transduced into the cell when the DNA becomes stably replicated by the cell, either by incorporation of the nucleic acid into the cellular genome, or by episomal replication. As used herein, the terms transduced and transfected encompass all techniques by which a nucleic acid molecule might be introduced into such a cell, including transfection with viral vectors, transfection with plasmid vectors, and introduction of naked DNA by electroporation, lipofection, injection, and particle gun acceleration.

Transgene: A foreign gene that is placed into an organism by introducing the foreign gene into embryonic stem (ES) cells, newly fertilized eggs or early embryos. In one embodiment, a transgene is a gene sequence, for example a sequence that encodes a marker polypeptide that can be detected using methods known to one of skill in the art. In another embodiment, the transgene encodes a therapeutic polypeptide that can be used to alleviate or relieve a symptom of a disorder. In yet another embodiment, the transgene encodes a therapeutically effective oligonucleotide, for example an antisense oligonucleotide, wherein expression of the oligonucleotide inhibits expression of a target nucleic acid sequence. In a further embodiment, the transgene encodes an antisense nucleic acid or a ribozyme. In yet another embodiment, a transgene is a stop cassette.

In other embodiments, a transgene contains regulatory sequences operably linked to the transgene (e.g. a promoter, such as a POMC promoter). Thus, the transgene can include regulatory sequences operably linked to a nucleic acid sequence encoding a polypeptide, such as a marker.

Transgenic Cell: Cells that contain foreign, non-native DNA.

Transgenic Animal: An animal, for example, a non-human animal such as a mouse, that has had DNA introduced into one or more of its cells artificially. By way of example, this is commonly done by random integration or by targeted insertion. DNA can be integrated in a random fashion by injecting it into the pronucleus of a fertilized ovum. In this case, the DNA can integrate anywhere in the genome, and multiple copies often integrate in a head-to-tail fashion. There is no need for homology between the injected DNA and the host genome.

Targeted insertion, the other common method of producing transgenic animals, is accomplished by introducing the DNA into embryonic stem (ES) cells and selecting for cells in which the DNA has undergone homologous recombination with matching genomic sequences. For this to occur, there often are several kilobases of homology between the exogenous and genomic DNA, and positive selectable markers are often included. In addition, negative selectable markers are often used to select against cells that have incorporated DNA by non-homologous recombination (random insertion).

Vector: A nucleic acid molecule as introduced into a cell, thereby producing a transformed cell. A vector can include nucleic acid sequences that permit it to replicate in the cell, such as an origin of replication. A vector can also include one or more marker or therapeutic transgenes and other genetic elements known in the art.

In some embodiments, the vector is a non-viral vector, such as a bacterial vector. In other embodiments, the vector is a viral vector. Examples of viral vectors include, but are not limited to adenoviral vectors, retroviral vectors, and Herpes viral vectors.

Voltage: An electric potential or potential difference, expressed in volts.

Unless otherwise explained, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. The singular terms "a", "an", and "the" include plural referents unless context clearly indicates otherwise. Similarly, the word "or" is intended to include "and" unless the context clearly indicates otherwise. It is further to be understood that all base sizes or amino acid sizes, and all molecular weight or molecular mass values, given for nucleic acids or polypeptides are approximate, and are provided for

description. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of this disclosure, suitable methods and materials are described below.

In the claims of this application and in the description of the invention, except where the context requires otherwise due to express language or necessary implication, the words "comprise" or variations such as "comprises" or "comprising" are used in an inclusive sense, i.e. to specify the presence of the stated features but not to preclude the presence or addition of further features in various embodiments of the invention. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In case of conflict,

the present specification, including explanations of terms, will control. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

Screening Methods

Methods for screening for an agent that affects caloric intake, food intake, appetite, and/or energy expenditure are disclosed herein. The methods include contacting a histological section of an arcuate nucleus from a non-human animal expressing a marker in proopiomelanocortin (POMC) neurons with the agent to be tested. The expression of the marker distinguishes the proopiomelanocortin neurons from the other neurons (and other cells) in the arcuate nucleus, such that electrophysiolological measurements can be made on the POMC neurons. An electrophysiological parameter of the POMC neurons is measured. The effect of the agent on this parameter indicated if the agent has an effect on appetite, caloric intake, food intake, or energy expenditure upon administration of a therapeutically effective amount of the agent to a subject.

In one embodiment, in order to distinguish the POMC neurons from all other cells within the arcuate nucleus, a non-human animal is generated that carries a transgene comprising a nucleic acid encoding the marker operably linked to a POMC nucleic acid sequence. The POMC nucleic acid sequence directs expression of the marker in POMC neurons in the arcuate nucleus of the non-human animal. The marker can be any marker, including, but not limited to, fluorescent markers (e.g., green fluorescent protein, *Aequoria victoria*, or *Discosoma* DSRed), antigenic markers (e.g., human growth hormone, human insulin, human HLA antigens), cell surface markers (e.g., CD4, or a any cell surface receptor), or enzymatic markers (e.g., lacZ).

The cDNA that encodes the marker can be fused in proper reading frame under the transcriptional and translational control of regulatory sequence of interest, such as a POMC regulatory sequence that directs expression of the marker in the POMC neurons of the arcuate nucleus. The sequences include, but are not limited to, the regulatory and coding sequences of the POMC gene, and suitable fragments thereof, wherein the regulatory and/or coding POMC sequence directs expression of

the marker in the POMC neurons of the arcuate nucleus. Specific, non-limiting examples of POMC sequences of use include, but are not limited to, transgenes carrying variable length or deletions of the 5' flanking sequences of a mammalian POMC gene, including, but not limited to, the mouse or human POMC gene. Specific, non-limiting examples of POMC sequences of use include, but are not limited to, murine, human, bovine, hamster, and rabbit POMC sequences. Variants of these POMC sequences, such as, but not limited to deletions, insertions, and additions are also of use, provided that these variants direct expression to the arcuate nucleus. In one embodiment, the POMC sequences can include the nPOMC1, nPOMC2 sequences, and/or the POMC promoter (see the Examples section below). Regions of homology for nPOMC1 and nPOMC2 are indicated in Fig. 10. In one embodiment, a POMC sequence of use includes the nPOMC1 and nPOMC2 and the POMC promoter, and is at least 70%, at least 80%, at least 90%, at least 95%, at least 98%, or at least 99% homologous with the corresponding originating POMC sequence. Additionally, such a sequence can include point mutation. Regions of homology for the human, cow, hamster, mouse, rabbit, and rat the nPOMC1 and nPOMC2 regions are shown in Fig. 10. One of skill in the art can readily use this information to design suitable sequences of interest. For example, conserved regions (shown in black) can be retained, while non-conserved regions (shown in grey or white) can be substituted. Additional exemplary sequences are described in the Examples section below.

This construct can be introduced into a vector to produce a product that is then amplified, for example, by preparation in a bacterial vector, according to conventional methods (see, for example, Sambrook et al., *Molecular Cloning: a Laboratory Manual*, Cold Spring Harbor Press, 1989). The amplified construct is thereafter excised from the vector and purified for use in producing transgenic animals.

Any animal can be of use in the methods disclosed herein, provided the animal is any non-human animal. A "non-human animal" includes, but is not limited to, a non-human primate, a farm animal such as swine, cattle, and poultry, a sport animal or pet such as dogs, cats, horses, hamsters, rodents, or a zoo animal such as lions, tigers, or bears. In one specific, non-limiting example, the non-human

animal is a transgenic animal, such as, but not limited to, a transgenic mouse, cow, sheep, or goat.

A transgenic animal contains cells that bear genetic information received, directly or indirectly, by deliberate genetic manipulation at the subcellular level, such as by microinjection or infection with recombinant virus, such that a recombinant DNA is included in the cells of the animal. This molecule can be integrated within the animal's chromosomes, or can be included as an extrachromosomally replicating DNA sequences, such as might be engineered into yeast artificial chromosomes. A transgenic animal can be a "germ cell line" transgenic animal, such that the genetic information has been taken up and incorporated into a germ line cell, therefore conferring the ability to transfer the information to offspring. If such offspring in fact possess some or all of that information, then they, too, are transgenic animals.

Transgenic animals can readily be produced by one of skill in the art. For example, transgenic animals can be produced by introducing into single cell embryos DNA encoding a marker, in a manner such that the polynucleotides are stably integrated into the DNA of germ line cells of the mature animal and inherited in normal Mendelian fashion. Advances in technologies for embryo micromanipulation permit introduction of heterologous DNA into fertilized mammalian ova. For instance, totipotent or pluripotent stem cells can be transformed by microinjection, calcium phosphate mediated precipitation, liposome fusion, retroviral infection or other means, the transformed cells are then introduced into the embryo, and the embryo then develops into a transgenic animal. In one non-limiting method, developing embryos are infected with a retrovirus containing the desired DNA, and transgenic animals produced from the infected embryo.

In another, specific, non-limiting example, the appropriate DNA(s) are injected into the pronucleus or cytoplasm of embryos, preferably at the single cell stage, and the embryos allowed to develop into mature transgenic animals. These techniques are well known. For instance, reviews of standard laboratory procedures for microinjection of heterologous DNAs into mammalian (mouse, pig, rabbit, sheep, goat, cow) fertilized ova include: Hogan et al., *Manipulating the Mouse Embryo*, Cold Spring Harbor Press, 1986; Krimpenfort et al., *Bio/Technology* 9:86,

1991; Palmiter et al., *Cell* 41:343, 1985; Kraemer et al., *Genetic Manipulation of the Early Mammalian Embryo*, Cold Spring Harbor Laboratory Press, 1985; Hammer et al., *Nature*, 315:680, 1985; Purcel et al., *Science*, 244:1281, 1986; Wagner et al., U.S. Patent No. 5,175,385; Krimpenfort et al., U.S. Patent No. 5,175,384.

A histological section of the arcuate nucleus from a non-human animal expressing a marker in the POMC neurons is prepared using methods known to one of skill in the art, and the section is contacted with a test agent of interest. An electrophysiological parameter of a POMC neuron is then assessed. Suitable electrophysiological parameters include, but are not limited to, hyperpolarization of the membrane potential of the POMC neuron and/or an increase in IPSCs in the POMC neuron. In one non-limiting example, an agonist is selected that causes hyperpolarization of the membrane potential of a POMC neuron, and increases IPSCs in a POMC neuron.

One of skill in the art can readily assesses neuron firing rate, membrane voltage, depolarization, action potentials, and IPSC frequency. Exemplary methods are described in the examples section below. However, the methods disclosed herein are not limited to the devices and measurements described in the Examples section. For example, any electrophysiology amplifier can be utilized, such as, but not limited to, devices produced by Dagan Instruments, Minneapolis, MN, or Heka Elektronik, Lambrecht/Pfalz, Germany.

In one embodiment, the membrane potential, action potential rate, and/or the frequency of IPSCs in a POMC neuron treated with an agent is compared to a control. Suitable controls include, but are not limited to, a section contacted with a buffer alone, in the absence of an agent, a sample contact with a control agent, such as an agent known to have an effect on the frequency of IPSCs, action potential rate, or to alter membrane potential of a POMC neuron, or an agent known not to have an effect on IPSCs, action potential rate, or membrane potential of a POMC neuron.

In one specific, non-limiting example, a section of the arcuate nucleus is contacted with an agent, and the effect on the membrane potential of a POMC neuron is measured. In this example, a change in the membrane potential of about 2 to about 50 mV indicates that the agent affects food intake, caloric intake, appetite, and/or energy expenditure. In another specific, non-limiting example, a change in

IPSC frequency is measured. In this example, a change in the IPSC frequency is measured. In this example, a change of the IPSC frequency from about 2% to a ten fold increase, or completely stopping IPSCs indicates that the agent affects food intake, caloric intake, appetite, and/or energy expenditure. In another embodiment, a change in the action potential rate of a POMC neuron is measured. In this example, a change in the action potential rate of about 2% to completely stopping, or a change in the action potential rate of greater than, or equal to, about a 1-fold, about a 2-fold, about a 20-fold, about a 50-fold, or about a 100-fold, increase indicates that the agent affects food intake, caloric intake, appetite, and/or energy expenditure. Alternatively, a change from no firing to activity of a POMC neuron indicates that the agent affects food intake, caloric intake, appetite, and/or energy expenditure. Other approaches to measuring activity includes, but not be limited to, an analysis of the expression of c-fos.

One of skill in the art can readily identify a statistically analysis of use in assessing data obtained from the methods disclosed herein. The statistical analyses are standard, such as tests for repeatability, for example analysis of variance, or wilcoxin signed rank test, are performed, using an appropriate confidence level, such as, but not limited to, $p<0.05$.

It should be noted that parameters of a POMC neuron, such as, but not limited to, ion fluxes (e.g., a potassium flux), enzyme activation (e.g., a serine/threonine kinase), changes in cyclic nucleotides (e.g., cAMP, cADP, cGMP, cGDP, etc.), among others, can also be measured. A specific, nonlimiting example of a signaling event is the generation of a K^+ flux following the interaction of an agent with a POMC neuron. A "physiological indicator," which is any compound in which a measurable property changes in a response to a physical parameter of the cell, can be used to measure the signaling event. One specific, non-limiting example of a measurable property is a change is in fluorescence of a physiological indicator in response to an ion flux.

Fluorescence is one spectral property of which can be used as the means of detecting a physiological parameter of a cell. A "fluorescent property" refers to the molar extinction coefficient at an appropriate excitation wavelength, the fluorescence quantum efficiency, the shape of the excitation spectrum or emission

spectrum, the excitation wavelength maximum and emission wavelength maximum, the ratio of excitation amplitudes at two different wavelengths, the ratio of emission amplitudes at two different wavelengths, the excited state lifetime, or the fluorescence anisotropy. A measurable difference in any one of these properties between a cell contacted with an agent as compared to a control cell suffices to identify a compound as being of interest. A measurable difference can be determined by determining the amount of any quantitative fluorescent property, e.g., the amount of fluorescence at a particular wavelength, or the integral of fluorescence over the emission spectrum. Optimally, the physiological indicator is selected to have fluorescent properties that are easily distinguishable. A specific, non-limiting example of a fluorescent indicator of use is fura-2. This dye measures intracellular calcium. Increased intracellular calcium is an indicator of increased neuronal activity, while decreased intracellular calcium is an indicator of decreased neural activity.

Any agent can be screened using the methods disclosed herein to determine if it affects appetite, food intake, caloric intake, and/or energy metabolism. Suitable test agents include, but are not limited to, agents that bind, or are suspected of binding a receptor on either a POMC neuron, or a NPY neuron, or both. Receptors on a POMC neuron include, but are not limited to a melanocortin receptor, a μ-opioid receptor, a leptin receptor, and an insulin receptor. Receptors on a NPY neuron include, but are not limited to, a Y2 receptor, a leptin receptor, an insulin receptor, a melanocortin receptor, or an opiod receptor. In one specific, non-limiting example the agent is a receptor agonist, or is suspected of being a receptor agonist. In another specific, non-limiting example, the agent is a Y2 receptor agonist, or is suspected of being a Y2 receptor agonist.

In one specific, non-limiting example, the agent is an antagonist for a receptor on an NPY neuron, or a POMC neuron. Thus, the agent can be, but is not limited to, an antagonist of a Y2 receptor. An electrophysiological property of the POMC neurons is measured. Increased activity of NPY neurons, measured as increased frequency of IPSCs in POMC neurons, hyperpolarization of POMC neurons, and/or a decrease in the action potential firing rate of POMC neurons indicates the antagonist is of use in increasing feeding behavior. Without being

bound by theory, antagonists, such as Y2 antagonists, can stimulate NPY neurons by reducing the tonic inhibition of those neurons mediated by the Y2 R and as such will be of use in treating anorexia and cachexia. Thus, the methods described herein can be use to screen for agents that increase appetite, food intake, caloric intake and decrease energy expenditure.

Agents that can be tested using the methods disclosed herein include polypeptides, chemical compounds; biological agents such as, but not limited to polypeptides, cytokines, and small molecules, peptidomimetics; antibodies; and synthetic ligands, amongst others. Receptor agonists and antagonists can be screened.

"Incubating" includes conditions that allow contact between the test compound and the histological section. "Contacting" includes in solution and solid phase. The test compound may also be a combinatorial library for screening a plurality of compounds. Compounds identified in the method of the invention can be further evaluated, detected, cloned, sequenced, and the like, either in solution of after binding to a solid support, by any method usually applied to the detection of a specific DNA sequence, such as PCR, oligomer restriction (Saiki et al., *Bio/Technology* 3:1008-1012, 1985), allele-specific oligonucleotide (ASO) probe analysis (Conner et al., *Proc. Natl. Acad. Sci. U.S.A.* 80:278, 1983), oligonucleotide ligation assays (OLAs) (Landegren et al., *Science* 241:1077, 1988), and the like. Molecular techniques for DNA analysis have been reviewed (Landegren et al., *Science* 242:229-237, 1988).

The binding affinities of receptor agonists (or antagonists) can also be determined in either cells or a membrane preparation expressing the receptor. For example, assays are utilized in which a labeled ligand is employed. A number of labels have been indicated previously (e.g., radiolabels, fluorescence labels, among others) to be of use. The candidate compound is added in an appropriate buffered medium. After an incubation to ensure that binding has occurred, the surface may be washed free of any nonspecifically bound components of the assay medium, particularly any nonspecifically bound labeled ligand, and any label bound to the

surface determined. The label may be quantitatively measured. By using standards, the relative binding affinity of a candidate compound can be determined.

Following screening using the methods disclosed herein, further testing can be performed, either in animal models or in clinical trials, to confirm that the agent affects food intake, caloric intake, appetite, or energy expenditure. Exemplary *in vivo* assays are described in the Examples section below. However, one of skill in the art can readily design alternative *in vivo* assays or clinical trials.

PYY Agonists

A PYY agonist can be screened using the methods disclosed herein, in order to determine if the PYY agonist will affect caloric intake, food intake, appetite, and/or energy metabolism. A PYY agonist is a molecule that binds to a receptor that specifically binds PYY, and elicits an effect of PYY. Suitable PYY agonists that can be screened using the methods disclosed herein include compounds that bind specifically in a Y receptor assay or competes for binding with PYY, such as in a competitive binding assay with labeled PYY. Suitable PYY agonists include, but are not limited to, compounds that bind to the Y2 receptor.

Thus, in one embodiment, a PYY agonist is selected using the methods disclosed herein that binds to a NPY neuron in the arcuate nucleus, and results in an electrophysiological effect on an NPY neuron. The electrophysiological effect on the NYP neuron can result in a further electrophysiological effect on a POMC neuron. Thus, one specific, non-limiting example, a PYY agonist is selected, using the methods disclosed herein, that causes depolarization of the membrane potential of a POMC neuron. In another specific, non-limiting example, a PYY agonist is selected using the method disclosed herein that causes an decrease in IPSCs in a POMC neuron, and/or an increased activity of a POMC neuron. In several non-limiting examples, agonists that cause hyperpolarization of the membrane potential of a POMC neuron, increase in IPSCs in a POMC neuron, are selected using the methods disclosed herein.

In one embodiment, these PYY agonists do not include NPY. In another specific, non-limiting example, a PYY agonist is tested using the methods disclosed herein that binds NPY neurons, but does not cross the blood/brain barrier. The

arcuate nucleus neurons upon which PYY exerts its effects are not protected by the blood/brain barrier, and thus are readily accessible to peripherally available molecules. In addition, other brain sites that express the Y2 receptor are protected by the blood/brain barrier. Without being bound by theory, agents able to bind to the arcuate Y2R, but that do not cross the blood/brain barrier following peripheral administration, are selected using the methods disclosed herein. In one embodiment, the ability of an agent to cross the blood brain barrier is assessed by the ability of the agent to induce the expression of c-fos in the arcuate nucleus following peripheral administration of the agent to a subject.

PYY and agonists that can be screened using the methods disclosed herein include, but are not limited to, polypeptides comprising, or alternatively consisting of, the amino acid sequence for PPY and agonists thereof, e.g., mutants, fragments and/or variants thereof. Variants include deletions, insertions, inversions, repeats and substitutions (e.g., conservative substitutions and non-conservative substitutions; see, e.g., Tables 1 and 2, *infra*). More than one amino acid (e.g., 2, 3, 4, 5, 6, 7, 8, 9, 10, etc.) can be deleted or inserted or substituted with another amino acid. Typically conservative substitutions are the replacements, one for another, among the aliphatic amino acids Ala, Val, Leu and Ile; interchange of Ser and Thr containing hydroxy residues, interchange of the acidic residues Asp and Glu, interchange between the amide residues Asn and Gln, interchange of the basic residues Lys and Arg, interchange of the aromatic residues Phe and Tyr, and interchange of the small-sized amino acids Ala, Ser, Thr, Met and Gly.

As another example, polypeptide fragments may contain a continuous series of deleted residues from the amino (N)- or the carboxyl (C)- terminus, or both (see, e.g., Tables 1 and 2, *infra*). Any number of amino acids, ranging from 1 to 24, can be deleted from the N-terminus, the C-terminus or both.

Furthermore, the agonist polypeptides that are screened using the methods disclosed herein, also include, but are not limited to, polypeptides comprising, or alternatively consisting of, internal deletions of the amino acid sequences for PPY and/or agonist thereof (see, e.g., Table 2, *infra*). Such deletions may comprise one or more amino acid residue deletions (e.g., one, two, three, four, five, six, seven, eight, nine, ten, etc.) and may begin at any amino acid position (e.g., two, three,

four, five, six, seven, eight, nine, ten, etc.). In addition, polypeptides can be screened that contain one or more such internal deletions. Such deletions are can be made in PPY, NPY and PP.

Also contemplated is the screening of agonist peptides that are PPY, NPY and/or PP chimeras having high affinity and/or selectivity for the Y2 receptor. These chimeras may comprise amino acid substitutions of one or more amino acids (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, etc.) from PPY, NPY and/or PP, variants, mutants and/or deletions thereof, with one or more amino acids (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, etc.) from a second PPY, NPY, or PP, variants, mutations and/or deletions thereof. These substitutions may begin at any amino acid position (e.g., two, three, four, five, six, seven, eight, nine, ten, etc.).

In one embodiment, the agents that are screened using the methods disclosed herein are selective for the Y2 receptor. That is, they bind with higher affinity to Y2 compared to other receptors, such as Y1, Y2, Y3, Y4, Y5 and Y6. In another embodiment, the peptides are selective for the Y2 and Y5 receptors over the Y1, Y3, Y4 and Y6 receptors.

Other polypeptide fragments that can be screened are fragments comprising structural or functional domain of the polypeptides of this disclosure. Such fragments include amino acid residues that comprise a polyproline-type II helix (residues 1-8), beta-turn (residues 9-14), amphipathic alpha-helix (residues 15-32) and/or a C-terminal turn structure (residues 33-36). See, Kirby et al., *J Med Chem* 36:385-393, 1993.

In addition, this disclosure includes the screening of a polypeptide or agonist comprising, or alternatively consisting of, the amino acid sequence for PPY, NPY and PP species variants (see Table 1, *infra*) and/or mutants, and fragments thereof.

Also contemplated is the screening of fusion proteins, whereby a PYY or PYY agonist will be fused to another protein or polypeptide (the fusion partner) using recombinant methods known in the art, to identify fusion proteins of use in reducing appetite, caloric intake, food intake, and/or energy expenditure. These fusion proteins can be synthetically synthesized by any known method. Any known peptide or protein can be used as the fusion partner (e.g., serum albumin, carbonic anhydrase, glutathione-S-transferase or thioredoxin, etc.). Such fusion proteins can

be designed linking the carboxy-terminus of the fusion partner to the amino-terminus of the PYY or agonist peptide, or vice versa. Optionally, a cleavable linker region can be used linking the PYY or PYY agonist to the fusion partner, and can be cleaved *in vivo* thereby resulting in the release of an active form of PYY or a PYY agonist. Examples of such cleavage regions include, but are not limited to, the linker regions D-D-D-D-Y (SEQ ID NO: 330), G-P-R (SEQ ID NO: 331), A-G-G (SEQ ID NO: 332) and H-P-F-H-L (SEQ ID NO 333), which can be cleaved by enterokinase, thrombin, ubiquitin cleaving enzyme and renin, respectfully. See, e.g., U.S. Patent No. 6,410,707.

Also contemplated is the screening of PYY agonists that Y2 specific peptide agonists as described in U.S. Patent No. 5,026,685; U.S. Patent No. 5,574,010; U.S. Patent No. 5,604,203; U.S. Patent No. 5,696,093; U.S. Patent No. 6,046,167.

PPY agonists that can be screened using the assays disclosed herein are described herein as follows.

TABLE 1 - PYY: Variation Among Species

PEPTIDE YY	AA SEQUENCE
Human	YPIKPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 1)
Rat	YPAKPEAPGEDASPEELSRYYASLRHYLNLVTRQRY (SEQ ID NO: 5)
Pig	YPAKPEAPGEDASPEELSRYYASLRHYLNLVTRQRY (SEQ ID NO: 6)
Guinea pig	YPSKPEAPGSDASPEELARYYASLRHYLNLVTRQRY (SEQ ID NO: 7)
Frog	YPPKPENPGEDASPEEMTKYLTALRHYINLVTRQRY (SEQ ID NO: 8)
Raja	YPPKPENPGDDAAPEELAKYYSALRHYINLITRQRY (SEQ ID NO: 9)
Dogfish	YPPKPENPGEDAPPEELAKYYSALRHYINLITRQRY (SEQ ID NO: 10)
Lampetra	FPPKPDNPGDNASPEQMARYKAAVRHYINLITRQRY (SEQ ID NO: 11)
Petromyzon	MPPKPDNPSPDASPEELSKYMLAVRNYINLITRQRY (SEQ ID NO: 12)

NEUROPEPTIDE Y	AA SEQUENCE
Human	YPSKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 2)
Rat	YPSKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 13)
Rabbit	YPSKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 14)
Dog	YPSKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 15)
Pig	YPSKPDNPGEDAPAEDLARYYSALRHYINLITRQRY (SEQ ID NO: 16)

	Cow	YPSKPDNPGEDAPAEDLARYYSALRHYINLITRQRY (SEQ ID NO: 17)
	Sheep	YPSKPDNPGDDAPAEDLARYYSALRHYINLITRQRY (SEQ ID NO: 18)
	Guinea pig	YPSKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 19)
	Avian	YPSKPDSPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 20)
5	Rana	YPSKPDNPGEDAPAEDMAKYYSALRHYINLITRQRY (SEQ ID NO: 21)
	Goldfish	YPTKPDNPGEGAPAEELAKYYSALRHYINLITRQRY (SEQ ID NO: 22)
	Dogfish	YPSKPDNPGEGAPAEDLAKYYSALRHYINLITRQRY (SEQ ID NO: 23)
	Lampetra	PPNKPDSPGEDAPAEDLARYLSAVRHYINLITRQRY (SEQ ID NO: 24)

10	PANCREATIC POLYPEPTIDE	AA SEQUENCE
	Human	ASLEPEYPGDNATPEQMAQYAAELRRYINMLTRPRY (SEQ ID NO: 3)
	Sheep	APLEPVYPGDNATPEQMAQYAADLRRYINMLTRPRY (SEQ ID NO: 25)
	Pig	APLEPVYPGDDATPEQMAQYAAELRRYINMLTRPRY (SEQ ID NO: 26)
	Dog	APLEPVYPGDDATPEQMAQYAAELRRYINMLTRPRY (SEQ ID NO: 27)
15	Cat	APLEPVYPGDNATPEQMAQYAAELRRYINMLTRPRY (SEQ ID NO: 28)
	Cow	APLEPEYPGDNATPEQMAQYAAELRRYINMLTRPRY (SEQ ID NO: 29)
	Rat	APLEPMYPGDYATHEQRAQYETQLRRYINTLTRPRY (SEQ ID NO: 30)
	Mouse	APLEPMYPGDYATPEQMAQYETQLRRYINTLTRPRY (SEQ ID NO: 31)
	Guinea pig	APLEPVYPGDNATPEQQMAQYAAEMRRYINMLTRPRY (SEQ ID NO: 32)
20	Chicken	GPSQPTYPGDDAPVEDLIRFYNDLQQYLNVVTRHRY (SEQ ID NO: 33)
	Alligator	TPLQPKYPGDGAPVEDLIQFYNDLQQYLNVVTRPRF (SEQ ID NO: 34)
	Bullfrog	APSEPHHPGDQATPDQLAQYYSDLYQYITFITRPRF (SEQ ID NO: 35)

Ref: Beck-Sickinger, A.G., Jung, G., *Biopolymers 37:123-142, 1995.*

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TABLE 2 – PEPTIDE AGONIST OF PYY

PEPTIDE	SEQUENCE
PPY(3-36)(human)	
30	IKPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 1)

Ref: Eberlein et al., *Peptides* 10:797-803, 1989; Grandt et al., *Peptides* 15(5):815-20, 1994.

Variations of PPY(3-36)

35 N-Terminal Deletions of PYY, including but not limited to: PYY(26-36), PYY(25-36), PYY(24-36), PYY(23-36), PYY(22-36), PYY(21-36), PYY(20-36), PYY(19-36), PYY(18-36), PYY(17-36), PYY(16-36), PYY(15-36), PYY(14-36), PYY(13-

36), PYY(12-36), PYY(11-36), PYY(10-36), PYY(9-36), PYY(8-36), PYY(7-36), PYY(6-36), PYY(5-36), PYY(4-36), PYY(3-36).

Ref: See, e.g., Balasubramaniam et al., *Pept Res* 1(1):32-5, Sep-Oct 1998; Liu et al., *J Gastrointest Surg* 5(2):147-52, Mar-Apr 2001.

PEPTIDE SEQUENCE

NPY (human)

YPSKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 2)

Ref: Tatemoto et al., *Proc Natl Acad Sci U.S.A.* 79:5485-9, 1982.

Variations of NPY

N-Terminal Deletions of NPY, including but not limited to: NPY(26-36), NPY(25-36), NPY(24-36), NPY(23-36), NPY(22-36), NPY(21-36), NPY(20-36), NPY(19-36), NPY(18-36), NPY(17-36), NPY(16-36), NPY(15-36), NPY(14-36), NPY(13-36), NPY(12-36), NPY(11-36), NPY(10-36), NPY(9-36), NPY(8-36), NPY(7-36), NPY(6-36), NPY(5-36), NPY(4-36), NPY(3-36).

Ref: See e.g., Gehlert et al., *Proc Soc Exp Biol Med* 218:7-22, 1998; Sheikh et al., *Am J Physiol* 261:G701-15, Nov. 1991.

Internal Deletions, including but not limited to: (1-4)-Aca-(14-36)pNPY, (1-4)-Aca-(15-36)pNPY, (1-4)-Aca-(16-36)pNPY, (1-4)-Aca-(17-36)pNPY, (1-4)-Aca-(18-36)pNPY, (1-4)-(31-36)pNPY11, (1-4)-Aca-(31-36)pNPY, (4-1)-(31-36)pNPY, (4-1)-Aca-(31-36)pNPY, $(4\text{-}1)_D$-(31-36)pNPY, $(4\text{-}1)_D$-Aca-(31-36)pNPY.

Ref: Fournier et al., *Mol Pharmacol* 45(1):93-101, Jan 1994.

Additional Internal Deletion Mutants, including but not limited to: des-$AA^{10\text{-}17}$-NPY, des-$AA^{10\text{-}17}$, Ac-[D-Lys^9(ε-Ac-Ala)]NPY, des-$AA^{10\text{-}17}$, Ac[D-Lys^9(ε-Ac-Ala)]NPY, des-$AA^{10\text{-}17}$[$Ala^{7,21}$]NPY, des-$AA^{10\text{-}17}$[$Cys^{7,21}$]NPY, des-$AA^{10\text{-}17}$[Glu^7,Lys^{21}]NPY, des-$AA^{11\text{-}17}$[D-Lys^{10}(ε-Ac), $Cys^{7,21}$]NPY, des-$AA^{10\text{-}17}$[D-Cys^7, D-Lys(ε-Ac), Cys^{21}]NPY,des-$AA^{10\text{-}17}$[D-Cys^7, Lys^9(ε-Ac), Cys^{21}]NPY, des-$AA^{10\text{-}17}$[$Cys^{7,21}$, Pro^{34}]NPY, des-$AA^{10\text{-}17}$[Asp^7, Dpr^{21}, Pro^{34}]NPY, des-$AA^{10\text{-}17}$[Glu^7, Lys^{21}, Pro^{34}]NPY, des-$AA^{10\text{-}17}$[$Cys^{7,21}$, Leu^{31}, Pro^{34}]NPY, des-$AA^{10\text{-}20}$[$Cys^{7,21}$, Pro^{34}]NPY, des-$AA^{10\text{-}17}$[$Cys^{2,27}$]NPY, des-$AA^{10\text{-}17}$[Cys^2, D-Cys^{27}]NPY.

Ref: Kirby et al., *J Med Chem* 38:4579-86, 1995.

Cyclic agonist of NPY, including but not limited to: [Lys 25-Glu 29]NPY(Ac-25-36), [Glu 25-Lys 29]NPY(Ac-25-36), [Lys 26-Glu31]NPY(Ac-25-36), [Glu 27-Lys 31]NPY(Ac-25-36), [Lys28-Glu 32]NPY(Ac-25-36), [Lys27-Glu34]NPY(Ac-25-36).

Ref: Rist et al., *Eur J Biochem* 247:1019-1028, 1997.

D-amino acid substitutions: [D-Tyr^1]NPY, [D-Pro^2]NPY, [D-Ser^3]NPY, [D-Lys^4]NPY, [D-Pro^5]NPY, [D-Asp^6]NPY, [D-Asn^7]NPY, [D-Pro^8]NPY, [D-Ala^9]NPY, [D-Glu^{10}]NPY, [D-Asp^{11}]NPY, [D-Ala^{12}]NPY, [D-Pro^{13}]NPY, [D-Ala^{14}]NPY, [D-Glu^{15}]NPY, [D-Asp^{16}]NPY, [D-Leu^{17}]NPY, [D-Ala^{18}]NPY, [D-Arg^{19}]NPY, [D-Tyr^{20}]NPY, [D-Tyr^{21}]NPY, [D-Ser^{22}]NPY, [D-Ala^{23}]NPY, [D-Leu^{24}]NPY, [D-Arg^{25}]NPY, [D-His^{26}]NPY, [D-Tyr^{27}]NPY, [D-Ile^{28}]NPY, [D-Asn^{29}]NPY, [D-Leu^{30}]NPY, [D-Ile^{31}]NPY, [D-Thr^{32}]NPY, [D-Arg^{33}]NPY, [D-Gln^{34}]NPY, [D-Arg^{35}]NPY, [D-Tyr^{36}]NPY, [D-Tyr^1, D-Pro^2]NPY, [D-Ser^3, D-Lys^4]NPY, [D-Pro^5, D-Asp^6]NPY, [D-Asn^7, D-Pro^8]NPY, [D-Glu^{10}, D-Asp^{11}]NPY, [D-Asp^{11}, D-Ala^{12}]NPY, [D-Pro^{13}, D-Ala^{14}]NPY, [D-Glu^{15}, D-Asp^{16}]NPY, [D-Met^{17}, D-Ala^{18}]NPY, [D-Arg^{19}, D-Tyr^{20}]NPY, [D-Tyr^{21}, D-Ser^{22}]NPY, [D-Ala^{23}, D-Leu^{24}]NPY, [D-Arg^{25}, D-His^{26}]NPY, [D-Tyr^{27}, D-Ile^{28}]NPY, [D-Asn^{29}, D-Leu^{30}]NPY, [D-Ile^{31}, D-Thr^{32}]NPY, [D-Arg^{33}, D-Gln^{34}]NPY, [D-Arg^{35}, D-Tyr^{36}]NPY.

Ref: Kirby et al., *J Med Chem* 36:3802-08, 1993; Grundemar et al., *Regulatory Peptides* 62:131-136, 1996.

Other NPY Agonist and Analogs

PEPTIDE	SEQUENCE
NPY(3-36)	SKPDNPGEDAPAEDMARYYSALRHYINLITRQRY (SEQ ID NO: 5)

Ref: Grandt et al., *Regulatory Peptides* 67(1):33-7, 1996.

PEPTIDE SEQUENCE

N-Acetyl NPY(24-36)

LRHYINLITRQRY (SEQ ID NO: 213)

Ref: Potter et al., *Eur J Pharmacol* 267(3):253-262, May 17, 1994.

PEPTIDE SEQUENCE

N-Acetyl [Leu^{28}, Leu^{31}] NPY(24-36)

LRHYLNLLTRQRY (SEQ ID NO: 214)

Ref: Potter et al., *Eur J Pharmacol* 267(3):253-262, May 17, 1994.

PEPTIDE SEQUENCE

[Leu^{28}, Leu^{31}] NPY(24-36)

LRHYLNLLTRQRY (SEQ ID NO: 215)

Ref: Potter et al., *Eur J Pharmacol* 267(3):253-262, May 17, 1994.

PEPTIDE SEQUENCE

[Leu^{17}, Gln^{19}, Ala^{21}, Ala^{22}, Glu^{23}, Leu^{28}, Leu^{31}] NPY(13-36)

PAEDLAQYAAELRIIYLNLLTRQRY (SEQ ID NO: 216)

Ref: Potter et al., Eur J Pharmacol 267(3):253-262, May 17, 1994.

PEPTIDE SEQUENCE

Cyclo S-S [Cys^{20},Cys^{24}]pNPY

SKPDNPGEDAPAEDMARCYSACRHYINLITRQRY (SEQ ID NO: 315)

Ref: Soll et al., *Eur J Biochem* 268(10):2828-37, May 2001.

PEPTIDE SEQUENCE

Cyclo-(28/32)-Ac-[Lys^{28}-Glu^{32}]-(25-36)-pNPY

RHYLNLIGRQRY (SEQ ID NO: 316)

Ref: Cabrele et al., *J Pept Sci* 6(3):97-122, Mar 2000.

PEPTIDE SEQUENCE

Cyclo-(27/31)-Ac-[Glu^{27}-Lys^{31}]-(25-36)-pNPY

RHGLNLLGRQRY (SEQ ID NO: 317)

Ref: Cabrele et al., *J Pept Sci* 6(3):97-122, Mar 2000.

PEPTIDE SEQUENCE

[Tyr^{32}, Leu^{34}]NPY(27-36)

YINLIYRLRY (SEQ ID NO: 318)

Ref: Leban et al., *J Med Chem* 38:1150-57, 1995.

PEPTIDE SEQUENCE

[Tyr^{32}, Leu^{34}]NPY(26-36)

HYINLIYRLRY (SEQ ID NO: 319)

Ref: Leban et al., *J Med Chem* 38:1150-57, 1995.

PEPTIDE SEQUENCE

[Tyr^{32}, Leu^{34}]NPY(25-36)

RHYINLIYRLRY (SED ID NO: 320)

Ref: Leban et al., *J Med Chem* 38:1150-57, 1995.

[Leu^{31}]NPY(27-36)

YINLLYRQRY (SEQ ID NO: 321)

Ref: Leban et al., *J Med Chem* 38:1150-57, 1995.

PEPTIDE SEQUENCE

[Tyr^{32}, Leu^{34}] (1-4)-Ahr-(27-36)NPY

YPSL-Aha-YINLIYRLRY (SED ID NO: 322)

Ref: Leban et al., *J Med Chem* 38:1150-57, 1995.

PEPTIDE SEQUENCE

[Tyr^{32}, Leu^{34}]NPY(28-36)

INLIYRLRY (SEQ ID NO: 323)

Ref: Leban et al., *J Med Chem* 38:1150-57, 1995.

PEPTIDE SEQUENCE

PP (human)

ASLEPEYPGDNATPEQMAQYAAELRRYINMLTRPRY (SEQ ID NO: 3)

Ref: Kimmel et al., *Endocrinology* 83:1323-30, 1968.

Variations of PP

N-Terminal Deletions including but not limited to: PP(26-36), PP(25-36), PP(24-36), PP(23-36), PP(22-36), PP(21-36), PP(20-36), PP(19-36), PP(18-36), PP(17-36), PP(16-36), PP(15-36), PP(14-36), PP(13-36), PP(12-36), PP(11-36), PP(10-36), PP(9-36), PP(8-36), PP(7-36), PP(6-36), PP(5-36), PP(4-36), PP(3-36).

TABLE 3 – EXAMPLES OF CONSERVATIVE AMINO ACID SUBSTITUTIONS OF PYY

Single point mutations of PYY(25-36)

PEPTIDE	SEQUENCE
[Lys^{25}]PPY(25-36)	KHYLNLVTRQRY (SEQ ID NO: 36)
[Thr^{27}]PPY(25-36)	RHTLNLVTRQRY (SEQ ID NO: 37)
[Phe^{27}]PPY(25-36)	RHFLNLVTRQRY (SEQ ID NO: 38)
[Ile^{28}]PYY (25-36)	RHYINLVTRQRY (SEQ ID NO: 39)
[Val^{28}]PYY (25-36)	RHYVNLVTRQRY (SEQ ID NO: 40)
[Gln^{29}]PYY (25-36)	RHYLQLVTRQRY (SEQ ID NO: 41)
[Ile^{30}]PYY (25-36)	RHYLNIVTRQRY (SEQ ID NO: 42)
[Val^{30}]PYY (25-36)	RHYLNVVTRQRY (SEQ ID NO: 43)
[Ile^{31}]PYY (25-36)	RHYLNLITRQRY (SEQ ID NO: 44)
[Leu^{31}]PYY (25-36)	RHYLNLLTRQRY (SEQ ID NO: 45)
[Ser^{32}]PYY (25-36)	RHYLNLVSRQRY (SEQ ID NO: 46)
[Lys^{33}]PYY (25-36)	RHYLNLVTKQRY (SEQ ID NO: 47)
[Asn^{34}]PYY (25-36)	RHYLNLVTRNRY (SEQ ID NO: 48)
[Lys^{35}]PYY (25-36)	RHYLNLVTRQKY (SEQ ID NO: 49)
[Thr^{36}]PYY (25-36)	RHYLNLVTRQRT (SEQ ID NO: 50)
[Phe^{36}]PYY (25-36)	RHYLNLVTRQRF (SEQ ID NO: 51)

Double point mutations

PEPTIDE	SEQUENCE
[Lys^{25}, Thr^{27}]PPY(25-36)	KHTLNLVTRQRY (SEQ ID NO: 52)
[Lys^{25}, Phe^{27}]PPY(25-36)	KHFLNLVTRQRY (SEQ ID NO: 53)
[Lys^{25}, Ile^{28}]PPY(25-36)	KHYINLVTRQRY (SEQ ID NO: 54)
[Lys^{25}, Val^{28}]PPY(25-36)	KHYVNLVTRQRY (SEQ ID NO: 55)
[Lys^{25}, Gln^{29}]PPY(25-36)	KHYLQLVTRQRY (SEQ ID NO: 56)

[Lys^{25}, Ile^{30}]PPY(25-36)	KHYLNIVTRQRY (SEQ ID NO: 57)
[Lys^{25}, Val^{30}]PPY(25-36)	KHYLNVVTRQRY (SEQ ID NO: 58)
[Lys^{25}, Ile^{31}]PPY(25-36)	KHYLNLITRQRY (SEQ ID NO: 59)
[Lys^{25}, Leu^{31}]PPY(25-36)	KHYLNLLTRQRY (SEQ ID NO: 60)
[Lys ^{25}, Ser ^{32}]PPY(25-36)	KHYLNLVSRQRY (SEQ ID NO: 61)
[Lys^{25}, Lys^{33}]PPY(25-36)	KHYLNLVTKQRY (SEQ ID NO: 62)
[Lys^{25}, Asn^{34}]PPY(25-36)	KHYLNLVTRNRY (SEQ ID NO: 63)
[Lys^{25}, Lys^{35}]PPY(25-36)	KHYLNLVTRQKY (SEQ ID NO: 64)
[Lys^{25}, Thr^{36}]PPY(25-36)	KHYLNLVTRQRT (SEQ ID NO: 65)
[Lys^{25}, Phe^{36}]PPY(25-36)	KHYLNLVTRQRF (SEQ ID NO: 66)
[Thr^{27}, Ile^{28}]PPY(25-36)	RHTINLVTRQRY (SEQ ID NO: 67)
[Thr^{27}, Val^{28}]PPY(25-36)	RHTVNLVTRQRY (SEQ ID NO: 68)
[Thr^{27}, Gln^{29}]PPY(25-36)	RHTLQLVTRQRY (SEQ ID NO: 69)
[Thr^{27}, Ile^{30}]PPY(25-36)	RHTLNIVTRQRY (SEQ ID NO: 70)
[Thr^{27}, Val^{30}]PPY(25-36)	RHTLNVVTRQRY (SEQ ID NO: 71)
[Thr^{27}, Ile^{31}]PPY(25-36)	RHTLNLITRQRY (SEQ ID NO: 72)
[Thr^{27}, Leu^{31}]PPY(25-36)	RHTLNLLTRQRY (SEQ ID NO: 73)
[Thr^{27}, Ser^{32}]PPY(25-36)	RHTLNLVSRQRY (SEQ ID NO: 74)
[Thr^{27}, Lys^{33}]PPY(25-36)	RHTLNLVTKQRY (SEQ ID NO: 75)
[Thr^{27}, Asn^{34}]PPY(25-36)	RHTLNLVTRNRY (SEQ ID NO: 76)
[Thr^{27}, Lys^{35}]PPY(25-36)	RHTLNLVTRQKY (SEQ ID NO: 77)
[Thr^{27}, Thr^{36}]PPY(25-36)	RHTLNLVTRQRT (SEQ ID NO: 78)
[Thr^{27}, Phe^{36}]PPY(25-36)	RHTLNLVTRQRF (SEQ ID NO: 79)
[Phe^{27}, Ile^{28}]PPY(25-36)	RHFINLVTRQRY (SEQ ID NO: 80)
[Phe^{27}, Val^{28}]PPY(25-36)	RHFVNLVTRQRY (SEQ ID NO: 81)
[Phe^{27}, Gln^{29}]PPY(25-36)	RHFLQLVTRQRY (SEQ ID NO: 82)
[Phe^{27}, Ile^{30}]PPY(25-36)	RHFLNIVTRQRY (SEQ ID NO: 83)
[Phe^{27}, Val^{30}]PPY(25-36)	RHFLNVVTRQRY (SEQ ID NO: 84)
[Phe^{27}, Ile^{31}]PPY(25-36)	RHFLNLITRQRY (SEQ ID NO: 85)
[Phe^{27}, Leu^{31}]PPY(25-36)	RHFLNLLTRQRY (SEQ ID NO: 86)
[Phe^{27}, Ser^{32}]PPY(25-36)	RHFLNLVSRQRY (SEQ ID NO: 87)
[Phe^{27}, Lys^{33}]PPY(25-36)	RHFLNLVTKQRY (SEQ ID NO: 88)
[Phe^{27}, Asn^{34}]PPY(25-36)	RHFLNLVTRNRY (SEQ ID NO: 89)
[Phe^{27}, Lys^{35}]PPY(25-36)	RHFLNLVTRQKY (SEQ ID NO: 90)
[Phe^{27}, Thr^{36}]PPY(25-36)	RHFLNLVTRQRT (SEQ ID NO: 91)
[Phe^{27}, Phe^{36}]PPY(25-36)	RHFLNLVTRQRF (SEQ ID NO: 92)
[Gln^{29}, Ile^{30}]PYY (25-36)	RHYLQIVTRQRY (SEQ ID NO: 93)
[Gln^{29}, Val^{30}]PYY (25-36)	RHYLQVVTRQRY (SEQ ID NO: 94)
[Gln^{29}, Ile^{31}]PYY (25-36)	RHYLQLITRQRY (SEQ ID NO: 95)

[Gln^{29}, Leu^{31}]PYY (25-36)	RHYLQLLTRQRY (SEQ ID NO: 96)
[Gln^{29}, Ser^{32}]PYY (25-36)	RHYLQLVSRQRY (SEQ ID NO: 97)
[Gln^{29}, Leu^{33}]PYY (25-36)	RHYLQLVTKQRY (SEQ ID NO: 98)
[Gln^{29}, Asn^{34}]PYY (25-36)	RHYLQLVTRNRY (SEQ ID NO: 99)
[Gln^{29}, Leu^{35}]PYY (25-36)	RHYLQLVTRQKY (SEQ ID NO: 100)
[Gln^{29}, Thr^{36}]PYY (25-36)	RHYLQLVTRQRT (SEQ ID NO: 101)
[Gln^{29}, Phe^{36}]PYY (25-36)	RHYLQLVTRQRF (SEQ ID NO: 102)
[Ile^{30}, Ile^{31}]PYY (25-36)	RHYLNIITRQRY (SEQ ID NO: 103)
[Ile^{30}, Leu^{31}]PYY (25-36)	RHYLNILTRQRY (SEQ ID NO: 104)
[Ile^{30}, Ser^{32}]PYY (25-36)	RHYLNIVSRQRY (SEQ ID NO: 105)
[Ile^{30}, Lys^{33}]PYY (25-36)	RHYLNIVTKQRY (SEQ ID NO: 106)
[Ile^{30}, Asn^{34}]PYY (25-36)	RHYLNIVTRNRY (SEQ ID NO: 107)
[Ile^{30}, Lys^{35}]PYY (25-36)	RHYLNIVTRQKY (SEQ ID NO: 108)
[Ile^{30}, Thr^{36}]PYY (25-36)	RHYLNIVTRQRT (SEQ ID NO: 109)
[Ile^{30}, Phe^{36}]PYY (25-36)	RHYLNIVTRQRF (SEQ ID NO: 110)
[Val^{30}, Ile^{31}]PYY (25-36)	RHYLNVITRQRY (SEQ ID NO: 111)
[Val^{30}, Leu^{31}]PYY (25-36)	RHYLNVLTRQRY (SEQ ID NO: 112)
[Val^{30}, Ser^{32}]PYY (25-36)	RHYLNVVSRQRY (SEQ ID NO: 113)
[Val^{30}, Lys^{33}]PYY (25-36)	RHYLNVVTKQRY (SEQ ID NO: 114)
[Val^{30}, Asn^{34}]PYY (25-36)	RHYLNVVTRNRY (SEQ ID NO: 115)
[Val^{30}, Lys^{35}]PYY (25-36)	RHYLNVVTRQKY (SEQ ID NO: 116)
[Val^{30}, Thr^{36}]PYY (25-36)	RHYLNVVTRQRT (SEQ ID NO: 117)
[Val^{30}, Phe^{36}]PYY (25-36)	RHYLNVVTRQRF (SEQ ID NO: 118)
[Ile^{31}, Ser^{32}]PYY (25-36)	RHYLNLISRQRY (SEQ ID NO: 119)
[Ile^{31}, Lys^{33}]PYY (25-36)	RHYLNLITKQRY (SEQ ID NO: 120)
[Ile^{31}, Asn^{34}]PYY (25-36)	RHYLNLITRNRY (SEQ ID NO: 121)
[Ile^{31}, Lys^{35}]PYY (25-36)	RHYLNLITRQKY (SEQ ID NO: 122)
[Ile^{31}, Thr^{36}]PYY (25-36)	RHYLNLITRQRT (SEQ ID NO: 123)
[Leu^{31}, Phe^{36}]PYY (25-36)	RHYLNLITRQRF (SEQ ID NO: 124)
[Leu^{31}, Ser^{32}]PYY (25-36)	RHYLNLLSRQRY (SEQ ID NO: 125)
[Val^{31}, Lys^{33}]PYY (25-36)	RHYLNLLTKQRY (SEQ ID NO: 126)
[Leu^{31}, Asn^{34}]PYY (25-36)	RHYLNLLTRNRY (SEQ ID NO: 127)
[Leu^{31}, Lys^{35}]PYY (25-36)	RHYLNLLTRQKY (SEQ ID NO: 128)
[Leu^{31}, Thr^{36}]PYY (25-36)	RHYLNLLTRQRT (SEQ ID NO: 129)
[Leu^{31}, Phe^{36}]PYY (25-36)	RHYLNLLTRQRF (SEQ ID NO: 130)
[Ser^{32}, Lys^{33}]PYY (25-36)	RHYLNLVSKQRY (SEQ ID NO: 131)
[Ser^{32}, Asn^{34}]PYY (25-36)	RHYLNLVSRNRY (SEQ ID NO: 132)
[Ser^{32}, Lys^{35}]PYY (25-36)	RHYLNLVSRQKY (SEQ ID NO: 133)
[Ser^{32}, Thr^{36}]PYY (25-36)	RHYLNLVSRQRT (SEQ ID NO: 134)

[Ser^{32}, Phe^{36}]PYY (25-36)	RHYLNLVSRQRY (SEQ ID NO: 135)
[Lys^{33}, Asn^{34}]PYY (25-36)	RHYLNLVTKNRY (SEQ ID NO: 136)
[Lys^{33}, Lys^{35}]PYY (25-36)	RHYLNLVTKQKY (SEQ ID NO: 137)
[Lys^{33}, Thr^{36}]PYY (25-36)	RHYLNLVTKQRT (SEQ ID NO: 138)
[Lys^{33}, Phe^{36}]PYY (25-36)	RHYLNLVTKQRF (SEQ ID NO: 139)
[Asn^{34}, Lys^{35}]PYY (25-36)	RHYLNLVTRNKY (SEQ ID NO: 140)
[Asn^{34}, Thr^{36}]PYY (25-36)	RHYLNLVTRNRT (SEQ ID NO: 141)
[Asn^{34}, Phe^{36}]PYY (25-36)	RHYLNLVTRNRF (SEQ ID NO: 142)
[Lys^{35}, Thr^{36}]PYY (25-36)	RHYLNLVTRQKT (SEQ ID NO: 143)
[Lys^{35}, Phe^{36}]PYY (25-36)	RHYLNLVTRQKF (SEQ ID NO: 144)

Point Mutations of PYY(24-36)

PEPTIDE	SEQUENCE
PYY(24-36)	LRHYLNLVTRQRY (SEQ ID NO: 145)
[Ile^{24}]PYY(24-36)	IRHYLNLVTRQRY (SEQ ID NO: 146)
[Val^{24}]PYY(24-36)	VRHYLNLVTRQRY (SEQ ID NO: 147)

Also included as PYY(24-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of any of these three mutants with any of the above listed mutants for PYY(25-36), e.g., [Lys^{25}]PPY(24-36) (Amino acid sequence=LKHYLNLVTRQRY (SEQ ID NO: 191)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 145.

Point Mutations of PYY(23-36)

PEPTIDE	SEQUENCE
PYY(23-36)	SLRHYLNLVTRQRY (SEQ ID NO: 148)
[Thr^{23}]PYY(23-36)	TLRHYLNLVTRQRY (SEQ ID NO: 149)

Also included as PYY(23-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(24-36), e.g., [Lys^{25}]PPY(23-36) (Amino acid sequence=SLKHYLNLVTRQRY (SEQ ID NO: 192)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 148.

Point Mutations of PYY(22-36)

PEPTIDE	SEQUENCE
PYY(22-36)	ASLRHYLNLVTRQRY (SEQ ID NO: 150)
[Ser^{22}]PYY(22-36)	SSLRHYLNLVTRQRY (SEQ ID NO: 151)

Also included as PYY(22-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(23-36), e.g., [Lys^{25}]PPY(22-36) (Amino acid sequence=ASLKHYLNLVTRQRY (SEQ ID NO: 193)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 150.

Point Mutations of PYY(21-36)

PEPTIDE	SEQUENCE
PYY(21-36)	YASLRHYLNLVTRQRY (SEQ ID NO: 152)
[Thr^{21}]PYY(21-36)	TASLRHYLNLVTRQRY (SEQ ID NO: 153)
[Phe^{21}]PYY(21-36)	FASLRHYLNLVTRQRY (SEQ ID NO: 154)

Also included as PYY(21-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of any of these three mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(22-36), e.g., [Lys^{25}]PPY(21-36) (Amino acid sequence=YASLKHYLNLVTRQRY (SEQ ID NO: 194)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 152.

Point Mutations of PYY(20-36)

PEPTIDE	SEQUENCE
PYY(20-36)	YYASLRHYLNLVTRQRY (SEQ ID NO: 155)
[Thr^{20}]PYY(20-36)	TYASLRHYLNLVTRQRY (SEQ ID NO: 156)
[Phe^{20}]PYY(20-36)	FYASLRHYLNLVTRQRY (SEQ ID NO: 157)

Also included as PYY(20-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of any of these three mutants with any of the above listed mutants for PYY(25-36), and/or any of the

above listed mutants for PYY(21-36), e.g., [Lys^{25}]PPY(20-36) (Amino acid sequence=YYASLKHYLNLVTRQRY (SEQ ID NO: 195)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 155.

Point Mutations of PYY(19-36)

PEPTIDE	SEQUENCE
PYY(19-36)	RYYASLRHYLNLVTRQRY (SEQ ID NO: 158)
[Lys^{19}]PYY(19-36)	KYYASLRHYLNLVTRQRY (SEQ ID NO: 159)

Also included as PYY(19-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(20-36), e.g., [Lys^{25}]PPY(19-36) (Amino acid sequence=RYYASLKHYLNLVTRQRY (SEQ ID NO: 196)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 158.

Point Mutations of PYY(18-36)

PEPTIDE	SEQUENCE
PYY(18-36)	NRYYASLRHYLNLVTRQRY (SEQ ID NO: 160)
[Gln^{18}]PYY(18-36)	QRYYASLRHYLNLVTRQRY (SEQ ID NO: 161)

Also included as PYY(18-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(19-36), e.g., [Lys^{25}]PPY(18-36) (Amino acid sequence=NRYYASLKHYLNLVTRQRY (SEQ ID NO: 197)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 160.

Point Mutations of PYY(17-36)

PEPTIDE	SEQUENCE
PYY(17-36)	LNRYYASLRHYLNLVTRQRY (SEQ ID NO: 162)
[Ile^{17}]PYY(17-36)	INRYYASLRHYLNLVTRQRY (SEQ ID NO: 163)
[Val^{17}]PYY(17-36)	VNRYYASLRHYLNLVTRQRY (SEQ ID NO: 164)

Also included as PYY(17-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of any of these three mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(18-36), e.g., [Lys^{25}]PPY(17-36) (Amino acid sequence=LNRYYASLKHYLNLVTRQRY (SEQ ID NO: 198)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 162.

Point Mutations of PYY(16-36)

PEPTIDE	SEQUENCE
PYY(16-36)	ELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 165)
[Asp^{16}]PYY(16-36)	DLNRYYASLRHYLNLVTRQRY (SEQ ID NO: 166)

Also included as PYY(16-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(17-36), e.g., [Lys^{25}]PPY(16-36) (Amino acid sequence=ELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 199)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 165.

Point Mutations of PYY(15-36)

PEPTIDE	SEQUENCE
PYY(15-36)	EELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 167)
[Asp^{15}]PYY(15-36)	DELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 168)

Also included as PYY(15-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(16-36), e.g., [Lys^{25}]PPY(15-36) (Amino acid sequence=EELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 200)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 167.

Point Mutations of PYY(14-36)

PEPTIDE	SEQUENCE
PYY(14-36)	PEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 169)

Also included as PYY(14-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of this PYY(14-36) mutant with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(15-36), e.g., [Lys^{25}]PPY(23-36) (Amino acid sequence=PEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 201) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 169.

Point Mutations of PYY(13-36)

PEPTIDE	SEQUENCE
PYY(13-36)	SPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 170)
[Thr^{13}]PYY(13-36)	TPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 171)

Also included as PYY(13-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(14-36), e.g., [Lys^{25}]PPY(13-36) (Amino acid sequence=SEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 202)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 170.

Point Mutations of PYY(12-36)

PEPTIDE	SEQUENCE
PYY(12-36)	ASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 172)
[Ser^{12}]PYY(12-36)	SSPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 173)

Also included as PYY(12-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(13-36), e.g., [Lys^{25}]PPY(12-36) (Amino acid sequence=ASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 203)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 172.

Point Mutations of PYY(11-36)

PEPTIDE	SEQUENCE
PYY(11-36)	DASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 174)
[Glu^{11}]PYY(11-36)	EASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 175)

Also included as PYY(12-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(12-36), e.g., [Lys^{25}]PPY(11-36) (Amino acid sequence=DASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 204)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 174.

Point Mutations of PYY(10-36)

PEPTIDE	SEQUENCE
PYY(10-36)	EDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 176)
[Asp^{10}]PYY(10-36)	DDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 177)

Also included as PYY(10-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(11-36), e.g., [Lys^{25}]PPY(10-36) (Amino acid sequence=EDASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 205)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 176.

Point Mutations of PYY(9-36)

PEPTIDE	SEQUENCE
PYY(9-36)	GEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 178)

Also included as PYY(9-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of this PPY(9-36) mutant with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(10-36), e.g., [Lys^{25}]PPY(9-36) (Amino acid sequence=GEDASPEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 206))

would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 178.

Potin Mutations of PYY(8-36)

PEPTIDE	SEQUENCE
PYY(8-36)	PGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 179)

Also included as PYY(8-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of this PPY(8-36) mutant with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(9-36), e.g., [Lys^{25}]PPY(8-36) (Amino acid sequence= SEQ ID NO: 207)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 179.

Point Mutations of PYY(7-36)

PEPTIDE	SEQUENCE
PYY(7-36)	APGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 180)
[Ser^{9}]PYY(7-36)	SPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 181)

Also included as PYY(7-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(8-36), e.g., [Lys^{25}]PPY(7-36) (Amino acid sequence=APGEDASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 208)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 180.

Point Mutations of PYY(6-36)

PEPTIDE	SEQUENCE
PYY(6-36)	EAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 182)
[Asp^{6}]PYY(6-36)	DAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 183)

Also included as PYY(6-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of either of these two mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(7-36), e.g., [Lys^{25}]PPY(6-36) (Amino acid sequence=EAPGEDASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 209)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 182.

Point Mutations of PYY(5-36)

PEPTIDE	SEQUENCE
PYY(5-36)	PEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 184)

Also included as PYY(5-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of this PPY(5-36) mutant with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(6-36), e.g., [Lys^{25}]PPY(5-36) (Amino acid sequence=PEAPGEDASPEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 210)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 184.

Point Mutations of PYY(4-36)

PEPTIDE	SEQUENCE
PYY(4-26)	KPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 185)
[Arg^4]PYY(4-36)	RPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 186)
[Gln^4]PYY(4-36)	QPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 187)
[Asn^4]PYY(4-36)	NPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 188)

Also included as PYY(4-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of any of these four mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(5-36), e.g., [Lys^{25}]PPY(4-36) (Amino acid sequence=KPEAPGEDASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 211))

would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 185.

Point Mutations of PYY(3-36)

PEPTIDE	*SEQUENCE*
PYY(3-36)	IKPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 1)
[Leu^3]PYY(3-36)	LKPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 189)
[Val^3] PYY(3-36)	VKPEAPGEDASPEELNRYYASLRHYLNLVTRQRY (SEQ ID NO: 190)

Also included as PYY(3-36) mutations are polypeptide variations (amino acid sequence variations) resulting from the combination of any of these three mutants with any of the above listed mutants for PYY(25-36), and/or any of the above listed mutants for PYY(4-36), e.g., [Lys^{25}]PPY(3-36) (Amino acid sequence=IKPEAPGEDASEELNRYYASLKHYLNLVTRQRY (SEQ ID NO: 212)) would result from combining the mutations from SEQ ID NO: 36 with SEQ ID NO: 1.

Also contemplated are PYY agonists (NPY analogs) having the formula:

X-Q-R_{19} -R_{20} -R_{21} -R_{22} -R_{23} -Leu-R_{25} -R_{26} -R_{27} -R_{28} -R_{29} -R_{30} -R_{31} -R_{32} -Arg-R_{34} -Arg-R_{36} -Y

wherein X is H or C^a Me or N^a Me or desamino or an acyl group having 7 carbon atoms or less; Q is R_{17} -R_{18}, R_{18} or desQ; R_{17} is Met, Arg, Nle, Nva, Leu, Ala or D-Ala; R_{18} is Ala, Ser, Ile, D-Ala, D-Ser or D-Ile; R_{19} is Arg, Lys or Gln; R_{20} is Tyr or Phe; R_{21} is Tyr, Glu, His or Ala; R_{22} is Ser, Ala, Thr, Asn or Asp; R_{23} is Ala, Asp, Glu, Gln, Asn or Ser; R_{25} is Arg or Gln; R_{26} is His, Arg or Gln; R_{27} is Phe or Tyr; R_{28} is Ile, Leu, Val or Arg; R_{29} is Asn or Ile; R_{30} is Leu, Met, Thr or Val; R_{31} is Ile, Val or Leu; R_{32} is Thr or Phe; R_{34} is Gln, Pro or His; R_{36} is Phe or Tyr; and Y is NH_2 or OH; provided that when Q is R_{18}, then at least one of R_{27} and R_{36} is Phe. Analogs of NPY have the following applications: potent postsynaptic treatment of hypertension and cardiogenic shock, the treatment of acute cardiovascular

circulatory failure, and the elevation of intracellular calcium. See U.S. Patent No. 5,026,685.

Certain preferred NPY analogs have the formula: X-R_{18}-Arg-Tyr-Tyr-R_{22}-R_{23}-Leu-Arg-His-Tyr-R_{28}-Asn-Leu-R_{31}-Thr-Arg-Gln-Arg-Tyr-NH_2, wherein X is H or C^a Me or N^a Me or desamino or an acyl group having 7 carbon atoms or less; R_{18} is Ala or Ser; R_{22} is Ser or Ala; R_{23} is Ala or Ser; R_{27} is Phe or Tyr; R_{28} is Ile or Leu; R_{31} is Ile or Val; and R_{36} is Phe or Tyr; provided that at least one of R_{27} and R_{36} is Phe. See U.S. Patent No. 5,026,685.

Other contemplated NPY analogs have the formula:

X-R_{17} -R_{18} -Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-R_{27}-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-R_{36}-NH_2,

wherein R_{17} is Arg or Leu and R_{18} is Ser or Ala or Ile; and wherein X, R_{27} and R_{36} are as previously indicated.

Still other preferred NPY analogs have the formula:

X-R_{18}-Arg-Tyr-Tyr-Ala-Ser-Leu-R_{25}-His-R_{27}-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-R_{36}-NH_2,

wherein X is desamino or C^a Me or N^a Me and wherein R_{18}, R_{25}, R_{27} and R_{36} are as previously indicated.

Examples of such NPY agonists include:

pNPY (17-36) having the formula:
H-Leu-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 217)

The peptide hNPY (17-36) having the formula: H-Met-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 218)

The peptide [Phe^{27}]-NPY (18-36) having the formula: H-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Phe-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 219)

The peptide [Ac-D-Ala^{17}]-NPY (17-36) having the formula: Ac-D-Ala-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 220)

The peptide NPY (19-36) having the formula: H-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 221)

The peptide [Nle^{17}]-NPY (17-36) having the formula: H-Nle-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 222)

The peptide [D-Ser^{18}]-NPY (18-36) having the formula: H-D-Ser-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 223)

The peptide [Ala^{17}, His^{21}]-NPY (17-36) having the formula: H-Ala-Ala-Arg-Tyr-His-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 224)

The peptide [D-Ile^{18}]-NPY (18-36) having the formula: D-Ile-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 225)

The peptide [Ac-Arg^{17}]-NPY (17-36) having the formula: Ac-Arg-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 226)

The peptide [Gln^{19}]-NPY (19-36) having the formula: H-Gln-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 227)

The peptide [Phe^{20}]-NpY (18-36) having the formula: H-Ala-Arg-Phe-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 228)

The peptide [C^a MeLeu^{17}]-pNPY (17-36) having the formula: H-C^a MeLeu-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 229)

The peptide [N^a MeLeu^{17}]-pNPY (17-36) having the formula: H-N^a MeLeu-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln- Arg-Tyr-NH_2 (SEQ ID NO: 230)

The peptide [desamino Ala^{18}]-NpY (18-36) having the formula: desamino-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr- NH_2 (SEQ ID NO: 231)

The peptide [For-Ala^{18}, Glu^{23}, Arg^{26}]-NPY (18-36) having the formula: For-Ala-Arg-Tyr-Tyr-Ser-Glu-Leu-Arg-Arg-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 232)

The peptide [Nva^{17}, Ala^{21}, Leu^{28}]-NPY (17-36) having the formula: H-Nva-Ala-Arg-Tyr-Ala-Ser-Ala-Leu-Arg-His-Tyr-Leu-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 233)

The peptide [Thr^{22}, Gln^{23}]-NPY (18-36) having the formula: H-Ala-Arg-Tyr-Tyr-Thr-Gln-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 234)

The peptide [desamino Leu^{17}, Asn^{23}, Val^{30}]-NPY (17-36) having the formula: H-desamino Leu-Ala-Arg-Tyr-Tyr-Ser-Asn-Leu-Arg-His-Tyr-Ile-Asn-Val-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 235)

The peptide [Asp^{22}, Ser^{23}, Thr^{30}]-NPY (18-36) having the formula: H-Ala-Arg-Tyr-Tyr-Asp-Ser-Leu-Arg-His-Tyr-Ile-Asn-Thr-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 236)

The peptide [Gln^{25}, Leu^{31}, Pro^{34}]-NPY (18-36) having the formula: H-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Gln-His-Tyr-Ile-Asn-Leu-Leu-Thr-Arg-Pro-Arg-Tyr-NH_2 (SEQ ID NO: 237)

The peptide [Gln^2 Phe^{36}]-NPY (17-36) having the formula: H-Leu-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-Gln-Tyr-Arg-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Phe-NH_2 (SEQ ID NO: 238)

The peptide [Phe^{36}]-pPYY (19-36) having the formula: H-Arg-Tyr-Tyr-Ala-Ser-Leu-Arg-His-Tyr-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-Phe-NH_2 (SEQ ID NO: 239)

The peptide pPYY (18-36) having the formula: H-Ser-Arg-Tyr-Tyr-Ala-Ser-Leu-Arg-His-Tyr-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 240)

The peptide [Ac-Ser^{18}, Phe^{27}]-pPYY (I8-36) having the formula: Ac-Ser-Arg-Tyr-Tyr-Ala-Ser-Leu-Arg-His-Phe-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 241)

The peptide [Nle^{17}, Asn^{22}, Phe^{27}]-NPY (17-36) having the formula: H-Nle-Ala-Arg-Tyr-Tyr-Asn-Ala-Leu-Arg-His-Phe-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 242)

The peptide [D-Ala^{18}, Glu^{21}, His^{34}]-NPY (18-36) having the formula: H-D-Ala-Arg-Tyr-Glu-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-His-Arg-Tyr-NH_2 (SEQ ID NO: 243)

The peptide [Bz-Leu^{17}, Pro^{34}, Phe^{36}]-pNPY (17-36) having the formula: Bz-Leu-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Pro-Arg-Phe-NH_2 (SEQ ID NO: 244)

The peptide [Lys^{19}, Phe^{27}, Val^{28}]-NpY (18-36) having the formula: H-Ala-Lys-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Phe-Val-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 245)

The peptide [D-Ala^{17}, Val^{28}, Phe^{32}]-NPY (17-36) having the formula: D-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Val-Asn-Leu-Ile-Phe-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 246)

The peptide [C^{a} MeSer^{18}, Met^{30}, Phe^{36}]-NPY (18-36) having the formula: H-C^{a} MeSer-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Met-Ile-Thr-Arg-Gln-Arg-Phe-NH_2 (SEQ ID NO: 247)

The peptide [Arg^{17}, Ile^{18}, Phe^{27,36}]-NPY (17-36) having the formula: H-Arg-Ile-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Phe-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Phe-NH_2 (SEQ ID NO: 248)

The peptide [Ser^{18}, Phe^{27}]-pNPY (17-36) having the formula: H-Leu-Ser-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Phe-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 249)

The peptide [N^{a} MeIle^{18}, Gln^{25}, Phe^{27}]-NPY (18-36) having the formula: N^{a} MeIle-Arg-Tyr-Tyr-Ser-Ala-Leu-Gln-His-Phe-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 250)

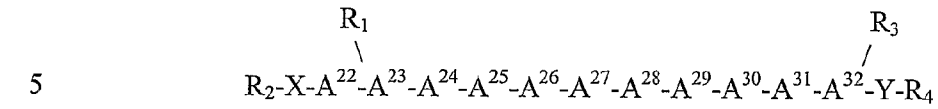
The peptide [D-Ser^{18}, Phe^{36}]-NPY (18-36) having the formula: H-D-Ser-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Phe-NH_2 (SEQ ID NO: 251)

The peptide [Asp^{23}, Arg^{26}]hNPY (17-36) having the formula: H-Met-Ala-Arg-Tyr-Tyr-Ser-Asp-Leu-Arg-Arg-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 252)

The peptide [Glu^{23}, Ile^{29}]-NPY (18-36) having the formula: H-Ala-Arg-Tyr-Tyr-Ser-Glu-Leu-Arg-His-Tyr-Ile-Ile-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 253)

The peptide [D-Ala^{17}]-NPY(17-36)-OH having the formula: D-Ala-Ala-Arg-Tyr-Tyr-Ser-Ala-Leu-Arg-His-Tyr-Ile-Asn-Leu-Ile-Thr-Arg-Gln-Arg-Tyr-OH (SEQ ID NO: 254).

Other peptide YY agonists have the formula:



wherein:

X is a chain of 0-5 amino acids, inclusive, the N-terminal one of which is bonded to R_1 and R_2

Y is a chain of 0-4 amino acids, inclusive, the C-terminal one of which is bonded to R_3 and R_4

R_1 is H, C_1 -C_2 alkyl (e.g., methyl), C_6 -C_{18} aryl (e.g., phenyl, napthaleneacetyl), C_1 --C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7 -C_{18} aralkyl (e.g., benzyl), or C_7 -C_{18} alkaryl (e.g., p-methylphenyl);

R_2 is H, C_1 -C_{12} alkyl (e.g., methyl), C_6 -C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1 -C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7 -C_{18} aralkyl (e.g., benzyl), or C_7 -C_{18} alkaryl (e.g., p-methylphenyl);

A^{22} is an aromatic amino acid, Ala, Aib, Anb, N-Me-Ala, or is deleted;

A^{23} is Ser, Thr, Ala, N-Me-Ser, N-Me-Thr, N-Me-Ala, or is deleted;

A^{24} is Leu, lie, Vat, Trp, Gly, Aib, Anb, N-Me-Leu, or is deleted;

A^{25} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1 -C_{10} alkyl group, or an aryl group), Orn, or is deleted;

A^{26} is His, Thr, 3-Me-His, 1-Me-His, β-pyrozolylalanine, N-Me-His, Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1 -C_{10} alkyl group, or an aryl group), Orn, or is deleted;

A^{27} is an aromatic amino acid other than Tyr;

A^{28} is Leu, Ile, Vat, Trp, Aib, Aib, Anb, or N-Me-Leu;

A^{29} is Asn, Ala, Gln, Gly, Trp, or N-Me-Asn;

A^{30} is Leu, Ile, Val, Trp, Aib, Anb, or N-Me-Leu;

A^{31} is Vat, Ile, Trp, Aib, Anb, or N-Me-Val;

A^{32} is Thr, Ser, N-Me-Set, or N-Me-Thr;

R_3 is H, C_1 -C_{12} alkyl (e.g., methyl), C_6 -C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1 -C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7 -C_{18} aralkyl (e.g., benzyl), or C_7 -C_{18} alkaryl (e.g., p-methylphenyl);

R_4 is H, C_1 -C_{12} alkyl (e.g., methyl), C_6 -C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1 -C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7 -C_{18} aralkyl (e.g., benzyl), or C_7 -C_{18} alkaryl (e.g., p-methylphenyl), or a pharmaceutically acceptable salt thereof. See U.S. Patent No. 5,574,010.

Particularly preferred agonists of this formula to be used in the method of the disclosure include:
N-α-Ala-Ser-Leu-Arg-His-Trp-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ ID NO: 255).

Other peptide YY agonists have the formula:

$$R_2\text{-}A^{25}\text{-}A^{26}\text{-}A^{27}\text{-}A^{28}\text{-}A^{29}\text{-}A^{30}\text{-}A^{31}\text{-}A^{32}\text{-}Y\text{-}R_4$$
(R_1 and R_3 are attached to the N-terminal amino acid)

wherein:

the N-terminal amino acid bonds to R_1 and R_2;

Y is a chain of 0-4 amino acids, inclusive the C-terminal one of which bonds to R3 and R4;

R_1 is H, C_1 -C_{12} alkyl, C_6 -C_{18} aryl, C_1 -C_{12} acyl, C_7 -C_{18} aralkyl, or C_7 -C_{18} alkaryl;

R_2 is H, C_1 -C_{12} alkyl, C_6 -C_{18} aryl, C_1 -C_{12} acyl, C_7 -C_{18} aralkyl, or C_7 -C_{18} alkaryl;

A^{25} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1 -C_{10} alkyl group, or an aryl group), Orn, or is deleted;

A^{26} is Ala, His, Thr, 3-Me-His, 1-Me-His, β-pyrozolylalanine, N-Me-His, Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1 -C_{10} alkyl group, or an aryl group), Orn or is deleted;

A^{27} is an aromatic amino acid;

A^{28} is Leu, Ile, Val, Trp, Aib, Anb, or N-Me-Leu;

A^{29} is Asn, Ala, Gln, Gly, Trp, or N-Me-Asn;

A^{30} is Leu, Ile, Val, Trp, Aib, Anb, or N-Me-Leu;

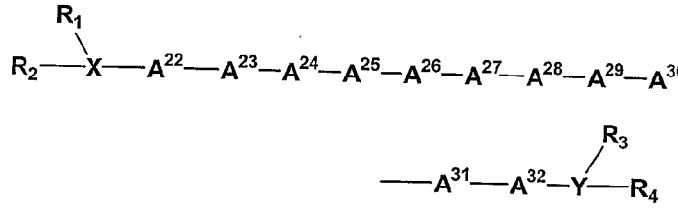
A^{31} is Val, Ile, Trp, Aib, Anb, or N-Me-Val;

A^{32} is Thr, Set, N-Me-Set, or N-Me-Thr or D-Trp;

R_3 is H, C_1-C_{12} alkyl, C_6 -C_{18} aryl, C_1 -C_{12} acyl, C_7 -C_{18} aralkyl, or C_7 -C_{18} alkaryl; and

R_4 is H, C_1 -C_{12} alkyl, C_6 -C_{18} aryl, C_1 -C_{12} acyl, C_7 -C_{18} aralkyl, or C_7 -C_{18} alkaryl, or a pharmaceutically acceptable salt thereof. Note that, unless indicated otherwise, for all peptide YY agonists described herein, each amino acid residue, e.g., Leu and A^1, represents the structure of NH--C(R)H--CO--, in which R is the side chain. Lines between amino acid residues represent peptide bonds which join the amino acids. Also, where the amino acid residue is optically active, it is the L-form configuration that is intended unless D-form is expressly designated.

Other PYY agonists have the formula:



wherein:

X is a chain of 0-5 amino acids, inclusive, the N-terminal one of which is bonded to R_1 and R_2;

Y is a chain of 0-4 amino acids, inclusive, the C-terminal one of which is bonded to R_3 and R_4;

R_1 is H, C_1-C_{12} alkyl (e.g. methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl);

R_2 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl);

A^{22} is an aromatic amino acid, Ala, Aib, Anb, N-Me-Ala, or is deleted;

A^{23} is Ser, Thr, Ala, Aib, N-Me-Ser, N-Me-Thr, N Me-Ala, or is deleted;

A^{24} is leu, Ile, Val, Trp, Gly, Nle, Nva, Aib, Anb, N-Me-Leu, or is deleted;

A^{25} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lye-e-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or an aryl group), Orn, or is deleted;

A^{26} is Ala, His, Thr, 3-Me-His, 1-Me-His, β-pyrozolylalanine, N-Me-His, Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl groups or an aryl group), Orn, or is deleted;

A^{27} is an aromatic amino acid other than Tyr;

A^{28} is Leu, Ile, Val, Trp, Nle, Nva, Aib, Anb, or N-Me-Leu;

A^{29} is Asn, Ala, Gin, Gly, Trp, or N-Me-Asn;

A^{30} is Leu, Ile, Val, Trp, Nle, Nva, Aib, Anb, or N-Me-Leu;

A^{31} is Val, Leu, Ile, Trp, Nle, Nva, Aib, Anb, or N-Me-Val;

A^{32} is Thr, Ser, N-Me-Ser, N-Me-Thr, or D-Trp;

R_3 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl); and

R_4 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl), or a pharmaceutically acceptable salt thereof.

In preferred embodiments, A^{27} is Phe, Nal, Bip, Pcp, Tic, Trp, Bth, Thi, or Dip.

In preferred embodiments X is A^{17}-A^{18}-A^{19}-A^{20}-A^{21} wherein

A^{17} is Cys, Leu, Ile, Val, Nle, Nva, Aib, Anb, or N-Me-Leu;

A^{18} is Cys, Ser, Thr, N-Me-Ser, or N-Me-Thr;

A^{19} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or C_6-C_{18} aryl group), Cys, or Orn;

A^{20} is an aromatic amino acid, or Cys; and

A^{21} is an aromatic amino acid, Cys, or a pharmaceutically acceptable salt thereof. In yet other preferred embodiments, Y is A^{33}-A^{34}-A^{35}-A^{36} wherein

A^{33} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or an aryl group), Cys, or Orn;

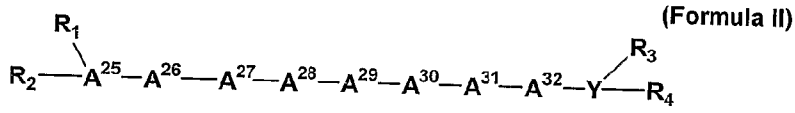
A^{34} is Cys, Gln, Asn, Ala, Gly, N-Me-Cln, Aib, or Anb;

A^{35} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or C_6-C_{18} aryl group), Cys, or Orn; and

A^{36} is an aromatic amino acid, Cys or a pharmaceutically acceptable salt thereof. See U.S. Patent No. 5,604,203.

Particular embodiments include compounds has the formula: N-α-Ac-Ala-Ser-Leu-Arg-His-Phe-Leu-Asn-Leu-Val-Thr-Arg-Gin-Arg-Tyr-NH_2 (SEQ. ID. NO: 325), H-Ala-Ser-Leu-Arg-His-Phe-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ. ID. NO: 326), N-α-Ac-Ala-Ser-Leu-Arg-Thr-Arg-Gin-Arg-Tyr-NH_2 (SEQ. ID. NO: 327), N-α-Ac-Ala-Ser-Leu-Arg-His-Thi-Leu-Asn-Leu-Val-Thr-Arg-Gin-Arg-Tyr-NH_2 (SEQ. ID. NO: 328), N-α-Ac-Tyr-Ser-Leu-Arg-His-Phe-Leu-Asn-Leu-Val-Thr-Arg-Gin-Arg-Tyr-NH_2 (SEQ. ID. NO: 329) or a pharmaceutically acceptable salt thereof.

Other PYY agonists have the formula:



wherein the N-terminal amino acid is bounded to R_1 and R_2; Y is a chain of 0-4 amino acids, inclusive the C-terminal one of which is bonded to R_3 and R_4;

R_1 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl);

R_2 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl);

A^{25} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or an aryl group), Orn, or is deleted;

A^{26} is Ala, His, Thr, 3-Me-His, 1-Me-His, β-pyrozolylalanine, N-Me-His, Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl groups or an aryl group), Orn, or is deleted;

A^{27} is an aromatic amino acid;

A^{28} is Leu, Ile, Val, Trp, Nle, Nva, Aib, Anb, or N-Me-Leu;

A^{29} is Asn, Ala, Gin, Gly, Trp, or N-Me-Asn;

A^{30} is Leu, Ile, Val, Trp, Nle, Nva, Aib, Anb, or N-Me-Leu;

A^{31} is Val, Ile, Trp, Nle, Nva, Aib, Anb, or N-Me-Val;

A^{32} is Thr, Ser, N-Me-Ser, N-Me-Thr, or D-Trp;

R_3 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl); and

R_4 is H, C_1-C_{12} alkyl (e.g., methyl), C_6-C_{18} aryl (e.g., phenyl, naphthaleneacetyl), C_1-C_{12} acyl (e.g., formyl, acetyl, and myristoyl), C_7-C_{18} aralkyl (e.g., benzyl), or C_7-C_{18} alkaryl (e.g., p-methylphenyl), or a pharmaceutically acceptable salt thereof. See U.S. Patent No. 5,604,203.

In particular embodiments, A^{27} is Phe, Nal, Bip, Pcp, Tic, Trp, Bth, Thi, or Dip.

In particular embodiments X is A^{33}-A^{34}-A^{35}-A^{36} wherein

A^{33} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or C_6-C_{18} aryl group), Cys, or Orn;

A^{34} is Gln, Asn, Ala, Gly, N-Me-Gin, Aib, Cys, or Anb;

A^{35} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R (where R is H, a branched or straight chain C_1-C_{10} alkyl group, or C_6-C_{18} aryl group), Cys, or Orn; and

A^{36} is an aromatic amino acid, Cys, or a pharmaceutically acceptable salt thereof.

Preferably, the compound has the formula: N-α-Ac-Arg-His-Phe-Leu-Asn-Leu-Val-Thr-Arg-Gln-Arg-Tyr-NH_2 (SEQ. ID. NO: 324).

Examplary PYY agonists include:

YPAKEAPGEDASPEELSTYYASLR [im-DNP-His^{26}] YLNLVTRZRY-NH_2	(SEQ ID NO: 256)
PYY(22-36) ASLRHYLNLVTRQRY-NH_2	(SEQ ID NO: 257)
[Ala^{32}]PYY ASLRHYLNLV[Ala]RQRY-NH_2	(SEQ ID NO: 258)
[$Ala^{23,32}$]PYY A[Ala]LRHYLNLV[Ala]RQRY-NN_2	(SEQ ID NO: 259)
[Glu^{28}]PYY(22-36) ASLRHY[Glu]NLVTRQRY-NH_2	(SEQ ID NO: 260)

N-α-Ac-PYY(22-36) N-α-Ac-ASLRHYLNLVTRORY-NH_2	(SEQ ID NO: 261)
N-α-Ac[p.CL.Phe^{26}]PYY N-α-Ac-ASLR[p.CL.Phe^{26}]YLNLVTRQRY-NH_2	(SEQ ID NO: 262)
N-α-Ac[Glu^{28}]PYY N-α-Ac-ASLRHY[Glu]NLVTRQRY-NH_2	(SEQ ID NO: 263)
N-α-Ac[Phe^{27}]PYY N-α-Ac-ASLRH[Phe]ENLVTRQR[N-Me-Tyr]-NH_2	(SEQ ID NO: 264)
N-α-Ac]8N-Me-Tyr^{36}]PYY N-α-Ac-ASLRHYENLVTR0R[N-Me-Tyr]-NH_2	(SEQ ID NO: 265)
N-α-myristoyl-PYY(2214 36) N-α-myristoyl-ASLRHYLNLVTRQRY-NH_2	(SEQ ID NO: 266)
N-α-naphthateneacetyl-PYY(22-36) N-α- naphthateneacetyl-ASLRHYLNLVTRQRY-NH_2	(SEQ ID NO: 267)
N-α-Ac[Phe^{27}]PYY N-α-Ac-ASLRH[Phe]ENLVTR0R[N-Me-Tyr]-NH_2	(SEQ ID NO: 268)
N-α-Ac-PYY (22-36) N-α-Ac-ASLRHYLNLVTRQRY-NH_2	(SEQ ID NO: 269)
N-α-Ac-[Bth^{27}]PYY (22-36) N-α-Ac-ASLRH[Bth]LNLVTRQRY-NH_2	(SEQ ID NO: 270)
N-α-Ac-[Bip^{27}]PYY (22-36) N-α-Ac-ASLRH[Bth]LNLVTRQRY-NH_2	(SEQ ID NO: 271) (SEQ ID NO: 272)
N-α-Ac-[Nal^{27}]PYY (22-36) N-α-Ac-ASLRH[Bth]LNLVTRQRY-NH_2	(SEQ ID NO: 273)
N-α-Ac-[Trp^{27}]PYY (22-36) N-α-Ac-ASLRH[Trp]LNLVTRQRY-NH_2	(SEQ ID NO: 274) (SEQ ID NO: 275)
N-α-Ac-[Thi^{27}]PYY (22-36) N-α-Ac-ASLRN[Thi]LNLVTRQRY-NH_2	(SEQ ID NO: 276)
N-α-Ac-[Tic^{27}]PYY (22-36) N-α-Ac-ASLRH[Tic]LNLVTRQRY-NH_2	(SEQ ID NO: 277)
N-α-Ac-[Phe^{27}]PYY (25-36) N-α-Ac-H[Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 279)
N-α-Ac-[Phe^{27},Thi^{27}]PYY (22-36) N-α-Ac-ASLRH[Phe]LNLVTRQR[Thi]-NH_2	(SEQ ID NO: 280)
N-α-Ac-[Thz^{26},Phe^{27}]PYY (22-36) N-α-Ac-ASLRH[Thz][Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 281)
N-α-Ac-[Phe^{27}]PYY (22-36) N-α-Ac-ASLRH[Thz][Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 282)
N-α-Ac-[Phe^{27}]PYY (22-36) N-α-Ac-[Phe]SLRN[Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 289)
N-α-Ac-[Tyr^{22},Phe^{27}]PYY (22-36) N-α-Ac-[Tyr]SLRH[Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 290)
N-α-Ac-[Trp^{28}]PYY (22-36) N-α-Ac-ASLRHY[Trp]NLVTRQRY-NH_2	(SEQ ID NO: 291)
N-α-Ac-[Trp^{28}]PYY (22-36)	

N-α-Ac-ASLRHYLN[Trp]VTRQRY-NH_2	(SEQ ID NO: 292)
N-α-Ac-[Ala^{26},Phe^{27}]PYY (22-36) N-α-Ac-ASLR[Ala][Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 293)
N-α-Ac-[Bth^{27}]PYY (22-36) N-α-Ac-ASLR[Bth]LNLVTRQRY-NH_2	(SEQ ID NO: 294)
N-α-Ac-[Phe^{27}]PYY (22-36) N-α-Ac-ASLRH[Phe]LNLVTRQRY-NH_2	(SEQ ID NO: 295)
N-α-Ac-[$Phe^{27,36}$]PYY (22-36) N-α-Ac-ASLRH[Phe]LNLVTRQR[Phe]-NH_2	(SEQ ID NO: 296)
N-α-Ac-[Phe^{27}, D-Trp^{32}]PYY (22-36) N-α-Ac-ASLRH[Phe]LNLV[D-Trp]RQRY-NH_2	(SEQ ID NO: 297)

Other PYY agonists include neurophilic Y Y2 receptor specific peptides having the formula:

$X1(-X2-X3-X4-X5-X6-X7-X8-X9-X10-X11-X12-X13-X14)_n-X15$

wherein

X1 is NH, CH_3CO or one or two naturally occurring amino acids.

X2 is Leu, Ile or Val.

X3 is Arg, Lys or His.

X4 is His, Lys or Arg.

X5 is Tyr or Phe.

X6 is Leu, Ile or Val.

X7 is Asn or Gln.

X8 is Leu, Ile or Val.

X9 is Leu, Ile or Val.

X10 is Thr or Ser.

X11 is Arg, His or Lys.

X12 is Gln or Asn.

X13 is Arg, His or Lys.

X14 is Tyr or Phe.

X15 is COOH, NH_2 or one or two naturally occurring amino acids with the terminal amino acid being in the normal or carboxamide form; and

n is 1 to 5. See U.S. Patent No. 5,696,093.

Examplary agonists include:

CH_3CO-L-R-H-Y-L-N-L-L-T-R-Q-R-Y-NH_2 (SEQ ID NO: 298)

CH_3CO-L-R-H-Y-I-N-L-I-T-R-Q-R-Y-NH_2 (SEQ ID NO: 299)

NH_2-L-R-H-Y-L-N-L-L-T-R-Q-R-Y-NH_2 (SEQ ID NO: 300)

NH_2-L-R-H-Y-I-N-L-I-T-R-Q-R-Y-NH_2 (SEQ ID NO: 301)

Other PYY agonists have the formula:

N-α-R^1 -[$Nle^{24,28,30}$, Trp^{27}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2,

N-α-R^1 -[$Nle^{24,28}$, $Trp^{27,30}$, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2,

N-α-R^1 -[$Nle^{24,28,30}$, Phe^{27}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2,

N-α-R^1 -[$Nle^{24,28}$, Phe^{27}, Trp^{30}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2,

N-α-R^1 -[Trp^{30}, $\psi^{35/36}$]PYY(25-36)-NH_2,

N-α-R^1 -[Trp^{30}]PYY(25-36)-NH_2,

N-α-R^1 -[$Nle^{24,28}$, Trp^{30}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2 and

N-α-R^1 -[Nle^{28}, Trp^{30}, Nva^{31}, $\psi^{35/35}$]PYY(22-36)-NH_2 or a pharmaceutically-acceptable salt thereof,

wherein R^1 is H, (C_1 -C_{12})alkyl or (C_1 -C_{12})acyl; and

ψ is a pseudopeptide bond selected from the group consisting of --CH_2 --NH--, --CH_2 --S--, --CH_2 --CH_2 --, --CH_2 --O-- and --CH_2 --CO--. See U.S. Patent No. 6,046,162.

Particular compounds of the immediately foregoing group of compounds are where R^1 is acetyl and ψ is --CH_2 --NH--.

A particular group of compounds is selected from a group consisting of

N-α-Ac-[$Nle^{24,28,30}$, Trp^{27}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2, (SEQ ID NO: 302)

N-α-Ac-[$Nle^{24,28}$, $Trp^{27,30}$, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2, (SEQ ID NO: 303)

N-α-Ac-[$Nle^{24,28,30}$, Phe^{27}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2, (SEQ ID NO: 304)

N-α-Ac-[$Nle^{24,28}$, Phe^{27}, Trp^{30}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2, (SEQ ID NO: 305)

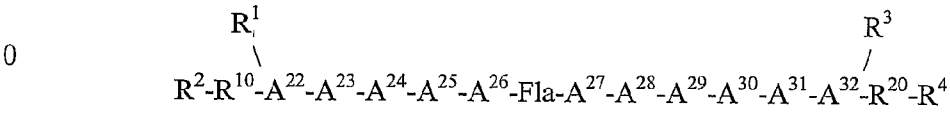
N-α-Ac-[Trp^{30}, $\psi^{35/36}$]PYY(25-36)-NH_2, (SEQ ID NO: 306)

N-α-Ac-[Trp^{30}]PYY(25-36)-NH_2 (SEQ ID NO: 307) and

N-α-Ac-[Nle^{28}, Trp^{30}, Nva^{31}, $\psi^{35/36}$]PYY(22-36)-NH_2, (SEQ ID NO: 308) or a pharmaceutically acceptable salt thereof.

Another particular compound has the formula N-α-Ac-[$Nle^{24,28}$, Trp^{30}, Nva.sup.31, $\psi^{35/36}$]PYY(22-36)-NH_2 (SEQ. ID. NO: 309) or a pharmaceutically acceptable salt thereof.

Another PYY agonist has the formula (A),



having one or two pseudopeptide bonds where each pseudopeptide bond is independently selected from the group consisting of --CH_2 --NH--, --CH_2 --S--, --CH_2 --CH_2 --, --CH_2 --O-- and -CH_2 --CO--; wherein:

R^{10} is a chain of 0-5 amino acids, inclusive, where the N-terminal amino acid is bonded to R^1 and R^2 by the side chain of the N-terminal amino acid or by the nitrogen of the amino group of the N-terminal amino acid;

R^{20} is a chain of 0-4 amino acids, inclusive, where the C-terminal amino acid is bonded to R^3 and R^4 by the side chain of the C-terminal amino acid or by the carbon of the carboxyl group of the C-terminal amino acid;

R^1, R^2, R^3 and R^4 are each independently selected from the group consisting of H, (C_1 -C_{12})alkyl, (C_6 -C_{18})aryl, (C_1 -C_{12})acyl, phenyl(C_1 -C_{12})alkyl and ((C_1 -C_{12})alkyl)$_{1-5}$ -phenyl;

A^{22} is an aromatic amino acid, Ala, Aib, Anb, N-Me-Ala or is deleted;

A^{23} is Ser, Thr, Ala, N-Me-Ser, N-Me-Thr, N-Me-Ala or is deleted;

A^{24} is Leu, Ile, Nle, Val, Trp, Gly, Aib, Anb, N-Me-Leu or is deleted;

A^{25} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-p.epsilon.-NH-Z, Orn or is deleted;

A^{26} is His, Thr, 3-Me-His, 1-Me-His, β-pyrazolylalanine, N-Me-His, Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-Z, Orn or is deleted;

A^{28} is Leu, Ile, Nle, Val, Trp, Aib, Anb or N-Me-Leu;

A^{29} is Asn, Ala, Gln, Gly, Trp or N-Me-Asn;

A^{30} is Leu, Ile, Nle, Fla, Val, Trp, Aib, Anb or N-Me-Leu;

A^{31} is Val, Nva, Ile, Trp, Aib, Anb or N-Me-Val; and

A^{32} is Thr, Ser, N-Me-Ser or N-Me-Thr;

where Z for each occurrence is independently selected from the group consisting of H, (C_1 -C_{10})alkyl and (C_6 -C_{18})aryl; or a pharmaceutically acceptable salt thereof. See U.S. Patent No. 6,046,167.

A particular group of compounds of the immediately foregoing group of compounds is where R^{10} is A^{17} -A^{18} -A^{19} -A^{20} -A^{21};

where A^{17} is Cys, Leu, Ile, Val, Nle, Nva, Aib, Anb or N-Me-Leu;

A^{18} is Cys, Ser, Thr, N-Me-Ser or N-Me-Thr;

A^{19} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R.sup.5, Cys or Orn;

A^{20} is an aromatic amino acid or Cys;

A^{21} is an aromatic amino acid or Cys;

R^{20} is A^{33} -A^{34} -A^{35} -A^{36},

A^{33} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R^5, Cys or Orn;

A^{34} is Cys, Gin, Asn, Ala, Gly, N-Me-Gln, Aib or Anb;

A^{35} is Arg, Lys, homo-Arg, diethyl-homo-Arg, Lys-ε-NH-R^5, Cys or Orn; and

A^{36} is an aromatic amino acid or Cys;

where R^5 for each occurrence is independently selected from the group consisting of H_1 (C_1 -C_{10})alkyl and (C_6 -C_{18}) aryl.

A particular group of compounds of the foregoing group of compounds are the compounds of the formula N-α-Ac-[Fla^{27})]PYY(25-36)-NH_2 and N-α-Ac-[Fla^{27}]PYY(22-36)-NH_2 or a pharmaceutically acceptable salt thereof.

Another group of PYY agonist has the formula:

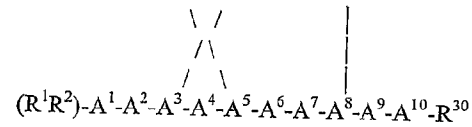
(I)

$(R^1 R^2)$-A^1 -A^2 -A^3 -A^4 -A^5 -A^6 -A^7 -A^8 -A^9 -A^{10} -R^{30},

(II)

(R^1R^2)-A^1-A^2-A^3-A^4-A^5-A^6-A^7-A^8-A^9-A^{10}-R^{30}

\ /

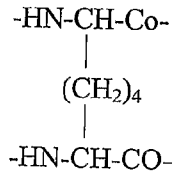


(III)

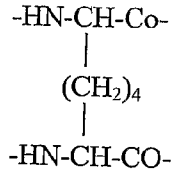
$$(R^1R^2)\text{-}[A^5\text{-}A^6\text{-}A^7\text{-}A^8\text{-}A^9\text{-}A^{10}]_m R^{30},$$

or a pharmaceutically acceptable salt thereof wherein

------represents an optional bond between the amino acids shown connected where each bond is independently selected from the group consisting of --S--S-- only when the amino acids connected are Cys-Cys, -CO-NH-, $-CH_2$ -NH- and



provided that when the optional bond is



it replaces the two amino acids that the optional bond is attached to; q is 1-4; m is 1 to 4;

R^{30} is OH or -O-R^1, provided that when A^1 to A^7 are deleted then R^{30} is also NH-R^1, where R^{30} is attached to the carbon atom of the carboxyl of the C-terminal amino acid;

R^1 and R^2 for each occurrence are each independently selected from the group consisting of H, (C_1 -C_{12})alkyl, (C_6 -C_{18})aryl, (C_1 -C_{12})acyl, phenyl(C_1 -C_{12})alkyl and ((C_1 -C_{12})alkyl)$_{1\text{-}5}$ -phenyl where R^1 and R^2 are attached to the nitrogen of the amine of the N-terminal amino acid;

A^1 is deleted or D- or L- of the following amino acids: Trp, Tyr, Fla, Bth, Nal, Tic, Tic-OH, Dip, Bip or optionally substituted Phe where the Phe is optionally

substituted with one to five substituents selected from the group consisting of (C_1 -C_4)alkyl, halo, (C_1 -C_4)alkoxy, amino and nitro;

A^2 is deleted or D- or L- of the following amino acids: Ile, Val, Leu, Nle, Anb, Aib, Pro, Gln or Asn;

A^3 is deleted or D- or L- of the following amino acids: Asn, Gln, Glu, Asp, Orn, Lys, Dpr or Cys;

A^4 is deleted or D- or L- of the following amino acids: Ile, Val, Leu, Nle, Anb, Aib or Pro;

A^5 is deleted or D- or L- of the following amino acids: Ile, Val, Leu, Nle, Anb, Aib, Pro, Glu, Asp, Orn, Lys, Dpr or Cys;

A^6 is deleted or D- or L- of the following amino acids: Thr, Ser, Trp, Tyr, Fla, Bth, Nal, Tic, Tic-OH, Dip, Bip or optionally substituted Phe where the Phe is optionally substituted with one to five substituents selected from the group consisting of (C_1 -C_4)alkyl, halo, (C_1 -C_4)alkoxy, amino and nitro;

A^7 is deleted or D- or L- of the following amino acids: Arg, Lys, homo-Arg, dialkyl-homo-Arg, Lys-ε-NH-R^7 or Orn;

A^8 is deleted or D- or L- of the following amino acids: Nva, Val, Ile, Leu, Nle, Anb, Aib, Pro, Gln, Asn, Glu, Asp, Orn, Lys, Dpr or Cys;

A^9 is deleted or D- or L- of the following amino acids: Arg, Lys, homo-Arg, dialkyl-homo-Arg, Lys-ε-NH-R^7 or Orn; and

A^{10} is deleted or D- or L- of the following amino acids: Tyr, Trp, Fla, Bth, Nal, Tic, Tic-OH, Dip, Bip, tyramine or optionally substituted Phe where the Phe is optionally substituted with one to five substituents selected from the group consisting of (C_1 -C_4)alkyl, halo, (C_1 -C_4)alkoxy, amino and nitro, or the corresponding decarboxylated optionally substituted Phe;

where R^7 for each occurrence is independently selected from the group consisting of H.sub.1 (C_1 -C_{10})alkyl and (C_6 -C_{18}) aryl, provided that not all of A_1 to A_{10} are deleted at the same time. See U.S. Patent No. 6,046,167.

A particular group of compounds of the immediately foregoing group of compounds is

(SEQ ID NO: 310)

H--Ile--Asn--Pro--Ile--Tyr--Arg--Leu--Arg--Tyr--OMe

(SEQ ID NO: 311)

H--Ile--Asn--Pro--Cys--Tyr--Arg--Leu--Arg--Tyr—Ome

H--Ile--Asn--Pro--Cys--Tyr--Arg--Leu--Arg--Tyr--Ome,

(SEQ ID NO: 312)

H--Cys--Tyr--Arg--Leu--Arg--Tyr—Ome

H--Cys--Tyr--Arg--Leu--Arg--Tyr--OMe,

(SEQ ID NO: 313)

H--Ile--Asn--Pro--NH--CH--CO--Tyr--Arg--Leu--Arg--Tyr--OMe

$(CH_2)_4$

H--Ile--Asn--Pro--NH--CH--CO--Tyr--Arg--Leu--Arg--Tyr--OMe,

(SEQ ID NO: 314)

H-[Tyr-Arg-Leu-Arg-Tyr]$_2$ –Ome

or a pharmaceutically acceptable salt thereof.

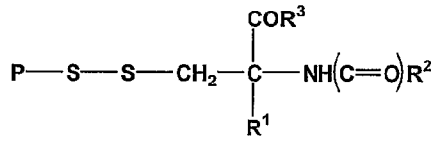
PYY and PYY agonists may be modified by well known processes such as amidation, glycosylation, acylation (e.g. acetylation), sulfation, phosphylation, cyclization, lipidization and pegylation. Methods for lipidization with fatty acid derivatives of sulfhydryl-containing compounds are disclosed in U.S. Patent No. 5,936,092; U.S. Patent No. 6,093,692; and U.S. Patent No. 6,225,445. Fatty acid derivatives of sulfhydryl-containing PYY and PYY agonists comprising fatty acid-conjugated products with a disulfide linkage are employed for delivery of the PYY and PYY agonists to neuronal cells and tissues. This modification markedly increases the absorption of the compounds relative to the rate of absorption of the unconjugated compounds, as well as prolonging blood and tissue retention of the compounds. Moreover, the disulfide linkage in the conjugate is quite labile in the

cells and thus facilitates intracellular release of the intact compounds from the fatty acid moieties.

Fatty acids, as constituents of phospholipids, make up the bulk of cell membranes. Due to their lipidic nature, fatty acids can easily partition into and interact with the cell membrane in a non-toxic way. Therefore, fatty acids represent potentially a useful carrier ligand for the delivery of proteins and peptides. Strategies that may use fatty acids in the delivery of proteins and peptides include the covalent modification of proteins and peptides and the use of fatty acid emulsions.

To prepare such conjugates, a sulfhydryl-containing PYY and PYY agonist is attached to a fatty acid derivative via a reversible, biodegradable disulfide bond. Such a conjugate is expected to bind to the apical side of a cell membrane, reach the basolateral membrane of the GI-epithelium as a result of membrane transport and turnover, and become released into interstitial fluid as the result of disulfide bond reduction.

Such lipidized PYY and PYY agonist compounds have the general formula



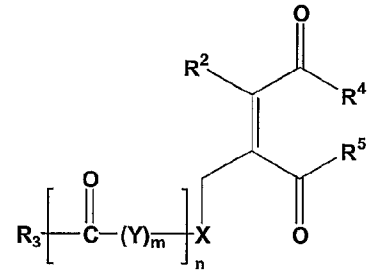
in which P is a residue derived from a PYY or PYY agonist; R^1 is hydrogen, lower alkyl or aryl; R^2 is a lipid-containing moiety and R^3 is --OH, a lipid-containing moiety or an amino acid chain comprising one or 2 amino acids and terminating in $-CO_2H$ or $-COR^2$. See U.S. Patent No. 5,936,092. These conjugates are particularly useful for increasing the absorption and prolonging blood and tissue retention of PYY and PYY agonists.

Typical alkyl groups include C_{1-6} alkyl groups including methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, 2-pentyl, 3-pentyl, neopentyl, hexyl, 2-hexyl, 3-hexyl, 2-methyl-1-pentyl, 3-methyl-1-pentyl, 4-methyl-1-pentyl, and the like.

Preferred aryl groups are C_{6-14} aryl groups and typically include phenyl, naphthyl, fluorenyl, phenanthryl, and anthracyl groups.

The term "lipid-containing moiety" refers to either a lipid group per se or a hydrocarbon-based group (in particular, one or more amino acids) comprising a lipid group. By the term "lipid group" is meant a hydrophobic substituent consisting of 4 to 26 carbon atoms, preferably 5 to 19 carbon atoms. Suitable lipid groups include, but are not limited to, the following: palmityl ($C_{15}H_{31}$,), oleyl ($C_{15}H_{29}$), stearyl ($C_{17}H_{35}$), cholate; and deoxycholate.

PCT Application No. WO 00/34236 describes drug-carrier conjugates and synthetic strategies for their production, as well as synthetic methods, intermediates, and final products useful for the uptake and release of biologically-active amino group containing compounds. Such lipidized PYY and PYY agonist compounds have general Formula I



in which R^2 is selected from the group consisting of hydrogen, halo, alkyl, or aryl, wherein the alkyl or aryl groups are optionally substituted with one or more alkoxy, alkoxyalkyl, alkanoyl, nitro, cycloalkyl, alkenyl, alkynyl, alkanoyloxy, alkyl or halogen atoms;

R^3 is a lipophilic group; one of R^4 and R^5 is a PYY or a PYY agonist and the other of R^4 and R^5 is OR^6 where R^6 is hydrogen, an alkali metal or a negative charge;

X is oxygen or sulfur;

Y is a bridging natural or unnatural amino acid; n is zero or 1; and m is an integer from zero to 10.

Typical alkyl groups include C_{1-6} alkyl groups including methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, 2-pentyl, 3-pentyl, neopentyl, hexyl, 2-hexyl, 3-hexyl, 2-methyl-1-pentyl, 3-methyl-1-pentyl, 4-methyl-1-pentyl, and the like.

Typical alkoxy groups include oxygen substituted by any of the alkyl groups

mentioned above.

Typical alkoxyalkyl groups include any of the above alkyl groups substituted by an alkoxy group, such as methoxymethyl, ethoxymethyl, propoxymethyl, butoxymethyl, pentoxymethyl, hexoxymethyl, methoxyethyl, methoxypropyl, methoxybutyl, methoxypentyl, methoxyhexyl, and the like.

Preferred aryl groups are C_{6-14} aryl groups and typically include phenyl, naphthyl, fluorenyl, phenanthryl, and anthracyl groups.

Typical alkoxy substituted aryl groups include the above aryl groups substituted by one or more of the above alkoxy groups, e.g., 3-methoxyphenyl, 2-ethoxyphenyl, and the like.

Typical alkyl substituted aryl groups include any of the above aryl groups substituted by any of the C_{1-6} alkyl groups, including the group $Ph(CH_2)n$, where n is 1-6, for example, tolyl, o-, m-, and p-xylyl, ethylphenyl, 1-propylphenyl, 2-propylphenyl, 1-butylphenyl, 2-butylphenyl, t-butylphenyl, 1-pentylphenyl, 2-pentylphenyl, 3-pentylphenyl.

Typical alkenyl groups include C_{2-6} alkenyl groups, e.g. ethenyl, 2-propenyl, isopropenyl, 2-butenyl, 3-butenyl, 4-pentenyl, 3-pentenyl, 2-pentenyl, 5-hexenyl, 4-hexenyl, 3-hexenyl, and 2-hexenyl groups.

Typical alkynyl groups include C_{2-6} alkynyl groups e.g. enthynyl, 2-propenyl, 2-butynyl, 3-butynyl, 4-pentynyl, 3-pentynyl, 2-pentynyl, 5-hexynyl, 4hexynyl, 3-hexynyl, and 2-hexynyl groups.

Typical alkenyl or alkynyl substituted aryl groups include any of the above C_{6-14} aryl groups substituted by any of the above C_{2-6} alkenyl or C_{2-6} alkynyl groups, e.g., ethenylphenyl, 1-propenylphenyl, 2-propenylphenyl, 1butenylphenyl, 2-butenylphenyl, 1-pentenylphenyl, 2-pentenylphenyl, 3-pentenylphenyl, 1-hexenylphenyl, 2-hexenylphenyl, 3-hexenylphenyl, ethynylphenyl, 1-propynylphenyl, 2-propynylphenyl, 1-butynylphenyl, 2-butynylphenyl, 1-pentynylphenyl, 2-pentynylphenyl, 3-pentynylphenyl, 1-hexynylphenyl, 2-hexynylphenyl, 3-hexynylphenyl groups.

Typical halo groups include fluorine, chlorine, bromine, and iodine.

Typical halo substituted alkyl groups include C_{1-6} alkyl groups substituted by one or more fluorine, chlorine, bromine, or iodine atoms, e.g., fluoromethyl,

difluoromethyl, trifluoromethyl, pentafluoroethyl, 1,1-difluoroethyl, and trichloromethyl groups.

Typical alkanoyl groups include $C_{1-5}C(=O)-$ alkanoyl groups, e.g., acetyl, propionyl, butanoyl, pentanoyl, and hexanoyl groups, or by an arylalkanoyl group, e.g., a $C_{1-5}C(=O)-$ alkanoyl group substituted by any of the above aryl groups.

Typical cycloalkyl groups include C_{3-8} cycloalkyl groups including cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl and cyclooctyl groups.

The term "lipophilic group" as used herein refers to either a naturally occurring lipid per se, a hydrophobic branched or unbranched hydrocarbon comprising about 4 to about 26 carbon atoms, preferably about 5 to about 19 carbon atoms, a fatty acid or ester thereof, or a surfactant. Suitable lipophilic groups include, but are not limited to, long chain alkanoyl groups including: palmityl ($C_{15}H_{31}$), oleyl ($C_{15}H_{29}$), stearyl ($C_{17}H_{35}$), lauryl ($C_{11}H_{23}$), cholyl, and myristyl ($C_{13}H_{27}$)

The term "natural or unnatural amino acid" as used herein refers to any of the 21 naturally occurring amino acids as well as D-form amino acids, blocked L-and D-form amino acids such as those blocked by amidation or acylation, substituted amino acids (e.g., those substituted with a sterically hindered alkyl group or a cycloalkyl group such as cyclopropyl or cyclobutyl) in which the substitution introduces a conformational restraint in the amino acid. The preferred naturally occurring amino acids for use in the present disclosure as amino acids or components of a peptide or protein are alanine, arginine, asparagine, aspartic acid, citrulline, cysteine, cystine, y-glutamic acid, glutamine, glycine, histidine, isoleucine, norleucine, leucine, lysine, methionine, ornithine, phenylalanine, proline, hydroxyproline, serine, threonine, tryptophan, tyrosine, valine, γ -carboxyglutamate, or O-phosphoserine. The preferred non-naturally occurring amino acids for use in the present disclosure as amino acids or components of peptides or proteins are any of the β-amino acids, e.g., α-alanine, γ-amino butyric acid, γ-amino butyric acid, γ-(aminophenyl)butyric acid, α-amino isobutyric acid, ε-amino caproic acid, 7-amino heptanoic acid, amino benzoic acid, aminophenyl acetic acid, aminophenyl butyric acid, cysteine (ACM), methionine sulfone, phenylglycine, norvaline, ornithine, δ-ornithine, ρnitro-phenylalanine, 1,2,3,4-terahydroisoquinoline-3-carboxylic acid and thioproline.

The present disclosure is also directed to methods of preparing lipidized conjugates of PYY and PYY agonists, pharmaceutical compositions comprising lipidized conjugates of PYY and PYY agonists, and methods of increasing the delivery of amino group-containing PYY and PYY agonists into a cell.

Also provided by the disclosure are chemically modified derivatives of PYY and PYY agonists which may provide additional advantages such as increased solubility, stability and circulating time of the polypeptide, or decreased immunogenicity (see U.S. Patent No. 4,179,337). Such modified derivatives include PYY and PYY agonists modified by pegylation. The terms "pegylated" and "pegylation" refer to the process of reacting a poly(alkylene glycol), preferably an activated poly(alkylene glycol), with a facilitator such as an amino acid, e.g. lysine, to form a covalent bond. Although "pegylation" is often carried out using poly(ethylene glycol) or derivatives thereof, such as methoxy poly(ethylene glycol), the term is not intended to be so limited here, but is intended to include any other useful poly(alkylene glycol), such as, for example poly(propylene glycol).

The chemical moieties for derivatization may also be selected from water soluble polymers such as polyethylene glycol, ethylene glycol/propylene glycol copolymers, carboxymethylcellulose, dextran, polyvinyl alcohol and the like. The polypeptides may be modified at random positions within the molecule, or at predetermined positions within the molecule and may include one, two, three or more attached chemical moieties.

The polymer may be of any molecular weight, and may be branched or unbranched. For polyethylene glycol, the preferred molecular weight is between about 1 kDa and about 100 kDa (the term "about" indicating that in preparations of polyethylene glycol, some molecules will weigh more, some less, than the stated molecular weight) for ease in handling and manufacturing. Other sizes may be used, depending on the desired therapeutic profile (e.g., the duration of sustained release desired, the effects, if any on biological activity, the ease in handling, the degree or lack of antigenicity and other known effects of the polyethylene glycol to a therapeutic protein or analog). For example, the polyethylene glycol may have an average molecular weight of about 200, 500, 1000, 1500, 2000, 2500, 3000, 3500, 4000, 4500, 5000, 5500, 6000, 6500, 7000, 7500, 8000, 8500, 9000, 9500, 10,000,

10,500, 11,000, 11,500, 12,000, 12,500, 13,000, 13,500, 14,000, 14,500, 15,000, 15,500, 16,000, 16,500, 17,000, 17,500, 18,000, 18,500, 19,000, 19,500, 20,000, 25,000, 30,000, 35,000, 40,000, 50,000, 55,000, 60,000, 65,000, 70,000, 75,000, 80,000, 85,000, 90,000, 95,000, or 100,000 kDa.

As noted above, the polyethylene glycol may have a branched structure. Branched polyethylene glycols are described, for example, in U.S. Patent No. 5,643,575; Morpurgo et al., *Appl. Biochem. Biotechnol.* 56:59-72, 1996; Vorobjev et al., *Nucleosides Nucleotides* 18:2745-2750, 1999; and Caliceti et al., *Bioconjug. Chem.* 10:638-646, 1999.

The polyethylene glycol molecules (or other chemical moieties) should be attached to the polypeptides or proteins with consideration of effects on functional or antigenic domains of the polypeptides or proteins. There are a number of attachment methods available to those skilled in the art, e.g., EP 0 401 384 (coupling PEG to G-CSF), see also Malik et al., *Exp. Hematol.* 20:1028-1035, 1992 (reporting pegylation of GM-CSF using tresyl chloride). For example, polyethylene glycol may be covalently bound through amino acid residues via a reactive group, such as, a free amino or carboxyl group. Reactive groups are those to which an activated polyethylene glycol molecule may be bound. The amino acid residues having a free amino group may include lysine residues and the N-terminal amino acid residues; those having a free carboxyl group may include aspartic acid residues glutamic acid residues and the C-terminal amino acid residue. Sulfhydryl groups may also be used as a reactive group for attaching the polyethylene glycol molecules. Preferred for therapeutic purposes is attachment at an amino group, such as attachment at the N-terminus or lysine group.

As suggested above, polyethylene glycol may be attached to proteins and polypeptides via linkage to any of a number of amino acid residues. For example, polyethylene glycol can be linked to proteins and polypeptides via covalent bonds to lysine, histidine, aspartic acid, glutamic acid, or cysteine residues. One or more reaction chemistries may be employed to attach polyethylene glycol to specific amino acid residues (e.g., lysine, histidine, aspartic acid, glutamic acid, or cysteine) of the polypeptide or protein or to more than one type of amino acid residue (e.g.,

lysine, histidine, aspartic acid, glutamic acid, cysteine and combinations thereof) of the protein or polypeptide.

One may specifically desire proteins and polypeptides chemically modified at the N-terminus. Using polyethylene glycol as an illustration, one may select from a variety of polyethylene glycol molecules (by molecular weight, branching, etc.), the proportion of polyethylene glycol molecules to protein (or peptide) molecules in the reaction mix, the type of pegylation reaction to be performed, and the method of obtaining the selected N-terminally pegylated protein. The method of obtaining the N-terminally pegylated preparation (i.e., separating this moiety from other monopegylated moieties if necessary) may be by purification of the N-terminally pegylated material from a population of pegylated protein molecules. Selective proteins chemically modified at the N-terminus modification may be accomplished by reductive alkylation which exploits differential reactivity of different types of primary amino groups (lysine versus the N-terminal) available for derivatization in a particular protein. Under the appropriate reaction conditions, substantially selective derivatization of the protein at the N-terminus with a carbonyl group containing polymer is achieved.

As indicated above, pegylation of the proteins and polypeptides may be accomplished by any number of means. For example, polyethylene glycol may be attached to the protein or polypeptide either directly or by an intervening linker. Linkerless systems for attaching polyethylene glycol to proteins and polypeptides are described in Delgado et al., *Crit. Rev. Thera. Drug Carrier Sys.* 9:249-304, 1992; Francis et al., *Intern. J. of Hematol.* 68:1-18, 1998; U.S. Patent No. 4,002,531; U.S. Patent No. 5,349,052; WO 95/06058; and WO 98/32466.

One system for attaching polyethylene glycol directly to amino acid residues of proteins and polypeptides without an intervening linker employs tresylated MPEG, which is produced by the modification of monmethoxy polyethylene glycol (MPEG) using tresylchloride ($ClSO_2CH_2CF_3$). Upon reaction of the protein or polypeptide with tresylated MPEG, polyethylene glycol is directly attached to amine groups of the protein or polypeptide. Thus, the disclosure includes protein-polyethylene glycol conjugates produced by reacting proteins and polypeptides with a polyethylene glycol molecule having a 2,2,2-trifluoreothane sulphonyl group.

Polyethylene glycol can also be attached to proteins and polypeptides using a number of different intervening linkers. For example, U.S. Patent No. 5,612,460 discloses urethane linkers for connecting polyethylene glycol to proteins. Protein-polyethylene glycol conjugates wherein the polyethylene glycol is attached to the protein or polypeptide by a linker can also be produced by reaction of proteins or polypeptides with compounds such as MPEG-succinimidylsuccinate, MPEG activated with 1,1'-carbonyldiimidazole, MPEG-2,4,5-trichloropenylcarbonate, MPEG- ρ -nitrophenolcarbonate, and various MPEG-succinate derivatives. A number of additional polyethylene glycol derivatives and reaction chemistries for attaching polyethylene glycol to proteins and polypeptides are described in WO 98/32466.

The number of polyethylene glycol moieties attached to each protein or polypeptide (i.e., the degree of substitution) may also vary. For example, the pegylated proteins and polypeptides may be linked, on average, to 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 12, 15, 17, 20, or more polyethylene glycol molecules. Similarly, the average degree of substitution within ranges such as 1-3, 2-4, 3-5, 4-6, 5-7, 6-8, 7-9, 8-10, 9-11, 10-12, 11-13, 12-14, 13-15, 14-16, 15-17, 16-18, 17-19, or 18-20 polyethylene glycol moieties per protein or polypeptide molecule. Methods for determining the degree of substitution are discussed, for example, in Delgado et al., *Crit. Rev. Thera. Drug Carrier Sys.* 9:249-304, 1992.

The proteins and polypeptides containing substantially non-antigenic polymers, preferably poly(alkylene glycols) may be prepared, for example, as described in U.S. Patent No. 5,428,128; U.S. Patent No. 6,127,355; and U.S. Patent No. 5,880,131.

To effect covalent attachment of poly(ethylene glycol) (PEG) to a protein or polypeptide, the hydroxyl end groups of the PEG must first be converted into reactive functional groups. This process is frequently referred to as "activation" and the product is called "activated PEG." Methoxy poly(ethylene glycol) (mPEG), distally capped with a reactive functional group is often used. One such activated PEG is succinimidyl succinate derivative of PEG (SS-PEG). See also Abuchowski et al., *Cancer Biochem. Biophys.* 7:175-186, 1984; and U.S. Patent No. 5,122,614 which discloses poly(ethylene glycol)-N-succinimide carbonate and its preparation.

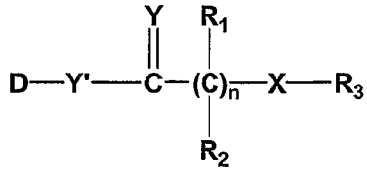
Alternative substantially non-antigenic polymers that may be employed in the practice of the present disclosure include materials such as dextran, polyvinyl pyrrolidones, polysaccharides, starches, polyvinyl alcohols, polyacrylamides, or other similar non-immunogenic polymers. Those of ordinary skill in the art will realize that the foregoing is merely illustrative and not intended to restrict the type of polymeric substances suitable for use herein.

In one aspect of the disclosure, the polymer is introduced into the peptide or protein molecule after being functionalized or activated for reaction and attachment to one or more amino acids. By activation, it is understood by those of ordinary skill in the art that the polymer is functionalized to include a desired reactive group. See, for example, U.S. Patent No. 4,179,337 and U.S. Patent No. 5,122,614. In this embodiment, the hydroxyl end groups of poly(alkylene glycols) are converted and activated into reactive functional groups.

In another aspect of the disclosure, the polymer is conjugated to a facilitator moiety prior to being introduced into the polypeptide or protein molecule. The facilitator moiety is preferably an amino acid such as lysine, however, non-amino acid moieties are also contemplated. Within the aspect, there are included multifunctionalized organic moieties such as alkyls or substituted alkyls. Such moieties can be prepared to have a nucleophilic functional group such as an amine and an electrophilic group such as an acid as well as a suitably functionalized region for conjugating with the desired polymer or polymers.

The facilitator moieties allow easier inclusion of a polymer into the peptide or protein molecule during synthesis. For example, poly(alkylene glycols) coupled to facilitator amino acids or amino acid residues in polypeptides or proteins by means of suitable coupling agents are illustrative. A useful review of a number of coupling agents known in the art appears in Dreborg et al., *Critical Reviews in Therapeutic Drug Carrier Systems* 6(4):315-165, 1990, see especially, pp. 317-320.

Pegylated PYY peptides and agonists can also be of the general formula



wherein:

D is a residue of a PYY peptide or agonist;

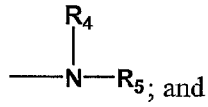
X is an electron withdrawing group;

Y and Y' are independently O or S;

(n) is zero (0) or a positive integer, preferably from 1 to about 12;

R_1 and R_2 are independently selected from the group consisting of H, C_{1-6} alkyls, aryls, substituted aryls, aralkyls, heteroalkyls, substituted heteroalkyls, and substituted C_{1-6} alkyls;

R_3 is a substantially non-antigenic polymer, C_{1-12} straight or branched alkyl or substituted alkyl, C_{5-8} cycloalkyl or substituted cycloalkyl, carboxyalkyl, carboalkoxy alkyl, dialkylaminoalkyl, phenylalkyl, phenylaryl or



; and

R_4 and R_5 are independently selected from the group consisting of H, C_{1-6} alkyls, aryls, substituted aryls, aralkyls, heteroalkyls, substituted heteroalkyls and substituted C_{1-6} alkyls or jointly form a cyclic C_5-C_7 ring. See U.S. Patent No. 6,127,355.

Typical alkyl groups include C_{1-6} alkyl groups including methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, 2-pentyl, 3-pentyl, neopentyl, hexyl, 2-hexyl, 3-hexyl, 2-methyl-1-pentyl, 3-methyl-1-pentyl, 4-methyl-1-pentyl, and the like.

Preferred aryl groups are C_{6-14} aryl groups and typically include phenyl, naphthyl, fluorenyl, phenanthryl, and anthracyl groups.

Typical alkyl substituted aryl groups include any of the above aryl groups substituted by any of the C_{1-6} alkyl groups, including the group $Ph(CH_2)_n$, where n is 1-6, for example, tolyl, o-, m-, and p-xylyl, ethylphenyl, 1-propylphenyl, 2-propylphenyl, 1-butylphenyl, 2-butylphenyl, t-butylphenyl, 1-pentylphenyl, 2-pentylphenyl, 3-pentylphenyl.

Typical cycloalkyl groups include C_{3-8} cycloalkyl groups including cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl and cyclooctyl groups.

Typical electron withdrawing groups include O, NR_1, S, SO and SO_2, wherein R_1 is defined above.

PYY Antagonists

Also contemplated, are the use of Y receptor antagonist. A Y receptor antagonist is a substance (typically a ligand) which binds to a Y receptor and blocks the physiological effect of a Y receptor agonist (such as, PYY, NPY, or PP (see Tables 1-3, *infra*). These antagonists could be either peptide antagonist or non-peptide antagonist of PYY, NPY, or PP.

Peptide antagonist include modifications, mutants, fragments, and/or variants thereof, of the PYY, NPY, or PP peptide's natural amino acid sequence (*e.g.*, by deletions, amino acid substitutions, deletions, insertions, and modifications of the N-terminal amino and/or C-terminal carboxyl group) resulting in a peptide which acts as an antagonist to a Y receptor. In addition, PYY, NPY, or PP amino acid sequences may be fusion or chimera proteins which act as antagonists at the Y receptor. These peptides may also be modified by processes such as, lipidation, pegylation, amidation, glycosylation, acylation, sulfation, phosphorylation, acetylation and cyclization.

Many non-peptide antagonist of the Y receptors are known in the art and are contemplated for use with this invention. (See Table 5, *infra*). Any known PYY, NPY, or PP non-peptide antagonist may be useful in this invention.

TABLE 5 – PYY AND NPY ANTAGONIST

Exemplary antagonists of the Y receptor include, but are not limited to the following:

BIBO3304

Ref: Berglund, MM. *Biochem Pharmacol* 60(12):1815-22, Dec 15, 2000.

SR120819A

1-[2-[2-(2-naphtylsulfamoyl)-3-phenylpropionamido]-3-[4-[N- [4-(dimethylaminomethyl)-cis-cyclohexylmethyl]amidino]phenyl]propiony l] pyrrolidine, (S,R) stereoisomer

Ref: Berglund, MM. *Biochem Pharmacol* 60(12):1815-22, Dec 15, 2000.

BIIE0246

(S)-N2-[[1-[2-[4-[(R,S)-5,11-dihydro-6(6h)-oxodibenz[b,e]azepin-11-yl]-1-piperazinyl]-2-oxoethyl]cyclopentyl]acetyl]-N-[2-[1,2-dihydro-3,5 (4H)-dioxo-1,2-diphenyl-3H-1,2,4-triazol-4-yl]ethyl]-argininamid

Ref: Malmstrom, *Life Sci* 69(17):1999-2005, Sep 14, 2001.

BIBP 3226

[(R)-N2-(diphenylacetyl)-N-[(4-hydroxyphenyl)methyl]-D-arginine-amide], and a recently described peptidic structure [Ile-Glu-Pro-Orn-Tyr-Arg-Leu-Arg-Tyr-NH2, cyclic (2,4'), (2',4)-diamide].

Ref: Doods, H.N. *J Pharmacol Exp Ther* 275(1):136-42, Oct, 1995.

BIBP 3435

Ref: Lundberg J.M., Modin A. *Br J Pharmacol* 116(7):2971-82, Dec, 1995.

H 394/84

1,4-Dihydro-4-[3-[[[[3-[spiro(indene-4,1'-piperidin-1-yl)]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethylester

Ref: Malmstrom, R.E. *Eur J Pharmacol* 418(1-2):95-104, Apr 20, 2001.

H 409/22

(2R)-5-([amino(imino)methyl]amino)-2-[(2,2-diphenylacetyl)amino]-N-[(IR)-1-(4-hydroxyphenyl)ethyl]-pentanamide

Ref: Malmstrom, R.E. *Life Sci* 69(17):1999-2005, Sep 14, 2001.

1229U91

Ref: Schober, DA. *Peptides* 19(3):537-42, 1998.

L-152,804

Ref: Kanatani, A. *Biochem Biophys Res Commun* 272(1):169-73, May 27, 2000.

Aminoalkyl substituted pyrazolo[1,5,-a]-1,5- pyrimidines and pyrazolo[1,5-a]-1,3,5-triazines

Ref: U.S. Patent No. 6,372,743

Alkyl and cycloalkyl derivatives of 1,4-dihydropyridine

(e.g., 1,4-dihydro-2,6-dimethyl-4-[4-[[[[3-[4-(3-methoxyphenyl)-1-piperidinyl]prop yl]amino]carbonyl]amino]butyl]-3,5-pyridine dicarboxylic acid, dimethyl ester)

Ref: U.S. Patent No. 6,444,675

4-(3-substituted-phenyl)-1,4-dihydropyridine derivatives

Ref: U.S. Pat. No. 5,635,503

Squarate derivatives of 4-phenyl-1,4-dihydropyridines

e.g., 1,4-dihydro-4-[3-[[2-[[3-[4-(3-methoxyphenyl)-1-piperidinyl]propyl]amino]-3 ,4-dioxo-1-cyclobuten-1-yl]amino]phenyl]-2,3-dimethyl-3,5-pyridinedicarboxy lic acid, dimethyl ester

Ref: U.S. Patent No. 6,432,960

Substituted amide Y receptor antagonist, such as:

N-(4-Diethylamino-phenyl)-2-phenyl-2-pyridin-4-yl-acetamide;

2-(4-Fluoro-phenyl)-2-pyridin-4-yl-N-(3,4,5,6-tetrahydro-2H-[1,2']bipyridin yl-5'-yl)-acetamide;

2-Phenyl-2-pyridin-4-yl-N-(3,4,5,6-tetrahydro-2H-[1,2']bipyridinyl-5'-yl)-acetamide;

N-(4-Diethylamino-phenyl)-2-phenyl-2-pyridin-2-yl-acetamide;

N-(6-Diethylamino-pyridin-3-yl)-2,2-diphenylacetamide;

N-(4-Diethyl-sulfamoyl-phenyl)-2-phenyl-2-pyridin-4-yl-acetamide;

2,2-Diphenyl-N-(6-pyrrolidin-1-yl-pyridin-3-yl)-acetamide;

2,2-Diphenyl-N-(3,4,5,6-tetrahydro-2H-[1,2']bipyridinyl-5'-yl)-acetamide;

N-[6-(2,5-Dimethyl-pyrrolidin-1-yl)-pyridin-3-yl]-2,2-diphenyl-acetamide;

N-(4-Diethylsulfamoyl-phenyl)-2,2-diphenyl-acetamide; and

N-(4-Dimethylsulfamoyl-phenyl)-2,2-diphenyl-acetamide.

Ref: U.S. Patent No. 6,407,120

Carbazole Y receptor antagonist, such as:

2-Dimethylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

3-Diethylamino-N-(9-ethyl-9H-carbazol-3-yl)-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-2-fluoro-benzamide;

4-Dimethylamino-N-(9-ethyl-9H-carbazol-3-yl)-butyramide;

N-(9-Ethyl-9H-carbazol-3-yl)-2-hydroxy-2,2-diphenyl-acetamide;

N-(9-Ethyl-9H-carbazol-3-yl)-2-hydroxy-2-methyl-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-2-hydroxy-2-methyl-butyramide;

N-(9-Ethyl-9H-carbazol-3-yl)-2-hydroxy-2-phenyl-propionamide;

(R)-N-(9-Ethyl-9H-carbazol-3-yl)-2-hydroxy-2-phenyl-propionamide;

2-Bromo-N-(9-ethyl-9H-carbazol-3-yl)-acetamide; and

3-Dimethylamino-N-(9-ethyl-9H-carbazol-3-yl)-propionamide.

2-[Bis-(2-hydroxy-ethyl)-amino]-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

2-Benzylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

3-Diphenylamino-N-(9-ethyl-9H-carbazol-3-yl)-propionamide; and

N-(9-Ethyl-9H-carbazol-3-yl)-3-(4-piperidin-1-ylmethyl-phenoxy)-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-3-[methyl-(1,2,3,4-tetrahydro-naphthalen-2-yl) -amino]-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-3-(quinolin-7-yloxy)-propionamide; and

2-[Bis-(2-hydroxy-ethyl)-amino]-N-(9-ethyl-9H-carbazol-3-yl)-acetamide.

3-Bromo-N-(9-ethyl-9H-carbazol-3-yl)-propionamide; N-(9-Isopropyl-9H-carbazol-3-yl)-trifluoroacetamide;

4-Dimethylamino-N-(9-ethyl-9H-carbazol-3-yl)-N-methyl-butyramide;

N-(9-Methyl-9H-carbazol-3-yl)-trifluoroacetamide;

1-Hydroxy-cyclopropanecarboxylic acid (9-ethyl-9H-carbazol-3-yl)-amide; and

2-(4-Chloro)-benzylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide.

2-(4-fluoro)-benzylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

(R)-N-(9-Ethyl-9H-carbazol-3-yl)-2-(1-phenyl-ethylamino)-acetamide;

(R)-N-(9-Ethyl-9H-carbazol-3-yl)-2-(1-(4-chloro)-phenyl-ethylamino)-acetamide;

2-(3-Diethylamino-2-hydroxy-propylamino)-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

2-(Benzyl-isopropyl-amino)-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

N-3-Bromo-(9-ethyl-9H-carbazol-6-yl)-trifluoroacetamide;

N-(9-Ethyl-6-formyl-9H-carbazol-3-yl)-trifluoroacetamide;

N-(9-Ethyl-6-hydroxymethyl-9H-carbazol-3-yl)-trifluoroacetamide;

N-(9-Ethyl-9H-carbazol-3-yl)-methanesulfonamide;

N-(9-Ethyl-9H-carbazol-3-yl)-chloromethanesulfonamide;

2-Bromo-N-(9-ethyl-9H-carbazol-3-yl)-acetamide; and

3-Dimethylamino-N-(9-ethyl-9H-carbazol-3-yl)-propionamide.

2-[Bis-(2-hydroxy-ethyl)-amino]-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

2-Benzylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

3-Diphenylamino-N-(9-ethyl-9H-carbazol-3-yl)-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-3-(4-piperidin-1-ylmethyl-phenoxy)-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-3-[methyl-(1,2,3,4-tetrahydro-naphthalen-2-yl) -amino]-propionamide;

N-(9-Ethyl-9H-carbazol-3-yl)-3-(quinolin-7-yloxy)-propionamide;

2-[Bis-(2-hydroxy-ethyl)-amino]-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

3-Bromo-N-(9-ethyl-9H-carbazol-3-yl)-propionamide; and

N-(9-Isopropyl-9H-carbazol-3-yl)-acetamide.

4-Dimethylamino-N-(9-ethyl-9H-carbazol-3-yl)-N-methyl-butyramide;

N-(9-Methyl-9H-carbazol-3-yl)-trifluoroacetamide;

1-Hydroxy-cyclopropanecarboxylic acid (9-ethyl-9H-carbazol-3-yl)-amide;

2-(4-Chloro)-benzylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide; and

2-(4-fluoro)-benzylamino-N-(9-ethyl-9H-carbazol-3-yl)-acetamide.

(R)-N-(9-Ethyl-9H-carbazol-3-yl)-2-(1-phenyl-ethylamino)-acetamide;

(R)-N-(9-Ethyl-9H-carbazol-3-yl)-2-(1-(4-chloro)-phenyl-ethylamino)-acetami de;

(R)-, (S)- or a mixture of (R)- and (S)-2-(3-Diethylamino-2-hydroxy-propylamino)-N-(9-ethyl-9H-carbazol-3-yl)- acetamide;

(S)-N-(6-tert-Butyl-9-ethyl-9H-carbazol-3-yl)-2-(3-diethylamino-2-hydroxy-p ropylamino)-acetamide, 2-(Benzyl-isopropyl-amino)-N-(9-ethyl-9H-carbazol-3-yl)-acetamide;

N-3-Bromo-(9-ethyl-9H-carbazol-6-yl)-trifluoroacetamide;

N-(9-Ethyl-6-formyl-9H-carbazol-3-yl)-trifluoroacetamide; and

N-(9-Ethyl-6-hydroxymethyl-9H-carbazol-3-yl)-trifluoroacetamide.

N-(9-Ethyl-9H-carbazol-3-yl)-methanesulfonamide; and

N-(9-Ethyl-9H-carbazol-3-yl)-chloromethanesulfonamide.

Ref: U.S. Patent No. 6,399,631

Various dihydropyridine derivatives:

Ref: U.S. Patent No. 4,829,076

Cyanoguanidine derivatives of the 4-(3-substituted-phenyl)-1,4-dihydropyridines

Ref: U.S. Patent No. 6,001,836

Amide derivatives that are NPY Y5 receptor antagonists

Ref: U.S. Patent No. 6,410,792

Thiourea linked piperazine and piperidine derivatives of 4-phenyl-1,4-dihydropyridines, such as:

1,4-dihydro-4-[3-[[[[3-[4-(3-methoxyphenyl)piperidinyl]propyl]amino]carbono thioyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester,

1,4-dihydro-4-[3-[[[[3-(4-phenylpiperidinyl)propyl]amino]carbonothioyl]amin o]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester, and

1,4-dihydro-4-[4-[[[[3-(4-cyclohexyl-1-piperazinyl)propyl]amino]carbonothio yl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester.

1,4-dihydro-4-[4-fluoro-3-[[[[3-(4-phenylpiperidinyl)propyl]amino]carbonoth ioyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester,

1,4-dihydro-4-[3-[[[[3-(4-methyl-1-piperidinyl)propyl]amino]carbonothioyl]a mino]-4-fluorophenyl)-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester,

1,4-dihydro-4-[3-[[[[3-(4-ethyl-1-piperidinyl]propyl]amino]carbonothioyl]am ino]-4-fluorophenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester,

1,4-dihydro-4-[3-[[[[3-(4-propyl-1-piperidinyl)propyl]amino]carbonothioyl]a

mino]-4-fluorophenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester,

1,4-dihydro-4-[3-[[[[3-[4-1,1-dimethylethyl)-1-piperidinyl]propyl]amino]carbonothioyl]amino]-4-fluorophenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester,

1,4-dihydro-4-[3-[[[[3-[4-(1-methylethyl)-1-piperidinyl]propyl]amino]carbonothioyl]amino]-4-fluorophenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester, and

1,4-dihydro-4-[4-[[[[3-(4-cyclohexyl-1-piperazinyl)propyl]amino]carbonothio yl]amino]-4-fluorophenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester.

Ref: U.S. Patent No. 6,391,881

Novel aryl sulfonamide and sulfamide compounds

Ref: U.S. Patent No. 6,391,877

Amine and amide derivative Y receptor antagonist, such as:

Amino-6-[(2-fluorophenylsulfonyl)amino]-N-[cis-1,2,3,4-tetrahydro-6-methoxy-1-(3-pyridinylmethyl)-2-naphthenyl]-(2S)-hexanamide bis-hydrochloride,

N-[5-amino-6-[[cis-1,2,3,4-tetrahydro-6-methoxy-1-(3-pyridinylmethyl)-2-naphthalenyl]amino]hexyl]-2-fluorobenzenesulfonamide tris-hydrochloride,

N-[5-amino-6-[[cis-1,2,3,4-tetrahydro-6-hydroxy-1-(3-pyridinylmethyl)-2-naphthalenyl]amino]hexyl]-2-fluorobenzenesulfonamide tris-hydrochloride,

(2S)-2-(Acetylamino)-6-[(2-fluorophenylsulfonyl)amino]-N-[cis-1,2,3,4-tetra hydro-6-methoxy-1-(3-pyridinylmethyl)-2-naphthenyl]hexanamide bis-hydrochloride,

(2S)-2-(Acetylamino)-6-[(2-fluorophenylsulfonyl)amino]-N-[cis-1,2,3,4-tetra hydro-6-hydroxy-1-(3-pyridinylmethyl)-2-naphthenyl]hexanamide bis-hydrochloride,

3-[(Phenylsulfonyl)amino]-N-[cis-1,2,3,4-tetrahydro-6-fluoro-1-(3-pyridinylmethyl)-2-naphthalenyl]-1-pyrrolidineacetamide bis-trifluoroacetate,

4-Oxo-1-phenyl-N-[cis-1,2,3,4-tetrahydro-1-(3-pyridinylmethyl)-2-naphthalenyl]-1,3,8-triazaspiro[4.5]decane-8-acetamide bis-hydrochloride,

trans-N-[2-(4-fluorophenyl)-3-(3-pyridinyl)propyl]-4-[((2-fluorophenylsulfo nyl)amino)methyl]-1-cyclohexanamide hydrochloride,
trans-N-[[[[[2-(4-fluorophenyl)-3-(3-pyridinyl)propyl]amino]methyl]-4-cyclo hexyl]methyl] 2-fluorobenzenesulfonamide bis-hydrochloride.
Ref: U.S. Patent No. 6,380,224.

Alkylene diamine-substituted pyrazlo (1,5-a)-1,5-pyrimidines and pyrazolo (1,5-a) 1,3,5-triazines, such as:

2-{2-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethylamino}-butan-1-ol;

N-{2-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethyl}-N'-methyl-cyclohexane-1,4-diamine;

N-{2-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethyl}-N'-ethyl-cyclohexane-1,4-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(4-morpholin-4-yl-cyclohexyl)-ethane-1,2-diamine;

4-{2-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethylamino}-cyclohexanol;

3-{2-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethylamino}-propane-1,2-diol;

N-{2-[3(2,6-dichloro-4-methoxy-phenyl)-2,5-dimnethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethyl}-N'-isobutyl-cyclohexane-1,4-diamine;

N-{2-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethyl}-N-isobutyl-cyclohexane-1,4-diamine;

4-{2-[3-(2,6-dichloro-4-ethoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-1-methyl-ethylamino}-cyclohexanol;

2-{2-[3-(2,6-dichloro-4-ethoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-ethylamino}-cyclohexanol;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazol[1,5-a]pyrimidin-7-yl]-N-(4,4,4-trifluoro-butyl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-ethoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(2,2,2-trifluoro-ethyl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(2-trifluoromethyl-cyclohexyl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(4-trifluoromethyl-cyclohexyl)-ethane 1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(2,2-difluoro-ethyl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N- (2-fluoro-1-methyl-ethyl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(2-fluoro-cyclohexyl)-ethane-1,2-diamine.

N-[3-(2,6-dichloro-phenyl)-2,5-dimethyl pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-ethyl-piperidin-5-a]pyrimidin-7-yl]-N -(2,2, 6, 6-tetramethyl-piperidin-4-yl)-ethane-1,2diamine;

N-[3-(2,6-dichloro-phenyl)-2,5-dimethyl-pyrazolo [1,5-a]pyrimidin-7-yl]-N-19 piperidin-4-yl-ethane-1,2-diamine;

N-[3-(2,6-dichloro-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-ethyl-piperidin-3-yl)-ethane-1,2-diamine;

N-(1benzyl-pyrrolidin-3-yl)-N'-[3-(2,6-dichloro-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-ethane-1,2-diamine;

N-[3-(2,6-dichloro-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N'-pyrimidin-2-yl-ethane-1,2-diamine;

N-(1-benzylpiperidin-4-yl)-N'-[3-(2,4-dichloro-6-methoxy-phenyl)-2,5-diethyl-pyrazolo [1,5-a]pyrimidin-7-yl]-ethane-1,2-diamine;

N-(1-benzyl-piperidin-4-yl)-N'-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-ethane-1,2-diamine;

N-[3(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-methyl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5 dimethyl-pyrazolo [1,5-a]pyrimidin-7-yl]-N-(1-ethyl-piperidin-4-yl)-ethane-1,2-di amine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-isopropyl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)2,5-dimethyl-pyrazolo [1,5-a]pyrimidin-7-yl]-N-(2,2,6,6-tetramethyl-piperidin-4-yl)ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-ethyl-piperidin-3-yl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4methoxy-phenyl)-2, 5-dimethyl-pyrazol to [1, 5-a] pyrimidin-7-yl]-N'-piperidin-4-yl-ethane 1,2-diamine;

N.sup.2 -(1-Benzyl-piperidin-4-yl)-N'-[3-(2,6-dichloro-phenyl)-2,5-dimethyl-pyrazo lo[1,5-ajpyrimidin-7-yl]-propane-1,2-diamine;

N-[3-(2,6-Dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N'-(1-pyridin-3-ylmethyl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-Dichloro-4-methoxyphenyl)-2,5-dimethyl-pyrazolo [1,5-a]pyrimidin-7-yl]-N'-(1-pyridin-4-ylmethyl-piperidin4-yl)-ethane-1,2-diamine;

3,5-Dichloro-4-12,5-dimethyl-7-[2-(1-phenyl-pyrrolidin-3-ylamino)-ethylamino]-pyrazolo[1,5-a]pyrimidin-3-yl]-phenol;

N-[3-(2,6-dicloro-4-methoxy-phenyl)-2,5-dimethyl-purazolo[1,5-a]pyrimdin-7-yl]-N'-(1-pyridin-2-ylmethyl-piperidin-4-yl)-ethane-1,2-diamine;

3,5-dichloro-4-(2,5-dimethyl-7-[2-(1-pyrimidin-2-yl-piperidin-4-ylamino)-ethylamino]-pyrazolo[1,5-a]pyrimidin-3-yl}-benzonitrile;

N-[3-(2,6-dichloro-4-ethoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5 a]pyrimidin-7-yl]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-(1-benzyl-piperidin-4-yl)-N'-[3(2,6-dichloro-4-ethoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-ethane-1,2-diamine;

N-[3-(2,6-dichloro-phenyl)-5-ethyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-phenyl)-5isopropyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)ethane-1,2-diamine;

N-[3-(2,4-dichloro-phenyl)-5-isopropyl-2-methyl-pyrazolo[1,5a]pyrimidin-7-y l]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-ethoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-yl-piperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-5isopropyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N2-(1-pyrimidin-2-yl-piperidin-4-yl)propane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-5-ethyl-2-methylpyrazoto [1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-dia mine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2-methyl-5-propyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N -(1-pyrimidin-2-yl-piperidin-4-yl)-propane-1,2-diamine;

N- [3-(2,6-dichloro-4-methoxy-phenyl)5-ethyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-ylpiperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,6-dichloro-phenyl)-2-methyl-5-propylpyrazoto [1,5-a]pyrimidin-7-yl]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-dia mine;

N-[3-(2,6-dichloro-phenyl)-2-methyl-5-propyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N2-(1-pyrimidin-2-yl-piperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,6-dichloro-phenyl)-5-ethyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl] -N.sup.2 -(1-pyrimidin-2-yl-piperidin-4-yl)-propane1,2-diamine;

N-[5-ethyl-2-methyl-3-(2,4,6-trimethyl-phenyl)-pyrazolo[1,5-a]pyrimidin-7-y l]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[5-ethyl-2-methyl-3-(2,4,6-trimethyl-phenyl)-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-yl-piperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,6dichloro-4-ethynyl-phenyl)-2,5-dimethylpyrazolo[1,5-a]pyrimidin-7-yl]-N'-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[2-methyl-5-propyl-3-(2,4,6-trimethyl-phenyl)-pyrazolo [1,5-a]pyrimidin-7-yl]-N'-(1pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[2,5-dimethyl-3-(2,4,6-trimethylphenyl)-pyrazolo[1,5-a]pyrimidin-7-yl]-N' -(1-pyridin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-Dimethyl-phenyl)-5-ethyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl] -N-(1-pyrimidin-2-yl-piperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,6-dimethyl-phenyl)-2-methyl-5-propyl-pyrazolo[1,5-a]pyrimidin-7-yl] –N- (1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,6-Dimethyl-phenyl)-2-methyl-5-propyl-pyrazolo[1,5-a]pyrimidin-7-yl]-NZ-(1-pyrimidin-2-yl-piperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,6dimethyl-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-ylpiperidin-4-yl)-propane-1,2-diamine;

N-[3-(2,4-dimethyl-phenyl)-5-ethyl-2-methyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine;

N-[3-(2,4-dimethyl-phenyl)-2-methyl-5-propyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(1-pyrimidin-2-yl-piperidin-4-yl)-ethane-1,2-diamine; and

1-[4-(1-{[3-(2,6-dichloro-4-methoxyphenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-ylamino]-methyl]-propylamino)piperidin-1-yl]-ethanone.

N-[2,5-dimethyl-3-(2,4,6-trimethylphenyl)-pyrazolo[1,5-a]pyrimidin-7-yl]-N-[2-(4-methoxy-phenyl)-ethyl]-ethane-1,2diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo1,5-a]pyrimidin-7-yl]-N-[2-(4-methoxy-phenyl)-ethyl]-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N'-[2-(3-ethoxy-4-methoxy-phenyl)-ethyl]-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo 1,5-a]pyrimidin-7-yl]-N-[2-(4-ethoxy-3-methoxy-phenyl)-ethyl]-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,a]pyrimidin-7-yl]-N'-(1,2,3,4-tetrahydro-naphthalen-2-yl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(2-pyridin-2-yl-ethyl)-ethane-1,2-diamine;

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo[1,5-a]pyrimidin-7-yl]-N-(2-pyridin-3-yl-ethyl)-ethane-1,2-diamine; and

N-[3-(2,6-dichloro-4-methoxy-phenyl)-2,5-dimethyl-pyrazolo [1,5-a]pyrimidin-7-yl]-N-(2-pyridin4-yl-ethyl)-ethane-1,2-diamine.

Ref: U.S. Patent No. 6,372,743

Spiroisoquinolinone derivative Y antaponist, such as:

2-(3-Chloropropyl)-2-phenyl-1,3-dioxolane,

2-(3-Chloropropyl)-2-(4-methoxyphenyl)-1,3-dioxolane,

2-(3-Chloropropyl)-2-(4-phenoxyphenyl)-1,3-dioxolane,

2-(3-Chloropropyl)-2-(4-bromophenyl)-1,3-dioxolane,

2-(3-Chloropropyl)-2-(4-chlorophenyl)-1,3-dioxolane,

N-3-Chloropropyl-N-methylbenzenemethanamine Hydrochloride,

N-(3-Chloropropyl)-N-(phenylmethyl)benzenemethanamine Hydrochloride,
N-(2-Hydroxyethyl)-N-methylbenzenemethanamine,
Chloro-1-(4-phenoxyphenyl)ethanone,
3-Chloro-1 -(4-phenoxyphenyl)propanone,
1'-[3-(4-Phenoxyphenyl)-3-oxopropyl]spiro[isoquinoline-1-(2H)-4'-piperidine -3-(4H)-one Hydrochloride,
1'-[3-(4-Bromophenyl)-3-oxopropyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3 -(4H)-one],
1'-[2-[(1,1'-Biphenyl)-4-yl]-2-oxoethyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one],
1'-[2-(4-Bromophenyl)-2-oxoethyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one],
1'-[2-(4-Phenoxyphenyl)-2-oxoethyl]spiro[isoquinoline-1-(2H-4'-piperidine-3 -(4H)-one], Hydrochloride,
1'-[2-[Bis(phenylmethyl)amino]ethyl]spiro[isoquinoline-1-(2H)-4'-piperidine -3-(4H)-one] Dihydrochloride,
1'-(4-Phenyl-4-oxobutyl)spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one] Hydrochloride,
1'-[4-(4-Methoxyphenyl)-4-oxobutyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one] Hydrochloride,
1'-[4-(4-Phenoxyphenyl)-4-oxobutyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one] Hydrochloride,
1'-[4-(4-Bromophenyl)-4-oxobutyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one],
1'-[4-(4-Chlorophenyl)-4-oxobutyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3 -(4H)-one] Hydrochloride,
1'-[2-[(1,1'-Biphenyl)-3-yl]-2-oxoethyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one] Hydrochloride,
1'-[3-[(1,1'-Biphenyl)-4-yl]-3-oxopropyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H)-one] Hydrochloride,
1'-[4-[(1,1'-Biphenyl)-4-yl]-4-oxobutyl]spiro[isoquinoline-1-(2H)4'-piperidine-3-(4H)-one] Hydrochloride,

1'-[2-[(1,1'-Biphenyl)-4-yl]-2-hydroxyethyl]spiro[isoquinoline-1-(2H)-4'-piperidine-3-(4H-one] Hydrochloride,
Ref: U.S. Patent No. 6,348,472

Triazine derivative Y receptor antagonists, such as:

N1-{[4-({[4-(Isopropylamino)-6-(methylamino)-1,3,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-[4-([4-(ethylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide)-6-(isopropylamino)-1,3,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-naphthalenesulfenamideN1-{[4-({[4,6-Di(isopropylamino)-1,3,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-[4-([4-(isopropylamino)-6-(propylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl3methyl-1-naphthalenesulfonamide,

N1-[4-([4-(butylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-(cyclobutylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-(cyclopropylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-(isopropylamino)-6-(pentylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-[(2-cyanoethyl)amino]-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-[(2-hydroxyethyl)amino]-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-(4-[(4-(isopropylamino)-6-((2-methoxyethyl)amino]-1,3,5-triazin-2-yl]amino)methyl]cyclohexylmethyl)-1-naphthalenesulfonamide,

N1-(4-[(4-(isopropylamino)-6-[(3-methoxypropyl)amino]-1,3,5-triazin-2-ylamino)methyl]cyclohexylmethyl)-1-naphthalenesulfonamide,

N1-{[4-({[4-}[2-(dimethylamino)ethyl]amino}-6-(isopropylamino)-1,3,5-triazin-2-yl]amino}methyl) cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-[4-([4-[3-(1H-1-imidazolyl)propyl]amino-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-({4-[({4-(isopropylamino)-6-1(4-methoxyphenethyl)amino]-1,3,5-triazin-2- yl}amino)methyl]cyclohexyl}methyl)-1-naphthalenesulfonamide,

N1-{[4-({[4-(dimethylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-[4-([4-[ethyl(methyl)amino]-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-(diethylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl) cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-(isopropylamino)-6-tetrahydro-1H-1-pyrrolyl-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-(4-[(4-(isopropylamino)-6-[(2S)-2-(methoxymethyl)tetrahydro-1H-1-pyrrolyl]-1,3,5-triazin-2-ylamino)methyl]cyclohexylmethyl)-1-naphthalenesulfonamide,

N1-{[4-({[4-(isopropylamino)-6-piperidino-1,3,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-napnthalenesulfonamide,

N1-4-([4-(isopropylamino)-6-(2-methylpiperidino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-[4-([4-(isopropylamino)-6-morpholino-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-{[4-({[4-[(2R,6S)-2,6-dimethyl-1,4-oxazinan-4-yl]-6-(isopropylamino)-1,3 ,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-[4-([4-[(2-hydroxyethyl)(methyl)amino]-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-naphthalenesulfonamide,

N1-{[4-({[4-(4-acetylpiperazino)-6-(isopropylamino)-1,3,5-triazin-2-yl]amnio}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-{[4-({[4-(isopropylamino)-6-(4-isopropylpiperazino)-1,3,5-triazin-2-yl]amino}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N1-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl -4-(tert-butyl)-1-benzenesulfonamide, N1-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl -4-fluoro-1-benzenesulfonamide,

N1-[4-([4,6-di (ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-2-methoxy-5-methyl-1-benzenesulfonamide, N1-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-2-fluoro-1-benzenesulfonamide,

N1-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-2-methyl-1-benzenesulfonamide, N3-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-3-pyridinesulfonamide, N1-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-4-methoxy-1-benzenesulfonamide,

N5-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-2,4-dimethyl-1,3-oxazole-5-sulfonamide, N2-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-2-thiophenesulfonamide, N4-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-methyl-1H-4-imidazolesulfonamide, N1-4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl- 4-methyl-1-benzenesulfonamide, N5-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl -2,1,3-benzothiadiazole-5-sulfonamide, N8-[4-([4,6-di(ethylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl -8-quinolinesulfonamide-yl]aminomethyl)cyclohexyl]methylme thanesulfonamide N1-[4-([4-(isopropylamino)-6-tetrahydro-1H-1-pyrrolyl-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-pyrrolidinesulfonamide,

N4-[4-([4-(isopropylamino)-6-morpholino-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-4-morpholinesulfonamide,

N1-[4-([4-(isopropylamino)-6-piperidino-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-1-piperidinesulfonamide,

N1-[(4-[(4,6-ditetrahydro-1H-1-pyrrolyl-1,3,5-triazin-2-yl)amino]methylcyclohexyl)methyl]-4-(tert-butyl)-1-benzenesulfonamide,

N-cyclopropyl-N'-[4-([4-(cyclopropylamino)-6-(isopropylamino)-1,3,5-triazin -2-yl]aminomethyl)cyclohexyl]methylsulfamide,

N'-[4-([4-(cyclopropylamino)-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohexyl]methyl-N,N-dimethylsulfamide,

N1-{[4-({[4-chloro-6-(isopropylamino)-1,3,5-triazin-2-yl]amnio}methyl)cyclohexyl]methyl}-1-naphthalenesulfonamide,

N'-[(4-[(4,6-dimorpholino-1,3,5-triazin-2-yl)amino]methylcyclohexyl)methyl] -N,N-dimethylsulfamide,

N1-[4-([4-chloro-6-(isopropylamino)-1,3,5-triazin-2-yl]aminomethyl)cyclohex yl]methyl-4-(tert-butyl)-1-benzenesulfonamnide,

N1-[4-([4-(cyclopropylamino)-6-tetrahydro-1H-1-pyrrolyl-1,3,5-triazin-2-yl] aminomethyl)cyclohexyl]methyl-4-fluoro-1-benzenesulfonamide,

N'-((4-(((4,6-dichloro-1,3,5-triazin-2-yl)amino)methyl)cyclohexyl)methyl)-N ,N-dimethylsulfamide,

N1-[(4-[(4,6-ditetrahydro-1H-1-pyrrolyl-1,3,5-triazin-2-yl)amino]methylcyclohexyl)methyl]-2-methoxy-5-methyl-1-benzenesulfonamide,

N1-[4-([4-(cyclopropylamino)-6-(2-pyridyl)-1,3,5-triazin-2-i]aminomethyl)cyclohexyl]methyl-4-fluoro-1-benzenesulfonamide,

N1-[4-(aminomethyl)cyclohexyl]methyl-4-fluoro-1-benzenesulfonamide,

N1-[4-(aminomethyl)cyclohexyl]methyl-4-fluoro-1-benzenesulfonamide,

N2, N4-diethyl-N6-[5-(1H-1-pyrazolyl)pentyl]-1,3,5-triazine-2,4,6-triamine

N2, N4-diethyl-N6-[3-(1H-1-imidazolyl)propyl]-1,3,5-triazine-2,4,6-triamine

N2, N4-diethyl-N6-(2-pyridylmethyl)-1,3,5-triazine-2,4,6-triamine

Ref: U.S. Patent No. 6,340,683

Tricyclic compound Y receptor antagonists, such as:

trans-N2-(4-Dimethylaminosulfonylaminomethyl)cyclohexyl-9-fluoro-5,6-dihydro -4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

1-Aza-9-fluoro-4,5-dihydro-2-{5-(dimethylaminosulfonyl-amino)pentyl}amino-3 -thia-benzo[e]azulene;

1-Aza-9-fluoro-2-(5-(2-fluorophenyl)sulfonylamino)pentylamino-4,5-dihydro-3 -thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(1-naphthyl)sulfonylamino)-pentylamino-3-th ia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(4-(methanesulfonylamino)-butyl)amino-3-thia-b enzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(4-(dimethylaminosulfonyl-amino)butyl)amino-3- thia-benzo[e]azulene;

1-Aza-9-fluoro-2-(4-(2-fluorophenyl)sulfonylamino)butylamino-4,5-dihydro-3- thia-benzo[e]azulene -3-thia -benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(4-((2(S)-methoxymethyl)-pyrrolidine-1-yl)sulfonyl)phenylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(methylsulfonylamino)-pentyl)amino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(methylsulfonylamino-methyl)cyclohexyl)amino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(2,4-difluorophenyl)sulfonylamino)pentylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-isopropylsulfonylamino)-pentylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(diethylaminosulfonyl-amino)pentyl)amino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(2-methoxy-5-methylphenyl)sulfonylamino)pen tylamino-3-thia-benzo[e]azulene;

1-Aza-2-(5-benzylsulfonylamino)pentylamino-9-fluoro-4,5-dihydro-3-thia-benz o[e]azulene;

1-Aza-2-(5-(3,4-difluorophenyl)sulfonylamino)pentylamino-9-fluoro-4,5-dihyd ro-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(4-methoxyphenyl)sulfonylamino)pentylamino- 3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(2-thienyl)sulfonylamino)-pentylamino-3-thi a-benzo[e]azulene;

1-Aza-9-fluoro-2-(5-(2-trifluoroethyl)sulfonylamino)pentylamino-4,5-dihydro -3-thia-benzo[e]azulene;

1-Aza-9-fluoro-2-(5-ethylsulfonylamino)pentylamino-4,5-dihydro-3-thia-benzo[e]azulene;

1-Aza-2-(4-diethylaminosulfonylamino)butylamino-9-fluoro-4,5-dihydro-3-thia -benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(1-methylimidazol-4-yl)sulfonylamino)pentylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(3,5-dimethylisoxazol-4-yl)sulfonylamino)pentylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-aminosulfonylamino)pentylamino-3-thia-benzo[e]azulene;

trans-1-aza-9-fluoro-2-(4-(2-fluorophenyl)sulfonylamino-methyl)cyclohexylam ino-4,5-dihydro-3-thia-benzo[e]-azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-{4-(4-methoxyphenyl)-sulfonylaminomethyl}cyclohexylamino-3-thia-benzo[e]azulene;

trans-N2-(4-(2,6-Difluorophenylsulfonyl)aminomethyl)cyclohexyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta-[d][1,3]-thiazol-2-amine;

trans-1-Aza-2-{4-benzylsulfonylaminomethyl}cyclohexylamino-9-fluoro-4,5-dih ydro-3-thia-benzo[e]azulene;

trans-N2-(4-(2-Thienylsulfonyl)aminomethyl)cyclohexyl-9-fluoro-5,6-dihydro- 4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-N2-(4-Ethylsulfonylaminomethyl)cyclohexyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-1-Aza-9-fluoro-4,5-dihydro-2-{4-(1-methylimidazolyl-4-yl)sulfonylaminomethyl}cyclohexylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-{4-(3,5-dimethylisoxazol-4-yl)sulfonylaminomethyl}cyclohexylamino-3-thia-benzo[e]azulene)-cyclohexylamino- 3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-diethylaminosulfonylamino)-cyclohexylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(4-methoxyphenyl)sulfonylamino)-cyclohexylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(2-thienyl)sulfonyl-amino)-cyclohexylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(2,2,2-trifluoro-ethyl)sulfonylamino)-cyclohexylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(4-(2,2,2-trifluoroethyl)-sulfonylamino)butylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-2-{4-(3,4-difluorophenyl)sulfonyl-aminomethy}cyclohexy lamino-4,5-dihydro-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-2-{4-trifluoromethylsulfonylaminomethyl}cyclohexylamino-4,5-dihydro-3-thiabenzo[e]-azulene;

trans-1-Aza-9-fluoro-2-{4-(2-fluoro)phenylsulfonylamino}-cyclohexylmethylamino-4,5-dihydro-3-thia-benzo[e]azulene;

trans-N2-(4-Methylsulfonylamino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine: A mixture of trans-N2-(4-amino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclo hepta[d][1,3]thiazol-2-aminedihydrochloride;

trans-N2-(4-Aminosulfonylamino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-N2-(4-Amino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-N2-(4-Aminosulfonylamino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

9-Fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine: 6-Bromo-3-fluoro-6,7,8,9-tetrahydro-5H-benzo[a]cyclohepten-5-one;

N1-(9-Fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)-5-bromopentanamide;

1-5-[(9-Fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]-thiazol-2-yl)amino]-5-oxopentyl-1,2-triazadien-2-ium;

N1-(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)-5-aminopentanamide;

N1-(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)-5-[(methylsulfonyl)amino]pentanamide;

trans-N2-(4-Aminosulfonylaminomethyl)cyclohexyl-4,5-dihydro-benzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

trans-N2-(4-Methylsulfonylaminomethyl)cyclohexyl-4,5-dihydro-benzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

trans-1-Aza-4,5-dihydro-2-{4-(2-methoxy-5-methyl)phenyl-sulfonylaminomethyl}cyclohexylamino-6-oxa-3-thia-benzo[e]azulene;

N1-(9-Fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]-thiazol-2-yl)-5-[(2-methoxy-5-methylphenyl)sulfonyl]-aminopentanamide;

N1-(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]-thiazol-2-yl)-5-aminopentanamide;

trans-N2-(4-Methylsulfonylamino)cyclohexylmethyl-4,5-dihydro-benzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

trans-1-Aza-4,5-dihydro-2-{4-(2-methoxy-5-methylphenyl)-sulfonylamino}cyclohexylmethylamino-6-oxa-3-thia-benzo[e]azulene;

trans-N2-(4-Ethylsulfonylamino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-ben zo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-1-Aza-9-fluoro-4,5-dihydro-2-{4-isopropylsulfonylamino}cyclohexylmethylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(3-pyridylsulfonylamino)cyclohexyl)amino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(5-(3-pyridyl)sulfonylamino)pentylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-(4-(3-pyridyl)sulfonylamino)butylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-{2-(2-methylsulfonylamino)ethoxy}ethylamino-3-thia-benzo[e]azulene;

1-Aza-9-fluoro-4,5-dihydro-2-{2-[2-(2-methoxy-5-methylphenyl)sulfonylamino] ethoxy}ethylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(3-pyridyl)sulfonylaminomethyl)cyclohexylamino-3-thia-benzo[e]azulene;

trans-N2-(4-Ethylsulfonylamino)cyclohexylmethyl-8-methoxy-4,5-dihydro-benzo [2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

trans-1-Aza-4,5-dihydro-8-methoxy-2-{4-methylsulfonyl-amino)cyclohexylmethylamino-6-oxa-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-{4-(3-pyridyl)sulfonylamino}cyclohexylmethylamino-3-thia-benzo[e]azulene;

trans-1-Aza-4,5-dihydro-9-methoxy-2-{4-methylsulfonyl-amino}cyclohexylmethylamino-6-oxa-3-thia-benzo[e]azulene;

trans-N2-(4-Ethylsulfonylamino)cyclohexylmethyl-9-methoxy-4,5-dihydro-benzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

trans-N2-(4-Methylsulfonylamino)cyclohexylmethyl-7-methoxy-4,5-dihydro-benzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine hydrochloride;

trans-1-Aza-4,5-dihydro-7-methoxy-2-{4-dimethylaminosulfonylamino}cyclohexylmethylamino-6-oxa-3-thia-benzo[e]azulene;

trans-N2-(4-Dimethylphosphonylamino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4 H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-N2-(4-Ethoxycarbonylamino)cyclohexylmethyl-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine hydrochloride;

1-Aza-9-fluoro-4,5-dihydro-2-(2-(2-isopropylsulfonylamino)-ethoxy)ethylamino-3-thia-benzo[e]-azulene;

2-(4-Methylsulfonylaminomethyl)cyclohexylamino-4H-chromeno[4,3-d]thiazole;

trans-1-Aza-4,5-dihydro-8-methoxy-2-(4-methylsulfonyl-amino)cyclohexylmethylamino-3-thia-benzo[e]-azulene;

trans-1-Aza-4,5-dihydro-8-methoxy-2-(4-methylsulfonylamino-methyl)cyclohexylamino-3-thia-benzo[e]-azulene;

trans-1-Aza-4,5-dihydro-2-(4-isopropylsulfonylaminomethyl)-cyclohexylamino-8-methoxy-3-thia-benzo[e]-azulene;

trans-1-Aza-4,5-dihydro-2-(4-methylsulfonylaminomethyl)-cyclohexylamino-7-m ethoxy-3-thia-benzo[e]-azulene;

trans-1-Aza-4,5-dihydro-2-(4-ethylcarbonylaminomethyl)-cyclohexylamino-9-fl uoro-3-thia-benzo[e]azulen;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(4-morpholinyl)-sulfonylaminomethyl)cyclohexylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(2-methoxy)ethoxy-carbonylaminomethyl)cyclohexylamino-3-thia-benzo[e]azulene 2-methoxyethyl N-(t4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]-cyclohepta[d][1,3]thiazol-2-yl) amino]cyclohexyl}methyl)-carbamate;

tert-butyl N-[(4-{[(benzoylamino)carbothioyl]amino}cyclo-hexyl)methyl]carbamate;

tert-butyl-N-({4-[(aminocarbothioyl)amino]cyclohexyl}-methyl)carbamate;

6-Bromo-3-fluoro-6,7,8,9-tetrahydro-5H-benzo[a]cyclohepten-5-one;

tert-Butyl-N-({4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]-cyclohepta-[d][1,3]th iazol-2-yl)amino]cyclohexyl}methyl)-carbamate;

trans-N2-[4-(Aminomethyl)cyclohexyl]-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(2-methoxy)ethoxy-carbonylaminomethyl)cyclohexylamino-3-thia-benzo[e]azulene 2-methoxyethyl N-({4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)a mino]cyclohexyl}-methyl)carbamate;

trans-N2-(4-(1-Morpholinylsulfonylaminomethyl)cyclohexyl-8-methoxy-5,6-dihy dro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine hydrochloride;

3-({4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)am ino]cyclohexyl}methyl)-1,3-oxazolan-2-one;

2-chloroethyl-N-({4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]-cyclohepta[d][1,3] thiazol-2-yl)amino]cyclohexyl}methyl)-carbamate;

3-({4-[(9-Fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)am ino]cyclohexyl}methyl)-1,3-oxazolan-2-one;

N1-({4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta-[d][1,3]thiazol-2-yl) amino]cyclohexyl}methyl)-2-methoxyacetamide;

N1-({4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]-cyclohepta-[d][1,3]thiazol-2-yl)amino]cyclohexyl}methyl)acetamide;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(N-propylformamido)-methyl)cyclohexylamino-3-thia-benzo[e]azulene;

trans-1-Aza-9-fluoro-4,5-dihydro-2-(4-(N-isopropylformamido)methyl)cyclohex ylamino-3-thia-benzo[e]azulene;

N1-{4-[(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)methyl]cyclohexyl}-2-methoxyacetamide;

Benzyl-N-(4-{[(aminocarbothioyl)amino]methyl}cyclohexyl)-carbamate;

Benzyl-N-{4-[(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]-thiazol-2-ylamino)methyl]cyclohexyl}carbamate;

N2-[(4-aminocyclohexyl)methyl]-4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine

N-{[4-(4,5-Dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)cyclohexyl]methyl}-N-propylformamide;

N1-{[4-(4,5-Dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)cyclohexyl]methyl}propanamide;

N2-{4-[(Propylamino)methyl]cyclohexyl}-4,5-dihydrobenzo-[2,3]oxepino[4,5-d] [1,3]thiazol-2-amine;

N-{[4-(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)cyclohexyl]methyl}-N-propylformamide;

N-{4-[(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)methyl]cyclohexyl}-N-(2-methoxyethyl)formamide;

N2-({4-[(2-methoxyethyl)amino]cyclohexyl}methyl)-4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

N-{4-[(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)methyl]cyclohexyl}-N-(2-methoxyethyl)formamide;

trans-1-Aza-2-(4-(n-(ethyl)formamido)cyclohexyl)methyl-amino-4,5-dihydro-6-oxa-3-thia-benzo[e]azulene;

trans-2-(4-Acetamido)cyclohexylmethylamino-1-aza-4,5-dihydro-6-oxa-3-thia-benzo[e]azulene;

Benzyl-N-[4-({[(benzoylamino)carbothioyl]amino}methyl)-cyclohexyl]carbamate;

Benzyl-N-(4-{[(aminocarbothioyl)amino]methyl}cyclohexyl)-carbamate;

Benzyl-N-{4-[(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]-thiazol-2-ylamino)methyl]cyclohexyl}carbamate;

N2-[(4-aminocyclohexyl)methyl]-4,5-dihydrobenzo[2,3]-oxepino[4,5-d][1,3]thiazol-2-amine

N1-{4-[(4,5-dihydrobenzo[2,3]oxepino[4,5-d][1,3]-thiazol-2-ylamino)methyl]cyclohexyl}acetamide;

N2-{[4-(Ethylamino)cyclohexyl]methyl}-4,5-dihydrobenzo-[2,3]oxepino[4,5-d][1,3]thiazol-2-amine;

N-{4-[(4,5-Dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)methyl]cyclohexyl}-N-ethylformamide;N-(4-[(4,5-Dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)methyl]cyclohexyl}-N-propylformamide;

N2-{[4-(propylamino)cyclohexyl]methyl}-4,5-dihydrobenzo-[2,3]oxepino[4,5-d] [1,3]thiazol-2-amine;

N-{4-[(4,5-Dihydrobenzo[2,3]oxepino[4,5-d][1,3]thiazol-2-ylamino)methyl]cyclohexyl}-N-propylformamide;

N1-{4-[(9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)amino]benzyl}-2-methoxyacetamide;N-{4-[(9-Fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-yl)amino]benzyl}methanesulfonamide;

N2-[4-(Aminomethyl)phenyl]-9-fluoro-5,6-dihydro-4H-benzo[6,7]cyclohepta[d][1,3]thiazol-2-amine

Ref: U.S. Patent No. 6,225,330

Bicyclic compound Y receptor antagonists, such as:

2-(5-Diethylaminosulfonylamino)pentylamino-4-(2-pyridyl)-thiazole hydrogen chloride

4-(2-Pyridyl)-2-(5-(2-thienyl)sulfonylaminopentyl)-amino-thiazole hydrogen chloride

2-(5-(2-Fluorophenyl)sulfonylamino)pentylamino-4-(2-pyridyl)-thiazole hydrogen chloride

2-(5-(4-Methoxyphenyl)sulfonylamino)pentylamino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(3,5-Dimethylisoxazol-4-yl)sulfonylamino)pentylamino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(3,4-Difluorophenyl)sulfonylamino)pentylamino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(2-Methoxy-5-methylphenyl)sulfonylamino)pentylamino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(Benzylsulfonylamino)pentylamino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(Ethylsulfonylamino)pentyl)amino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(Trifluoromethylsulfonylamino)pentyl)amino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(Aminosulfonylamino)pentyl)amino-4-(2-pyridyl)thiazole hydrogen chloride

2-(5-(2-Fluorophenyl)sulfonylamino)pentylamino-4-(3-pyridyl)thiazole hydrogen chloride

2-(5-(3,5-Dimethylisoxazol-4-yl)sulfonylamino)pentylamino-4-(3-pyridyl)thiazole hydrogen chloride

2-(5-(2-Methoxy-5-methyl)phenylsulfonylamino)pentylamino-4-(3-pyridyl)thiazole hydrogen chloride

2-(5-(2-Fluoro)phenylsulfonylamino)pentylamino-4-(4-pyridyl)thiazole hydrogen chloride

2-(5-(3,5-Dimethylisoxazol-4-yl)sulfonylamino)pentylamino-4-(4-pyridyl)thiazole hydrogen chloride 2-(5-(2-Methoxy-5-methyl)phenylsulfonylamino)pentylamino-4-(4-pyridyl)thiazole hydrogen chloride

N1-{5-[(4-Benzo[b]thiophen-2-yl-1,3-thiazol-2-yl)amino]-pentyl}-2-methoxy-5 -methyl-1-benzenesulfonamide

N1-(5-{[4-(5-Chloro-3-methylbenzo[b]thiophen-2-yl)-1,3-thiazol-2-yl]amino}pentyl)-2-methoxy-5-methyl-1-benzene-sulfonamide

N1-(4-{[4-(5-Phenyl-3-isoxazolyl)-1,3-thiazol-2-yl)amino}-pentyl)-2-methoxy-5-methyl-1-benzenesulfonamide

N1-(5-{[4-(3-Thienyl)-1,3-thiazol-2-yl]amino}pentyl)-2-methoxy-5-methyl-1-benzenesulfonamide

N1-[5-({4-[1-(Phenylsulfonyl)-1H-3-pyrrolyl]-1,3-thiazol-2-yl}amino)pentyl] -2-methoxy-5-methyl-1-benzenesulfonamide

trans-N8-[(4-{[4-(3-Phenyl-5-isoxazolyl)-1,3-thiazol-2-yl]amino}cyclohexyl) methyl]-8-quinolinesulfonamide

N,N-Dimethyl-N'-(5-{[4-(3-Thienyl)-1,3-thiazol-2-yl]amino}pentyl)sulfamide

trans-2-(4-(2-Methoxy-5-methylphenyl)sulfonylamino)cyclohexylmethylamino-4- (2-pyridyl)thiazole dihydrogen chloride

trans-2-(4-(2-Fluorophenyl)sulfonylamino)cyclohexylmethyl-amino-4-(2-pyridyl)thiazole dihydrogen chloride

trans-2-(4-(3,5-Dimethyl-4-isoxazolyl)sulfonylamino)cyclohexylmethylamino-4-(2-pyridyl)thiazole dihydrogen chloride

trans-2-(4-(2-Fluorophenyl)sulfonylamino)cyclohexylmethyl-amino-4-(3-pyridyl)thiazole dihydrogen chloride

trans-2-(4-(2-Methoxy-5-methylphenyl)sulfonylamino)cyclohexylmethylamino-4-(4-pyridyl)thiazole dihydrogen chloride

N1-(5-[4-(1,3-thiazol-2-yl)-1,3-thiazol-2-yl]aminopentyl)-2-methoxy-5-methy l-1-benzenesulfonamide

trans-N1-[(4-[4-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminocyclo hexyl)methyl]-2-methoxy-5-methyl-1-benzenesulfonamide

trans-N,N-dimethyl-N'-[(4-[4-(-1,3-thiazol-2-yl)-1,3-thiazol-2-yl]aminocyclohexyl)methyl]sulfamide

N,N-Dimethyl-N'-(5-{[4-(2-thienyl)-1,3-thiazol-2-yl]amino}-pentyl)sulfamide

N1-(5-{[4-(2-Thienyl)-1,3-thiazol-2-yl]amino}pentyl)-2-methoxy-5-methyl-1-b enzenesulfonamide

N1-(5-[4-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminopentyl)-2-methoxy-5-methyl-1-benzenesulfonamide

N1-(5-[4-(2,5-Dimethyl-1,3-thlazol-4-yl)-1,3-thiazol-2-yl]aminopentyl)-4-fluoro-1-benzenesulfonamide

N1-(5-[4-(1,3-Thiazol-2-yl)-1,3-thiazol-2-yl]aminopentyl)-4-fluoro-1-benzenesulfonamide

N'-(5-[4-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminopentyl)-N,N-dimethylsulfamide

trans-N1-[(4-[4-(2,5-dimethyl-1,3-thiazol-4-yl])-1,3-thiazol-2-yl]aminocyclohexyl)methyl]-4-fluoro-1-benzene-sulfonamilde

trans-N'-[(4-[4-(2,5-dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminocyclohexyl)methyl]-N,N-dimethylsulfamide

trans-N'-[4-([5-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminomethyl)cyclohexyl]methyl-N,N-dimethyl-sulfamide

trans-N4-[4-([4-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminomethyl)cyclohexyl]methyl-4-morpholine-sulfonamide

trans-N-[4-([4-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminomethyl)cyclohexyl]-N-(2-methoxyethyl)formamide

trans-N-[4-([4-(2,5-Dimethyl-1,3-thiazol-4-yl)-1,3-thiazol-2-yl]aminomethyl)cyclohexyl]-N-isopropylformamide

Ref: U.S. Patent No. 6,218,408

N-aralkylaminotetralin Y receptor antagonist, such as:

rac-cis-1-(Phenylmethyl)-6-methoxy-N-(2-(3,4-dimethoxyphenyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine;

rac-cis-1-(Phenylmethyl)-6-methoxy-N-(2-(3-indolyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine hemifumarate;

rac-cis-1-(Phenylmethyl)-N-(4-fluorophenylmethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monohydrobromide;

rac-cis-1-(Phenylmethyl)-N-(2-methoxyphenylmethyl)-1,2,3,4-tetrahydro-2-naphthalenamine;

rac-cis-1-(Phenylmethyl)-N-(2-methoxyphenylmethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monohydrobromide;

rac-cis-1-(4-Fluorophenylmethyl)-N-(2-methoxyphenylmethyl)-1,2,3,4-tetrahyd ro-2-naphthalenamine monohydrobromide;

rac-trans-1-(4-Fluorophenylmethyl)-N-(2-methoxyphenylmethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monooxalate;

rac-cis-1-(Phenylmethyl)-N-(4-fluorophenylmethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monohydrobromide;

rac-cis-1-(Phenylmethyl)-7-methoxy-N-(2-methoxyphenylmethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monohydrobromide;

rac-trans-1-(4-Fluorophenylmethyl)-N-(2-(3-indolyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monooxalate;

rac-cis-1-(Phenylmethyl)-N-(2-methoxyphenyl-2-oxomethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monohydrobromide;

rac-cis-1-(Phenylmethyl)-7-methoxy-N-(2-(3-indolyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine 0.8 fumarate 0.8 methanol 0.2 hydrate;

rac-trans-1-(Phenylmethyl)-7-methoxy-N-(2(3-indolyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monooxalate;

rac-cis-1-(2-Naphthylmethyl)-N-(2-(3-indolyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine hemifumarate methanol;

rac-trans-1-(2-Naphthylmethyl)-N-(2-(3-indolyl)ethyl)-1,2,3,4-tetrahydro-2-naphthalenamine monooxalate;

rac-cis-1-(2-Naphthylmethyl)-N-(2-methoxyphenylmethyl)-1,2,3,4-tetrahydro-2 -naphthalenamine monohydrobromide;

rac-cis-1-(Phenylmethyl)-N-(2-methoxyphenyl-2-oxoethyl)-1,2,3,4-tetrahydro- 2-naphthalenamine;

rac-cis-1-(4-Fluorophenylmethyl)-N-(3-phenylpropyl)-1,2,3,4-tetrahydro-2-na phthalenamine monohydrobromide;

rac-cis-1-(3-pyridylmethyl)-N-(2-(3,4-dimethoxyphenyl)ethyl-1,2,3,4-tetrahydro-2-naphthalenamine monohydrobromide
Ref: U.S. Patent No. 6,201,025

Amide derivative Y receptor antagonist:
Ref: U.S. Patent No. 6,048,900

N-substituted aminotetralin Y receptor antagonist, such as:

rac-[1α,2α(trans)]-N-[[[[[1,2,3,4-tetrahydro-6-methoxy-1-(phenylmethyl)-2-naphthalenyl]amino]methyl]4-cyclohexyl]methyl]2-naphthalenesulfo namide;
rac-[1α,2α(trans)]-N-[[[[[1,2,3,4-tetrahydro-6-methoxy-1-(phenylmethyl)-2-naphthalenyl]amino]-5-pentyl]2-naphthalenesulfonamide;

rac-[1α,2α(trans)]-N-[[[[[1,2,3,4-tetrahydro-6-methoxy-1-(3-pyridinylmethyl)-2-naphthalenyl]amino]methyl]-4-cyclohexyl]methyl]2-naphthalenesulfonamide;

rac-[1α,2α(trans)]-N-[[[[[1,2,3,4-tetrahydro-6-fluoro-1-(phenylmethyl)-2-naphthalenyl]amino]methyl]-4-cyclohexyl]methyl]2-fluorobenzenesulfonamide;
rac-[1α,2α (trans)]-N-[[[[[1,2,3,4-tetrahydro-6-fluoro-1-phenyl-2-naphthalenyl]amino]methyl]-4-cyclohexyl]methyl]2-naphthalenesulfonamide;
rac-[1α,2α(trans)]-N-[[[[[1,2,3,4-tetrahydro-6-methoxy-1-(1-propene-3-yl)-2-naphthalenyl]amino]methyl]4-cyclohexyl]methyl] benzenesulfonamide;

rac-[1α,2α(trans)]-N-[[[[[1,2,3,4-tetrahydro-6-methoxy-1-(3-hydroxypropyl)-2-naphthalenyl]amino]methyl]-4-cyclohexyl]methyl] benzenesulfonamide;

rac-[1α,2 α (trans)]-N-[[[[[1,2,3,4-tetrahydro-6-methoxy-1-(n-propyl)-2-naphthalenyl]amino]methyl]-4-cyclohexyl]methyl] benzenesulfonamide.
Ref: U.S. Patent No. 6,140,354

4-phenyl-1,4-dihydropyrimidinone derivative Y receptor antagonist:
Ref: U.S. Patent No. 5,889,016

Piperidine derivative dihydropyridine Y receptor antagonist:

4-Dihydro-[3-[[[[3-[4-(3-methoxyphenyl)-1-piperidinyl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-hydroxy-4-(3-methoxyphenyl)piperidin-1-yl]propyl] amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-(2-methoxyphenyl)piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-(4-phenylpiperidin-1-yl)propyl]amino]carbonyl]amino] phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1. 4-Dihydro-4-[3-[[[[3-(4-hydroxy-4-phenylpiperidin-1-yl)propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-2,6-dimethyl-4-[3-[[[[3-[4-[3-(2-propynyloxy)phenyl]-1-piperidinyl]propyl]amino]carbonyl]amino]phenyl]-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-cyano-4-phenylpiperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-(3-hydroxyphenyl)piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-naphthalen-1-ylpiperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

4-[3-[[[[3-[4-(1,1'-Biphenyl-3-yl)piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-1,4-dihydro-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-(phenylmethyl)-piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

4-[3-[[[[3-(4-cyclohexyl-1-piperidinyl)propyl]amino]carbonyl]amino]phenyl]-1,4-dihydro-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-dihydro-4-[3-[[[[3-[4-hydroxy-4-(2-phenoxyphenyl)-1-piperidinyl]propyl] amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-(4-phenyl-1-piperidinyl)propyl]amino]carbonyl]amino] phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, ethyl methyl ester;

1,4-Dihydro-4-[3-[[[[3-[(4-phenylmethyl)-1-piperidinyl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, ethyl methyl ester;

1,4-Dihydro-4-[3-[[[3-[4-hydroxy-4-(2-methoxyphenyl)-piperidin-1-yl]propyl] amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, ethyl methyl ester;

1. 4-Dihydro-4-[3-[[[[3-[4-hydroxy-4-(3-methoxyphenyl)-piperidin-1-yl]propy l]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, ethyl methyl ester;

1,4-Dihydro-2,6-dimethyl-4-[3-[[[[3-[4-[3-(2-propoxy)phenyl]-1-piperidinyl]-propyl]amino]carbonyl]amino]phenyl]3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[2-[4-(3-methoxyphenyl)-1-piperidinyl]ethyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester hydrochloride;

1,4-Dihydro-4-[3-[[[[4-[4-(3-methoxyphenyl)-1-piperidinyl]butyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester hydrochloride;

1,4-Dihydro-4-[3-[[[[3-[4-(3-methoxyphenyl)-1-piperidinyl]propyl]methylamino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester hydrochloride;

4-Dihydro-4-[3-[[[[3-[1,2,3,6-tetrahydro -4-(3-methoxyphenyl)pyridin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-d imethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-(1,2,3,6-tetrahydro-4-phenylpyridin-1-yl)propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[1,2,3,6-tetrahydro-4-(3-hydroxyphenyl)pyridine]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridine dicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[1,2,3,6-tetrahydro-4-(1-naphthalenyl)-1-pyridinyl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[3-(4-phenylpiperidin-1-yl)-1-oxo-1-propyl]amino]phenyl]- 2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1. 4-Dihydro-4-[3-[[4-(4-phenylpiperidin-1-yl)-1-oxo-1-butyl]amino]phenyl]- 2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[5-(4-phenylpiperidin-1-yl)-1-oxo-1-pentyl]amino]phenyl]- 2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[6-(4-phenylpiperidin-1-yl)-1-oxo-1-hexyl]amino]phenyl]-2 ,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[5-(4-hydroxy-4-phenylpiperidin-1-yl)-1-oxo-1-pentyl]amin o]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[5-(4-cyano-4-phenylpiperidin-1-yl)-1-oxo-1-pentyl]amino] phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[4-[4-(3-methoxyphenyl)-1-piperidinyl]butyl]carbonyl]ami no]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-dihydro-4-[3-[[[[3-[4-(3-methoxyphenyl)-1-piperidinyl]propyl]oxy]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester hydrochloride;

1,4-Dihydro-4-[3-[[[[3-[4-(3-methoxyphenyl)piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-(2-methoxyphenyl)piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-(3-hydroxyphenyl)piperidin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[4-naphthalenylpiperidin-1-yl]propyl]amino]carbonyl] amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

4-[3-[[[[3-(4-cyclohexyl-1-piperidinyl)propyl]amino]carbonyl]amino]phenyl]- 1,4-dihydro-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[[3-[1,2,3,6-tetrahydro-4-(3-methoxyphenyl)pyridin-1-yl]propyl]amino]carbonyl]amino]phenyl]-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester;

1,4-Dihydro-4-[3-[[[3-[1,2,3,6-tetrahydro-4-(1-naphthalenyl)pyridin-1-yl]propyl]amino]carbonyl]amino]phenyl-2,6-dimethyl-3,5-pyridinedicarboxylic acid, dimethyl ester.

Ref: U.S. Patent No. 5,668,151

The invention is illustrated by the following non-limiting Examples.

EXAMPLES

Example 1

Material and Methods

Generation of POMC-EGFP mice: The *EGFP* cassette contains its own Kozak consensus translation initiation site along with *SV40* polyadenylation signals downstream of the *EGFP* coding sequences directing proper processing of the 3' end of the EGFP mRNA. The *EGFP* cassette was introduced by standard techniques into the 5' untranslated region of exon 2 of a mouse *Pomc* genomic clone containing 13 kb of 5' and 2 kb of 3' flanking sequences (Young et al., *J Neurosci* 18, 6631-40, 1998). The transgene was microinjected into pronuclei of one-cell stage embryos of C57BL/6J mice (Jackson Laboratories) as described (Young et al., *J Neurosci* 18, 6631-40, 1998). One founder was generated and bred to wildtype C57BL/6J to produce N_1 hemizygous mice. In addition, N_2 and subsequent generations of mice homozygous for the transgene were also generated. The mice are fertile and have normal growth and development.

Immunofluorescence and GFP co-localization: Anesthetized mice were perfused transcardially with 4% paraformaldehyde and free-floating brain sections prepared with a vibratome. Sections were processed for immunofluorescence and colocalization of GFP fluorescence using standard techniques. Primary antisera and their final dilutions were rabbit anti-β-endorphin, 1:2500 v/v; rabbit anti-NPY, 1:25,000 v/v (Alanex Corp.); rabbit anti-ACTH, 1:2000 v/v; and mouse anti-TH, 1:1000 v/v (Incstar). After rinsing, sections were incubated with 10mg/ml biotinylated horse anti-mouse/rabbit IgG (Vector Laboratories) followed by Cy-3 conjugated streptavidin, 1:500 v/v (Jackson Immunoresearch Laboratories). Photomicrographs were taken on a Zeiss Axioscop using FITC and RITC filter sets (Chroma Technology Corp.).

Electrophysiology (Example 2): 200 μm thick coronal slices were cut from the ARC of four-week old male POMC-EGFP mice. Slices were maintained in (in

mM) [NaCl, 126; KCl, 2.5; $MgCl_2$, 1.2; $CaCl_2.2H_2O$, 2.4; $NaH_2PO_4.H_2O$, 1.2; $NaHCO_3$, 21.4; Glucose, 11.1] (Krebs) at 35°C and saturated with 95% O_2 5% CO_2 for 1 hour(hr) prior to recordings. Recordings were made in Krebs at 35° C. Slices were visualized on an Axioskop FS2 (Zeiss) through standard infra red optics and using epifluorescence through a FITC filter set (see Fig. 1c). Whole cell recordings were made from fluorescent neurons using an Axopatch 1D amplifier (Axon Instruments) and Clampex 7 (Axon Instruments). Resting membrane potentials were determined using an event detection protocol on a PowerLab system (AD Instruments, Mountain View, CA) to average expanded traces of the membrane potential. Drugs were applied to the bath over the times indicated. The resting membrane potential was stable for up to an hour in cells treated with Krebs alone. I-V relationships for the Met-Enk currents were established using a step protocol; (–60 mV holding potential, sequentially pulsed (40 ms) from –120 to –50 mV, cells were returned to –60 mV for 2 s between voltage steps). The protocol was repeated after Met Enk addition. The net current was the difference between the two I-V relationships. This protocol was repeated in Krebs with 6.5 mM K^+. I-V relationships to identify the postsynaptic leptin current were performed similarly with slow voltage ramps (5 mV/ s from –100 to –20 mV) before and 10 minutes after the addition of leptin (100 nM). GABAergic IPSCs were recorded using a CsCl internal electrode solution (in mM) [CsCl, 140; Hepes, 10; $MgCl_2$, 5; Bapta, 1; (Mg)-ATP, 5; (Na)GTP, 0.3]. Both mini IPSCs and large amplitude (presumably multisynaptic) IPSCs were observed in the untreated slices. TTX (1 μM) abolished large IPSCs. Data were acquired before and after addition of drug for the times indicated on the figures at a –50 mV holding potential in 2 s. sweeps every 4 s. Mini postsynaptic currents were analyzed using Axograph 4 (Axon Instruments). IPSCs and excitatory postsynaptic currents (EPSCs) were distinguished on the basis of their decay constants; additionally picrotoxin (100 μM) blocked all IPSCs. POMC neurons receive a low EPSC tone and the frequency was not modulated by any of the treatments described here.

Immunostaining for light and electron microscopy: Double immunocytochemistry for NPY and POMC using different color

diaminobenzidine(DAB) chromogens was carried out on fixed mouse hypothalami according to published protocols (Horvath et al., *Neuroscience* 51, 391-9, 1992). For electron microscopy, preembedding immunostaining for β-endorphin was using an ABC Elite kit (Vector Laboratories) and a DAB reaction followed by post-embedding labeling of GABA and NPY using rabbit anti-GABA, 1:1000 v/v and gold conjugated (10 nm) goat anti-rabbit IgG or sheep anti-NPY and gold conjugated (25 nm) goat anti-sheep IgG. Finally, sections were contrasted with saturated uranyl acetate (10 minutes) and lead citrate (20-30 s) and examined using a Philips CM-10 electron microscope.

Animals: Male *Pomc-EGFP* mice were studied at 5-6 weeks of age and were generated as described above. Male mice aged 8-12 weeks and between 20-30 g bodyweight were kept under controlled temperature (21-23°C) and light conditions (lights on 06:00-18:00) with *ad libitum* access to water and food except where stated. All studies were performed in the early light-phase (0700-0800).

Intraperitoneal injections: Mice were accustomed to IP injection by injections of 0.5 ml saline on the two days prior to study. For all studies, animals received an IP injection of either PYY_{3-36} or saline in 100 µl .

Electrophysiology: Whole cell patch clamp recordings were made from POMC neurons in the hypothalamus of 180 µm thick coronal slices from Pomc-EGFP mice, as previously reported (Cowley et al., *Nature* 411, 480-484, 2001). "Loose cell-attached" recordings were made using extracellular buffer in the electrode solution, and maintaining seal resistance between 3-5Mohm throughout the recording. Firing rates were analyzed using mini-analysis protocols (MiniAnalysis, Jaejin Software, NJ). Vehicle controls were used in this system, previously validated for the electrophysiological actions of neuropeptides (Cowley et al., *Nature* 411, 480-484, 2001). Data were analyzed by ANOVA, Neuman-Keuls posthoc comparison, and Wilcoxon Signed Rank Test.

C-fos expression: C-fos expression was measured in Pomc-EGFP mice 2 hours after IP administration of saline or PYY_{3-36} (5μg/100g) using standard immunohistochemical techniques (Hoffman et al., *Front. Neuroendocrinol.* 14, 173-213, 1993). Data were obtained from 5 mice in each group. For the Pomc-EGFP mice 5 anatomically matched arcuate nucleus sections (Franklin et al., *The Mouse Brain in Stereotaxic Coordinates* (Academic Press, San Diego, 1997) were counted from each animal, and images acquired using a Leica TSC confocal microscope (Grove et al., *Neuroscience* 100, 731-40, 2000).

Measurements of Energy Expenditure: To determine the actions of PYY on energy expenditure the OXYMAX system (Columbus Instruments, Columbus, OH) is utilized with rodents following PYY injection into a treatment cohort. This system is also utilized with rodents following a saline injection (control cohort). The equipment measures O_2 consumption and CO_2 production; the efficiency with which the body produces CO_2 from O_2 gives a reliable index of caloric or metabolic efficiency. A similar system is used with human volunteers.

Example 2

Neural Network in the Arcuate Nucleus

A strain of transgenic mice was generated expressing green fluorescent protein (EGFP Clontech), under the transcriptional control of mouse *Pomc* genomic sequences that include a region located between –13 kb and –2 kb required for accurate neuronal expression (Young et al., *J Neurosci* 18, 6631-40, 1998) (Fig. 1a). Bright green fluorescence (509 nm) was seen in the two CNS regions where POMC is produced: the ARC and the nucleus of the solitary tract. Under ultraviolet (450–480 nm) excitation POMC neurons were clearly distinguished from adjacent, non-fluorescent neurons (Fig. 1b) visualized under infrared optics. Double immunofluorescence revealed >99% cellular co-localization of EGFP and POMC peptides within the ARC (Fig. 1c). There was close apposition of both tyrosine hydroxylase (TH)- and NPY-stained terminals on EGFP-expressing POMC neurons, but no evidence of co-localization of the TH or NPY immunoreactivity with EGFP. Total fluorescent cell counts performed on coronal hypothalamic sections revealed 3148 ± 62 (mean ± SEM: n=3) POMC-EGFP neurons distributed through the entire

ARC (Franklin et al., *The Mouse Brain in Stereotaxic Coordinates* (Academic Press, San Diego, 1997) (Fig. 1d). POMC neurons in the mouse are located both medially and ventrally within the ARC, in contrast to a predominantly lateral position in the rat ARC.

POMC-EGFP neurons in hypothalamic slices had a resting membrane potential of –40 to –45 mV and exhibited frequent spontaneous action potentials. The non-selective opioid agonist met-enkephalin (Met-Enk: 30 μM; Sigma) caused a rapid (35- 40 s), reversible hyperpolarization (10-20 mV) of the membrane potential of POMC cells (n=10) and prevented spontaneous action potential generation (Fig. 2a). In normal (2.5 mM K^+) Krebs buffer, the reversal-potential of the inwardly-rectifying opioid current was approximately –90mV, while in 6.5 mM K^+ Krebs the reversal-potential was shifted to approximately –60 mV (n=3: Fig. 2b). The μ opioid receptor (MOP-R) antagonist CTAP (1 μM; Phoenix Pharmaceuticals) completely prevented the current induced by Met-Enk in POMC cells (n=3: Fig. 2c). These characteristics indicate the opioid current was due to activation of MOP-R and increased ion conductance through G protein coupled, inwardly-rectifying potassium channels (GIRK) (Kelly et al., *Neuroendocrinology* 52, 268-75, 1990). The similar opioid responses in EGFP-labeled POMC neurons to that of guinea pig (Kelly et al., *Neuroendocrinology* 52, 268-75, 1990) or mouse (Slugg et al., *Neuroendocrinology* 72, 208-17, 2000). POMC cells, identified by post-recording immunohistochemistry, suggests that expression of the EGFP transgene does not compromise either expression of receptors nor their coupling to second messenger systems in POMC neurons.

Next, the direct effects of leptin on identified POMC cells in slice preparations were investigated. Leptin (0.1 – 100 nM) depolarized 72 of 77 POMC cells by 3-30 mV (Fig. 3a; mean ± SEM depolarization at 100 nM leptin = 9.7 ± 1.2 mV, n= 45) within 2-10 minutes, in a concentration responsive manner (Fig. 3b). There were two components to the depolarization and neither were fully reversible within 40 minutes. Firstly, the depolarization was due to a small inward current which reversed at approximately -20 mV (Fig. 3c), suggesting the involvement of a non-specific cation channel (Powis et al., *Am J Physiol* 274, R1468-72, 1998). Secondly, leptin treatment decreased the GABAergic tone onto POMC cells.

GABAergic inhibitory postsynaptic currents (IPSCs) were observed in POMC cells and leptin (100 nM) decreased their frequency by 25% (Fig. 3d) in 5 out of 15 cells suggesting that it acted presynaptically to reduce GABA release (leptin had no effect on IPSCs in10 out of 15 POMC neurons). The effect on IPSC frequency occurred with a similar lag to the effect on membrane potential. Thus, leptin not only directly depolarizes POMC neurons but also acts at GABAergic nerve terminals to reduce the release of GABA onto POMC neurons, allowing them to adopt a more depolarized resting potential. The consistent depolarization of POMC cells by leptin was specific because leptin had no effect on 5 of 13 adjacent non-fluorescent cells tested (Fig. 3e), while it hyperpolarized 5 (Fig. 3f) and depolarized 3 other non-POMC neurons in the ARC. The electrophysiological effects of leptin reported here are consistent with leptin's biological actions; leptin rapidly causes release of α-MSH from rat hypothalami (Kim et al., *J Clin Invest* 105, 1005-11, 2000), presumably by activating POMC neurons.

Previous reports of neuronal hyperpolarization by leptin (Glaum et al., *Mol Pharmacol* 50, 230-5, 1996; Spanswick et al., *Nature* 390, 521-5, 1997), and the demonstrated co-localization of GABA and NPY (Horvath et al., *Brain Res* 756, 283-6, 1997) within subpopulations of ARC neurons, suggested that leptin hyperpolarizes NPY/GABA cells that directly innervate POMC neurons, and thus reduces GABAergic drive onto POMC cells. Both the leptin and NPY Y2 receptors are expressed on NPY neurons in the ARC (Hakansson et al., *J Neurosci* 18, 559-72, 1998; Broberger et al., *Neuroendocrinology* 66, 393-408, 1997). Furthermore, activation of Y2 receptors inhibits NPY release from NPY neurons (King et al., *J Neurochem* 73, 641-6, 1999), and presumably would also diminish GABA release from NPY/GABA terminals. This provides an alternative pharmacological approach, independent of leptin, to test the hypothesized innervation of POMC neurons by GABAergic NPY neurons. Indeed, NPY (100 nM; Bachem) decreased the frequency of GABAergic IPSCs by 55% within 3 minutes, in all 12 POMC cells tested (Fig. 4a). Both NPY and leptin still inhibited IPSCs in the presence of tetrodotoxin (TTX) (6 of 6 and 3 of 5 cells respectively), indicating that some of the inhibition of IPSCs was occurring through direct effects at presynaptic nerve terminals. POMC neurons express the NPY Y1 receptor (Broberger et al.,

Neuroendocrinology 66, 393-408, 1997) and NPY also hyperpolarized all POMC neurons tested, by an average of 9±6 mV (n=3).

Another pharmacological test to confirm the origin of GABAergic innervation on POMC neurons from NPY/GABA terminals was to test the effect of the recently characterized and highly selective MC3-R agonist D-Trp^8-γMSH (Grieco et al., *J Med Chem* 43, 4998-5002, 2000) on local GABA release. D-Trp^8-γMSH (7 nM) increased the frequency of GABAergic IPSCs (280 ± 90%) recorded from 3 of 4 POMC neurons (Fig. 4b). It had no effect on one cell. The positive effect of MC3-R activation, together with the negative effects of NPY and leptin, demonstrate the dynamic range of the NPY/GABA synapse onto POMC neurons and point to the important role of this synapse in modulating signal flow within the ARC. D-Trp^8-γMSH (7 nM) also hyperpolarized (-5.5 ± 2.4 mV) 9 of 15 POMC neurons tested and decreased the frequency of action potentials (Fig 4c); the remaining cells showed no significant response to D-Trp^8-γMSH. These effects could be due entirely to increased GABA release onto the POMC cells, or could be due to an additional postsynaptic action of D-Trp^8-γMSH on POMC neurons, approximately half of which also express the MC3-R (Bagnol et al., *J Neurosci (Online)* 19, RC26, 1999). Thus, MC3-R acts in a similar autoreceptor manner to MOP-Rs on POMC neurons, diminishing POMC neuronal activity in response to elevated POMC peptides.

To further determine that the IPSCs in POMC neurons were due to local innervation by NPY/GABA cells, multi-label immunohistochemistry was performed using light and electron microscopy. Although independent NPY (Csiffary et al., *Brain Res* 506, 215-22, 1990) and GABA (Horvath et al., *Neuroscience* 51, 391-9, 1992) innervation of POMC cells has been reported, co-localization of NPY and GABA in nerve terminals forming synapses onto POMC cells has not been shown. Similar to the rat (Csiffary et al., *Brain Res* 506, 215-22, 1990), a dense innervation of POMC cells by NPY axon terminals was detected in the mouse (Fig. 4d). Electron microscopy confirmed the coexpression of NPY and GABA in axon terminals and revealed that these boutons established synapses on the perikarya of all 15 ARC POMC neurons analyzed (representative example, Fig. 4e).

A detailed model of regulation of this circuit shows dual mechanisms of leptin action in the ARC, interactions between NPY/GABA and POMC neurons, and autoregulatory feedback from opioid and melanocortin peptides as well as NPY (Fig. 4f). In this model, leptin directly depolarizes the POMC neurons and simultaneously hyperpolarizes the somata of NPY/GABA neurons, and diminishes release from NPY/GABA terminals. This diminished GABA release disinhibits the POMC neurons, and result in an activation of POMC neurons and an increased frequency of action potentials.

Example 3

Administration of PYY Inhibits Food Intake

The effect of PYY on feeding in rats, and mice has been established (Batterham et al., *Nature* 418:450, 2002).

Example 4

PYY Administration Affects c-fos Expression

To investigate whether this inhibition of food intake involved a hypothalamic pathway, c-fos expression was examined in the arcuate nucleus, an important center of feeding control (Schwartz et al., *Nature* 404, 661-671, 2000; Cowley et al., *Nature* 411, 480-484, 2001), following a single IP injection of PYY_{3-36} (Fig. 5). There was a 2-fold increase in the number of cells positive for c-fos in the lateral arcuate of the rat (PYY_{3-36} =168 ± 2, saline = 82.7 ± 5, n = 3, $P < 0.0001$). Likewise in *Pomc-EGFP*-transgenic mice (Cowley et al., *Nature* 411, 480-484, 2001) IP administration of PYY_{3-36} resulted in a 1.8-fold increase in the number of arcuate cells positive for c-fos (Fig. 5b), compared with saline control animals (Fig. 5a) (PYY_{3-36} = 250 ± 40, saline = 137 ± 15, n = 5, $P < 0.05$). IP PYY_{3-36} caused a 2.6 fold increase in the proportion of POMC neurons that express c-fos (PYY_{3-36} = 20.4 ± 2.9%, saline = 8 ± 1.4%, n = 5, $P < 0.006$) (Figs. 5c and d). These observations show that PYY_{3-36} can act via the arcuate nucleus

Example 5

Y2 receptors

The electrophysiological response of hypothalamic POMC neurons to administration of both PYY_{3-36} and Y2A was examined. The effect of PYY on feeding in rats and mice has been established (Batterham et al., *Nature* 418:450, 2002). POMC neurons were identified using mice with targeted expression of green fluorescent protein in POMC neurons (Cowley et al., *Nature* 411, 480-484, 2001). PYY_{3-36} disinhibited the POMC neurons, resulting in a significant depolarization of 19 of the 22 POMC neurons tested (Fig. 6a inset) (10.3 ± 2.1 mV depolarization, n = 22, $P < 0.0003$). A similar depolarization was seen with Y2A (8.7 ± 1.8 mV depolarization, n = 9, $P < 0.002$). The depolarization caused by PYY_{3-36} stimulated a significant increase in the frequency of action potentials in POMC neurons (Fig 6a) (93% increase over control, $P < 0.05$, n = 22). In the whole cell mode the effect of PYY_{3-36} was sometimes reversed upon washout, but only after a long latency (30 minutes). A similar washout of leptin effects upon these neurons was observed.

To exclude effects of cellular rundown, or seal deterioration, the effects of PYY_{3-36} in the "loose cell-attached" (or extracellular) configuration was examined. PYY_{3-36} caused a reversible 5-fold increase in the frequency of action potentials in loose cell-attached recordings of POMC neurons (Fig. 6b). This increase in firing rate occurred with the same latency as PYY_{3-36} reduced the frequency of inhibitory postsynaptic currents (IPSCs) onto all 13 POMC neurons tested (Fig. 6c) (51.9 ± 9.2 % reduction, n = 13, $P < 0.0001$), indicating a reduced frequency of GABA release onto POMC neurons. Interestingly, the firing rate of POMC neurons returned to basal, in spite of continued inhibition of IPSCs. A similar effect upon IPSC frequency was seen with Y2A (44.4 ± 9.3% reduction, n = 8, $P < 0.004$) suggesting this effect to be via Y2R. PYY_{3-36} (25 nM) caused a hyperpolarization (5.2 ± 1.16 mV, $P < 0.004$, n = 5) of unidentified, but presumably NPY-containing, non-POMC, neurons in the arcuate nucleus. There is a tonic GABAergic inhibition of POMC neurons by NPY neurons (Cowley et al., *Nature* 411, 480-484, 2001) and these results suggest that PYY_{3-36} acts by inhibiting NPY neurons, thus decreasing this GABAergic tone and consequentially disinhibiting POMC neurons. The effect of Y2A on peptide secretion was also examined using hypothalamic explants (Kim et

al., *J. Clin. Invest.* 105, 1005-11, 2000). Y2A significantly decreased NPY release, with a concomitant increase in α-MSH release from hypothalamic explants (Batterham et al., *Nature* 418:450, 2002). Taken together these observations show that PYY_{3-36} modulates both the NPY and melanocortin systems in the arcuate nucleus.

Example 6

Human Studies

The effect of PYY on feeding in humans has been established (Batterham et al., *Nature* 418:450, 2002).

Example 7

Generation of Additional Lines of Transgenic Mice

A strain of transgenic mice has been generated that expresses green fluorescent protein under the transcriptional control of the mouse POMC genomic sequences, including a region located between –13 kilobases (kb) and –2 kb that is required for accurate neuronal expression (see above, e.g. Example 2, and Cowley et al., Nature 411:480, 2001, which is herein incorporated by reference). Additional lines of transgenic mice were then generated. The starting material for these experiments was either a 4 kb fragment of the rat POMC gene extending from a position approximately -4000 base pairs (bp) 5' to the transcriptional start site through to position +64 in the first exon or a 10 kb mouse genomic clone including approximately 2 kb of 5' flanking sequences. The complete POMC gene is composed of 3 exons and 2 introns, and approximately 2 kb of 3' flanking sequences (see Rubinstein et al., Neuroendocrinol. 58:373, 1993, herein incorporated by reference).

A cosmid genomic library was constructed from 129S6 strain mouse genomic DNA partially cut with the EcoRI restriction endonuclease. This library was screened with probes generated from the original 10 kb mouse POMC clone. This screen resulted in the isolation of several overlapping POMC genomic clones, which were used to construct a transgene vector for microinjection that included approximately 13 kb of 5' flanking sequences, the 6 kb POMC gene, and 8 kb of 3' flanking sequences for a total size of 27 kb. An artificial oligonucleotide sequence

was introduced into exon 3 of the coding sequence to permit the unequivocal identification and quantification of mRNA transcribed from the transgene compared to the endogenous mouse POMC gene. Two additional transgenes were constructed, one that was truncated at the -2 kb side 5' to the POMC gene and the other that was truncated at the +2 kb side 3' to the POMC gene (see Young et al., J. Neurosci. 18:6631, 1998, herein incorporated by reference). Expression studies in this line of transgenic mice demonstrated that DNA sequences between -13 and -2 kb 5' to the POMC gene are necessary for eutopic, neuron-specific expression in the arcuate nucleus of the hypothalamus and the nucleus of the tractus solitarius. However, all the transgenes were appropriately expressed in the pituitary gland, consistent with the location of pituitary-specific DNA regulatory elements within the proximal -400 bp of the POMC promoter.

An additional series of transgenes containing the EGFP reporter gene were constructed as illustrated in Fig. 7. Truncation of the 5' flanking sequences to position -9 kb resulted in a loss of neuronal expression, but did not affect pituitary expression, suggesting that the essential neuron-specific regulatory elements are contained in the 4 kb between nucleotide positions -13 and -9 kb. Furthermore, an internal deletion of genomic 5' flanking genomic sequences between positions -6.5 to -0.8 did not affect the positive transgene expression in either hypothalamic neurons (see Fig. 8a for a representative histologic section illustrating the robust expression of the fluorescent protein in arcuate neurons) or the pituitary cells. This suggests the position independence of the neural regulatory elements relative to the basal promoter. Virtually every transgenic strain produced with the distal 4 kb of regulatory elements displayed a high penetrance of reporter transgene expression in the neurons of the arcuate nucleus, suggesting that the transcriptional regulatory elements contained within the 4 kb of DNA sequence are insulated from the effects of random chromosomal integration position in common with known locus-control type enhancer elements.

A transgene containing the distal 4 kb of mouse POMC 5' genomic sequences between -13 and -9 kb to a minimal herpes simplex viral thymidine kinase (TK) promoter and human growth hormone reporter transgene (Figs. 8b and 8c) was produced, and transengic mice carrying this construct were generated. This region

of the mouse POMC genomic sequences conferred hypothalamic arcuate neuronal-specific expression of the human growth hormone marker.

The minimal TK promoter has been used previously in conjunction with proximal pituitary-specific POMC regulatory elements (see Liu, et al 1992). In these studies, no intrinsic capacity of the TK promoter to cause reporter gene expression in POMC cells of transgenic mice was observed. Thus, expression of the hGH marker in these transgenic mice indicate that the 4 kb of distal mouse POMC genomic sequences contain a classically defined position- and promoter-independent transcriptional enhancer with specific activity for targeting high-level gene expression to POMC neurons, but not pituitary cells, in vivo.

The complete nucleotide sequence of the 4 kb of 129S6 substrain mouse POMC genomic DNA was obtained from the cosmid clones. A multiple alignment sequence comparison was performed with a public human data base BAC sequence containing the human POMC gene on chromosome 2 using the web-based program PIP Maker (Percentage Identity Plot) and the program named Dotter. The two programs, which use a completely different sequence comparison algorithm, found the same two highly conserved regions between mouse and human 5' flanking POMC gene sequences (Fig. 9). These two homology islands have been termed nPOMC1 and nPOMC2 for neural POMC regulatory elements 1 and 2.

Fig. 10 illustrates a sequence alignment of the nPOMC1 and nPOMC2 elements from a variety of mammalian species. In addition to the primary sequence, the distance between both sites to the TATA box is also conserved in the human and mouse POMC genes. The nPOMC1 site extends for approximately 450 bp with an overall mouse/human similarity of 65% having a 190 bp core with a maximal conservation of 80% (the human and bovine similarity is even higher, at 85%). This core is at -10.5 and -12.2 kb from the human and mouse TATA box, respectively. The core is located 1.7 or 2.3 kb upstream of human and mouse nPOMC2, respectively. The site nPOMC2 has a 153 bp region from which 138 are identical between mouse and human (90% of similarity). It is located at -8.9 and -9.9 kb from the human and mouse TATA box, respectively.

Without being bound by theory, these two small and highly conserved areas, embedded within several kb of heterogeneous genomic sequences, resemble the exon-intron differences within the transcriptional unit. Interestingly, the similarities between mouse and human exon 1, 2 and 3 of 64%, 87% and 82%, respectively, are not higher than the interspecies identity for nPOMC1 and nPOMC2 (Fig. 9).

A Clustall comparison of 280 bp of the proximal human and mouse POMC promoters show 68% of similarity in a region that contains all cis-acting elements necessary for basal (e.g. TATA and GC boxes) and pituitary specific expression (e.g. T-Pit, Ptx1 and PP1). Using degenerate oligonucleotide primers to amplify corresponding genomic regions from other mammalian species and sequencing of the PCR fragments, it was confirmed that nPOMC1 and nPOMC2 are also highly conserved in bovine, hamster, and rabbit genomic DNA. In addition, a bovine genomic library was screened using the bovine nPOMC1 PCR fragment as a radiolabeled probe. One of the positive phage clones also contains nPOMC2 and POMC exon 1 indicating that these two regulatory regions are syntenic with the TATA box within 15 kb, similar to the human and mouse. An internal portion of bovine nPOMC2 that was amplified from this clone shows 90% similarity with the human. The sequence of the rat nPOMC1 was obtained from a BLAST comparison of the draft genome project and is highly similar to the mouse sequence. Furthermore, the 129 mouse POMC genomic sequence is nearly identical to the C57BL/6J POMC genomic sequence now available on the public genomic data bases. The nucleotide sequences spanning the nPOMC1 and nPOMC2 elements together with the precise nucleotide positions on human chromosome 2 and mouse chromosome 12 are shown in Fig. 11. BLAST analyses indicate that the complete nPOMC1 and nPOMC2 elements are previously unidentified and uncharacterized sequences and appear to be unique to the POMC gene locus. Thus, one of skill in the art can readily generate transgenic mice carrying a transgene including any of these regions of the POMC gene, operably linked to a marker (such as, but not limited to, growth hormone or GFP).

The adipostatic hormone leptin not only activates POMC neurons but also stimulates transcription of the POMC gene in the hypothalamus presumably through a JAK kinase/STAT3-dependent pathway. The web-based program Mat Inspector

was used to localize STAT3 DNA binding sites within 11.5 kb of 5' flanking regions of the human POMC gene. Eight sites were detected that share high homology with the consensus core TTCCNGGAA. Interestingly, only one site entirely matches this consensus sequence and it is located within the highly conserved nPOMC1 site and 50 bp downstream of another STAT3-like site (Fig. 10). This similarity suggests that nPOMC1 may be a leptin-responsive element within the POMC gene. The sites are slightly less well conserved in the other genomic sequences available. Another potentially interesting short DNA sequence present in the 5' half of nPOMC1 that is 100% identical among all five mammalian species is CTAATGGATGTGCATTA (SEQ ID NO: 352). Excluding the 5' C, the remaining 16 nucleotides contain an imperfect (12/16) palindrome that could be a symmetrical DNA-protein binding site.

As disclosed herein, a number of lines of transgenic mice have been produced that carry a number of transgenes that including a POMC regulatory region operably linked to nucleic acid sequence encoding a marker. Histological sections can readily be prepared from these, or other lines of transgenic mice carrying a POMC regulatory region operably linked to a nucleic acid sequence encoding a marker. These sections can be used to screen agents, such as chemical compounds, proteins, small molecules or salts, in order to identify an agent that affects caloric intake, food intake, or appetite, as described herein.

Example 8

Exemplary Screening Protocol

Coronal slices, from about 140 to about 400 μm thick, containing neurons form the ARC of mice carrying a POMC gene or regulatory element operably linked to a maker, wherein the marker is expressed in the POMC neurons in the ARC. In one embodiment, suitable sections are produced from a male, four week old mouse expressing GFP from the POMC promoter, such as one of the lines of mice disclosed herein. It should be noted that the age and sex of the animal is not a limitation, as female mice as well as older and younger mice can be used. These sections are then incubated with an agent of interest, and an electrophysiological parameter of the POMC neurons is measured. A change in this electrophysiological

parameter indicates that the agent affects caloric intake, food intake, appetite and/or energy expenditure.

In one example, 180 μm thick slices of the ARC are maintained in (in mM) [NaCl, 126; KCl, 2.5; $MgCl_2$, 1.2; $CaCl2_2H2O$, 2.4; $NaH_2PO_4.H2O$, 1.2; $NaHCO_3$, 21.4; Glucose, 11.1] (Krebs) at 35°C and saturated with 95% O_2 5% CO_2 for 1 hour (hr) prior to recordings. Recordings are made in Krebs at 35°C. Slices are visualized on an Axioskop FS2 (Zeiss) through standard infra red optics and using epifluorescence through a FITC filter set (see Fig. 1c). Whole cell or loose cell attached recordings are made from fluorescent neurons using an Axopatch 1D amplifier (Axon Instruments) and Clampex 7 (Axon Instruments).

Resting membrane potentials and action potential frequencies are determined using an event detection protocol on a PowerLab system (AD Instruments, Mountain View, CA) to average expanded traces of the membrane potential. Alternatively, an event detection software package, such as Synaptosoft (Gaegin software), is used to plot instantaneous frequencies. Agents are applied to the bath over a specific time period, such as but not limited to, about one to about 15 minutes. The resting membrane potential is stable for up to an hour in cells treated with Krebs alone.

Current to voltage (I-V) relationships are established using a step protocol; (-60 mV holding potential, sequentially pulsed (40 ms) from -120 to -50 mV, cells were returned to -60 mV for 2 s between voltage steps). The protocol is repeated after addition of an agent of interest. The net current is the difference between the two I-V relationships are used to confirm that the agent is exerting a postsynaptic effect. Similarly slow voltage ramps (5 mV/ s from -100 to -20 mV) before and 10 minutes after the addition of the agent (such as, but not limited to, a concentration of 1nM –10 mM, e.g. at 100 nM) can be used to determine if a postsynaptic effect is occurring.

GABAergic IPSCs are recorded using a CsCl internal electrode solution (in mM) [CsCl, 140; Hepes, 10; $MgCl_2$, 5; Bapta, 1; (Mg)-ATP, 5; (Na)GTP, 0.3]. Addition of blockers of excitatory currents are used to allow the analysis of IPSC frequency in isolation. Both mini IPSCs and large amplitude (presumably multisynaptic) IPSCs are observed in the untreated slices. TTX (1 mM) abolishes

large IPSCs. Data is acquired before and after addition of agent at, for example, a -50 mV holding potential in 2 seconds sweeps every 4 seconds. Mini postsynaptic currents are analyzed using Axograph 4 (Axon Instruments) or Synaptosoft. IPSCs and excitatory postsynaptic currents (EPSCs) are distinguished on the basis of their decay constants and sensitivity to specific blocking agents; picrotoxin (100 mM) blocks IPSCs.

Exemplary parameters that can be analyzed are:

1. Analyzing membrane potential in POMC neurons as compared to a control, such as a POMC neuron in an untreated section or a section incubated with a control agent. This allows assessment of whether an agent increases or decreases the activity of POMC neurons.

2. Analyzing action potential firing rate in POMC neurons as compared to a control, such as a POMC neuron in an untreated section or a section incubated with a control agent. This allows assessment of whether an agent increases or decreases the activity of POMC neurons.

3. Analyzing IPSC frequency onto POMC neurons as compared to a control, such as an untreated section or a section incubated with a control agent. This allows assessment of whether an agent increases or decreases the activity of NPY neurons.

Any change in one or more of these parameters identifies the agent as affecting caloric intake, appetite, food intake, and/or energy expenditure when a therapeutically effective amount of the agent is administered to a subject. Thus, in one embodiment, these data are interpreted by determining the effect of the agent on the activity of POMC neurons (as shown by membrane potential or action potential firing rate) and/or NPY/AGRP neurons (as shown by the IPSC frequency in POMC neurons). Agents that increase the activity of NPY neurons and /or decrease the activity of POMC neurons will increase caloric intake, food intake and/or appetite, and decrease energy expenditure. Agents which decrease the activity of NPY neurons and/or increase the activity of POMC neurons will decrease caloric intake, food intake and/or appetite and/or increase energy expenditure.

Example 9

In Vitro Assessment of Ghrelin, A Known Appetite Stimulant

Slices are of the ARC from POMC-EGFP mice (see Example 1 for a description of the animals) were maintained in (in mM) [NaCl, 126; KCl, 2.5; $MgCl_2$, 1.2; $CaCl_2.2H_2O$, 2.4; $NaH_2PO_4.H_2O$, 1.2; $NaHCO_3$, 21.4; Glucose, 11.1] (Krebs) at 35°C and saturated with 95% O_2 5% CO_2 for 1 hour (hr) prior to recordings. Recordings were made in Krebs at 35°C. Slices were visualized on an Axioskop FS2 (Zeiss) through standard infra red optics and using epifluorescence through a FITC filter set. Whole cell (Fig. 12a and 12b) or loose cell attached (Fig. 12c) recordings were made from fluorescent neurons using an Axopatch 1D amplifier (Axon Instruments) and Clampex 7 (Axon Instruments).

GABAergic IPSCs were recorded using a CsCl internal electrode solution (in mM) [CsCl, 140; Hepes, 10; $MgCl_2$, 5; Bapta, 1; (Mg)-ATP, 5; (Na)GTP, 0.3]. Addition of blockers of excitatory currents allowed the analysis of IPSC frequency in isolation. Both mini IPSCs and large amplitude (presumably multisynaptic) IPSCs were observed in the untreated slices. TTX (1 mM) abolished large IPSCs. Data was acquired before and after addition of agent at a -50 mV holding potential in 2 second sweeps every 4 seconds. Picrotoxin (100 mM) blocked all IPSCs.

IPSC frequencies were analyzed using Synaptosoft (Gaegin software), to determine instantaneous IPSC frequencies (Fig. 12a). Resting membrane potentials are determined using an event detection protocol on a PowerLab system (AD Instruments, Mountain View, CA) to average expanded traces of the membrane potential (Fig. 12b). Action potential firing rates were determined in loose cell attached mode and the recorded data was analyzed using synaptosoft to determine instantaneous frequencies. Ghrelin (50 nM) was applied to the bath over three minutes. The resting membrane potential was stable for up to an hour in cells treated with Krebs alone.

The results of the addition of ghrelin on IPSC frequency of POMC neurons are shown in Fig. 12a. An increase in the frequency of IPSCs in POMC neurons was detected. This is caused by NPY neurons. Thus an increase in the activity of NPY neurons is demonstrated. Fig. 12b shows the effect of ghrelin on the resting membrane potential of POMC neurons. Gherlin hyperpolarized POMC neurons,

mean 1.47 ± 0.7mV; $p<0.03$. The activity of POMC neurons was decreased by the addition of ghrelin. Fig 12c shows the action potential firing rate in POMC neurons. The activity of POMC neurons was inhibited by ghrelin. Thus, ghrelin, an agent known to increase feeding, caloric intake, and appetite, and decrease energy expenditure, decreases the resting membrane potential (Fig. 12b) and action potential firing rate of POMC neurons (by 50%, see Fig. 12c), increases the frequency of IPSCs in POMC neurons (Fig. 12a) and increases the activity of NPY neurons.

Example 10

In-vitro assessment of fenfluramine, a known appetite suppressant and weight loss agent

Slices are of the ARC from POMC-EGFP expressing transgenic mice (Batterham et al., *Nature* 418:450, 2002) were maintained in (in mM) [NaCl, 126; KCl, 2.5; $MgCl_2$, 1.2; $CaCl_2.2H_2O$, 2.4; $NaH_2PO_4.H_2O$, 1.2; $NaHCO_3$, 21.4; Glucose, 11.1] (Krebs) at 35°C and saturated with 95% O_2 5% CO_2 for 1 hour (hr) prior to recordings. Recordings were made in Krebs at 35°C. Slices were visualized on an Axioskop FS2 plus (Zeiss) through standard infra red optics and using epifluorescence through a endow-GFP filter set (Chroma Technology Corp). Whole cell or loose cell attached recordings (both were used, results from whole cell recordings are shown in Fig. 13b; results from loose cell attached are shown in Fig. 13a) were made from fluorescent neurons using an Axopatch 200B amplifier (Axon Instruments) and Clampex 8 (Axon Instruments).

Action potential firing rates were determined in loose cell attached mode and the recorded data was analyzed using synaptosoft to determine instantaneous frequencies (Fig. 13a). Resting membrane potentials and depolarizations were determined using an event detection protocol on a PowerLab system (AD Instruments, Mountain View, CA) to average expanded traces of the membrane potential (Fig. 13b) Serotonin receptor agonists were applied to the bath at the specified concentrations over four minutes. The resting membrane potential was stable for up to an hour in cells treated with Krebs alone.

Fenfluramine (20μM) caused a two fold increase in the frequency of action potentials in POMC neurons (fig 13a; n=3). Thus fenfluramine increased the activity of POMC neurons. In separate experiments Fenfluramine also depolarized POMC neurons (Fig 13b) in a dose dependent manner. Thus, by another test Fenfluramine increases the activity of POMC neurons. The non-selective serotonin receptor agonist serotonin (5-HT) also increased the resting membrane potential (depolarized) POMC neurons. The effect of serotonin and fenfluramine on POMC neurons is likely mediated by the 5-HT 2C R because 5-HT 2C R selective agonists mCPP and MK 212 also depolarized POMC neurons (Fig 13b).

It will be apparent that the precise details of the methods or compositions described may be varied or modified without departing from the spirit of the described invention. We claim all such modifications and variations that fall within the scope and spirit of the claims below.

THE CLAIMS DEFINING THE INVENTION ARE AS FOLLOWS:

1. A method for screening for an agent that affects caloric intake, comprising

contacting a histological section of an arcuate nucleus from a mouse expressing a marker in proopiomelanocortin neurons with an agent to be tested, wherein the mouse comprises a transgene comprising a nucleic acid encoding the marker operably linked to a proopiomelanocortin nucleic acid sequence, and wherein the proopiomelanocortin nucleic acid sequence directs expression of the marker in proopiomelanocortin neurons in the arcuate nucleus of the mouse; and

assaying for an electrophysiological response of a proopiomelanocortin neuron in the histological section, thereby determining if the agent affects caloric intake.

2. A method for screening for an agent that affects appetite, comprising

contacting a histological section of an arcuate nucleus from a mouse expressing a marker in proopiomelanocortin neurons with an agent to be tested, wherein the mouse comprises a transgene comprising a nucleic acid encoding the marker operably linked to a proopiomelanocortin nucleic acid sequence, and wherein the proopiomelanocortin nucleic acid sequence directs expression of the marker in proopiomelanocortin neurons in the arcuate nucleus of the mouse; and

assaying for an electrophysiological response of a proopiomelanocortin neuron in the histological section, thereby determining if the agent affects appetite.

3. A method for screening for an agent that affects food intake, comprising

contacting a histological section of an arcuate nucleus from a mouse expressing a marker in

proopiomelanocortin neurons with an agent to be tested, wherein the mouse comprises a transgene comprising a nucleic acid encoding the marker operably linked to a proopiomelanocortin nucleic acid sequence, and wherein the proopiomelanocortin nucleic acid sequence directs expression of the marker in proopiomelanocortin neurons in the arcuate nucleus of the mouse; and

assaying for an electrophysiological response of a proopiomelanocortin neuron in the histological section, thereby determining if the agent affects food intake.

4. A method according to any one of claims 1 to 3, wherein the agent specifically binds a receptor on the proopiomelanocortin neuron.

5. A method according to any one of claims 1 to 4, wherein the agent specifically binds a melanocortin receptor or a μ-opioid receptor.

6. A method according to claim 5, wherein the agent specifically binds the melanocortin 3 receptor.

7. A method according to any one of claims 1 to 3, wherein the agent specifically binds a receptor on a neuropeptide Y neuron.

8. A method according to claim 7, wherein the agent specifically binds the Y2 receptor.

9. A method according to any one of claims 1 to 8, wherein the electrophysiological response comprises induction of current or a change in the membrane potential of the proopiomelanocortin neuron.

10. A method according to any one of claims 1 to 9, wherein the electrophysiological response comprises depolarization of the proopiomelanocortin neuron.

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11. A method according to any one of claims 1 to 10, wherein the marker is a fluorescent protein.

12. A method according to claim 11, wherein the fluorescent protein is green fluorescent protein.

13. A method according to any one of claims 1 to 12, wherein the proopiomelanocortin nucleic acid sequence comprises a proopiomelanocortin regulatory nucleic acid sequence.

14. A method according to any one of claims 1 to 13, wherein the agent is a polypeptide, a chemical compound, a salt, or a neurotransmitter.

15. A method for screening for an agent that affects caloric intake, energy expenditure, appetite, or food intake, comprising

contacting a histological section of an arcuate nucleus, with an agent to be tested, wherein proopiomelanocortin neurons in the histological section express a heterologous marker that distinguishes the proopiomelanocortin neurons from other cells in the histological section; and

assaying for an electrophysiological response of a proopiomelanocortin neuron in the histological section, thereby determining if the agent affects caloric intake, appetite, energy expenditure, or food intake.

16. A method according to claim 15, wherein the heterologous marker is expressed from a transgene.

17. A method according to claim 15 or claim 16, wherein the agent specifically binds a receptor on the proopiomelanocortin neuron.

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18. A method according to any one of claims 15 to 17, wherein the agent specifically binds a melanocortin receptor or μ-opioid receptor.

19. A method according to claim 18, wherein the agent specifically binds the melanocortin 3 receptor.

20. A method according to claim 15, wherein the agent specifically binds a receptor on a neuropeptide Y neuron.

21. A method according to claim 20, wherein the agent specifically binds the Y2 receptor.

22. A method according to any one of claims 15 to 21, wherein the electrophysiological response comprises induction of current or a change in the membrane potential of the proopiomelanocortin neuron.

23. A method according to any one of claims 15 to 22, wherein the electrophysiological response comprises depolarization of the proopiomelanocortin neuron.

24. A method according to any one of claims 15 to 23, wherein the heterologous marker is a fluorescent protein.

25. A method according to claim 24, wherein the fluorescent protein is green fluorescent protein.

26. A method according to any one of claims 15 to 25, wherein the proopiomelanocortin nucleic acid sequence comprises a proopiomelanocortin regulatory nucleic acid sequence and a nucleic acid sequence encoding proopiomelanocortin.

27. A method according to any one of claims 1 to 26, wherein assaying an electrophysiological response comprises: (a) a whole cell recording; (b) a loose cell

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attached patch recording; (c) determining a resting membrane potential of a proopiomelanocortin neuron; or (d) determining a GABA-mediated inhibitory postsynaptic current or determining the action potential frequency of a proopiomelanocortin neuron.

28. A method according to any one of claims 1 to 27, wherein the agent is not PYY and is not a polypeptide fragment of PYY that is more than 10 amino acids in length.

29. A method according to any one of claims 1 to 13 and 15 to 26, wherein the agent is not a polypeptide.

30. A method according to any one of claims 1 to 3 or 15, substantially as herein described with reference to any one of the examples and/or drawings.

FIG. 1

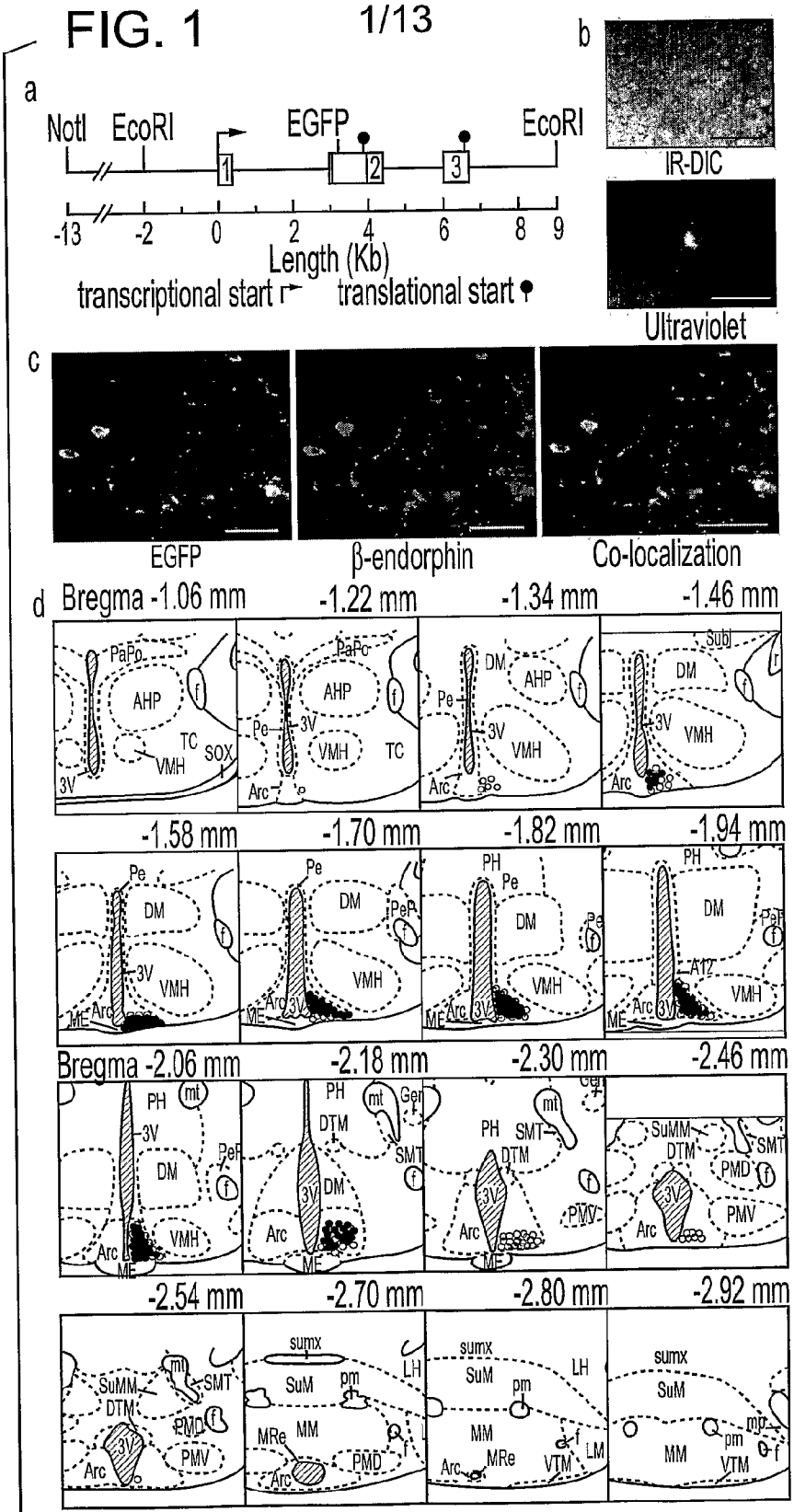

a
NotI
EcoRI
EGFP
EcoRI
1
2
3
-13
-2
0
2
4
6
8
9
Length (Kb)
transcriptional start
translational start
b
IR-DIC
Ultraviolet
c
EGFP
β-endorphin
Co-localization
d
Bregma -1.06 mm
-1.22 mm
-1.34 mm
-1.46 mm
-1.58 mm
-1.70 mm
-1.82 mm
-1.94 mm
Bregma -2.06 mm
-2.18 mm
-2.30 mm
-2.46 mm
-2.54 mm
-2.70 mm
-2.80 mm
-2.92 mm

FIG. 2

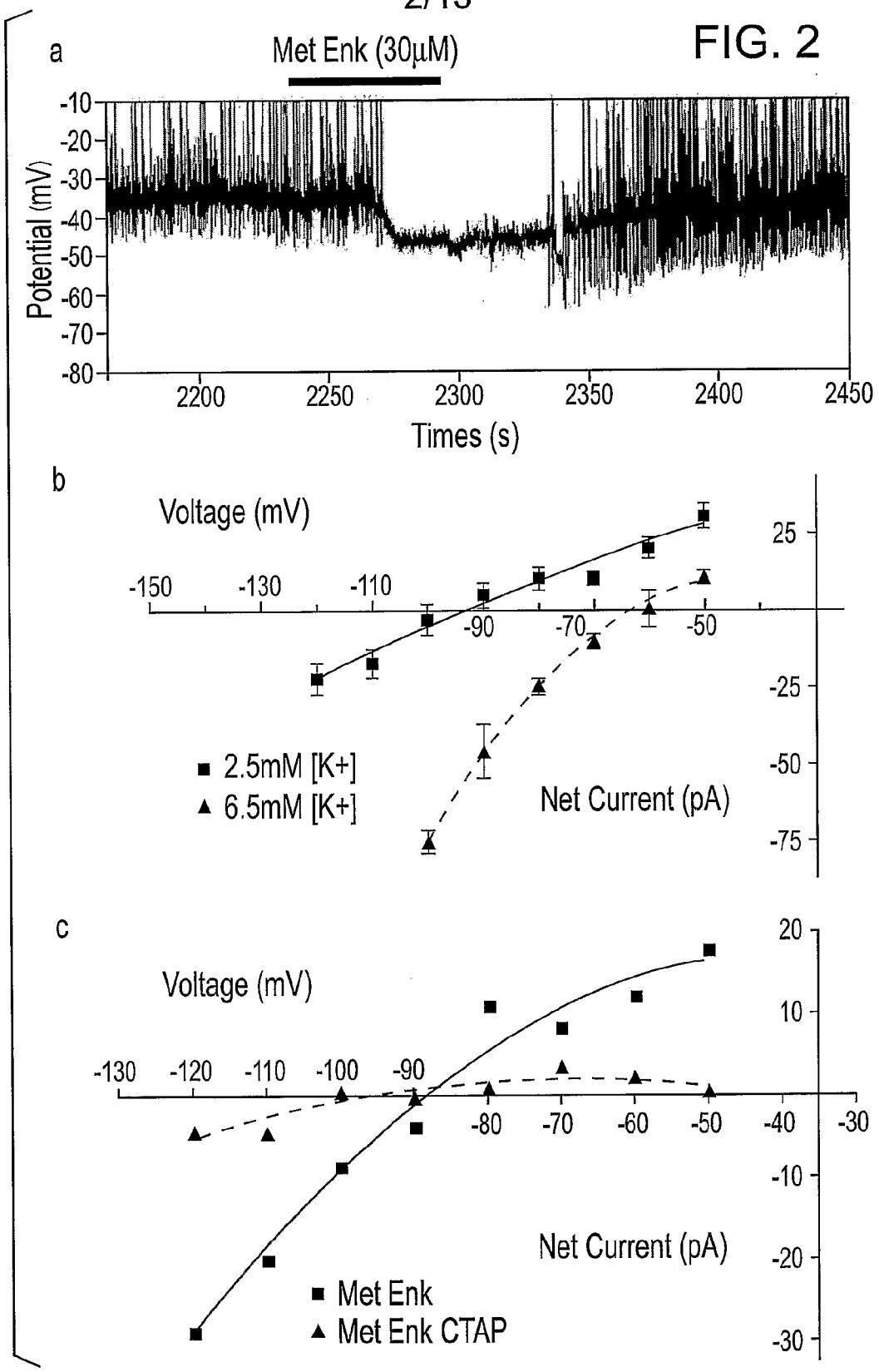

a
Met Enk (30μM)
Potential (mV)
-10
-20
-30
-40
-50
-60
-70
-80
2200
2250
2300
2350
2400
2450
Times (s)
b
Voltage (mV)
-150
-130
-110
-90
-70
-50
25
-25
-50
-75
2.5mM [K+]
6.5mM [K+]
Net Current (pA)
c
Voltage (mV)
-130
-120
-110
-100
-90
-80
-70
-60
-50
-40
-30
20
10
-10
-20
-30
Net Current (pA)
Met Enk
Met Enk CTAP

FIG. 3

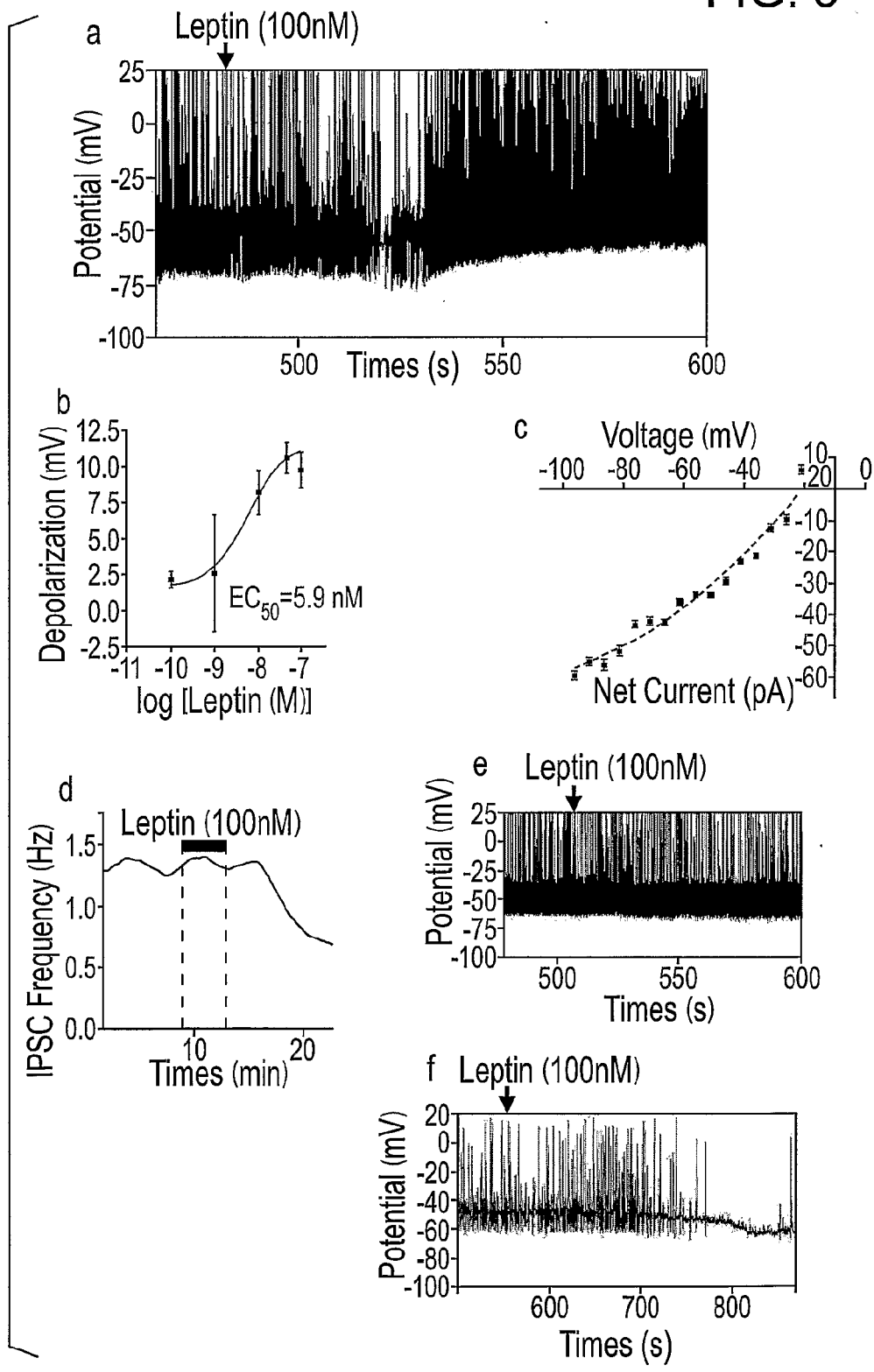
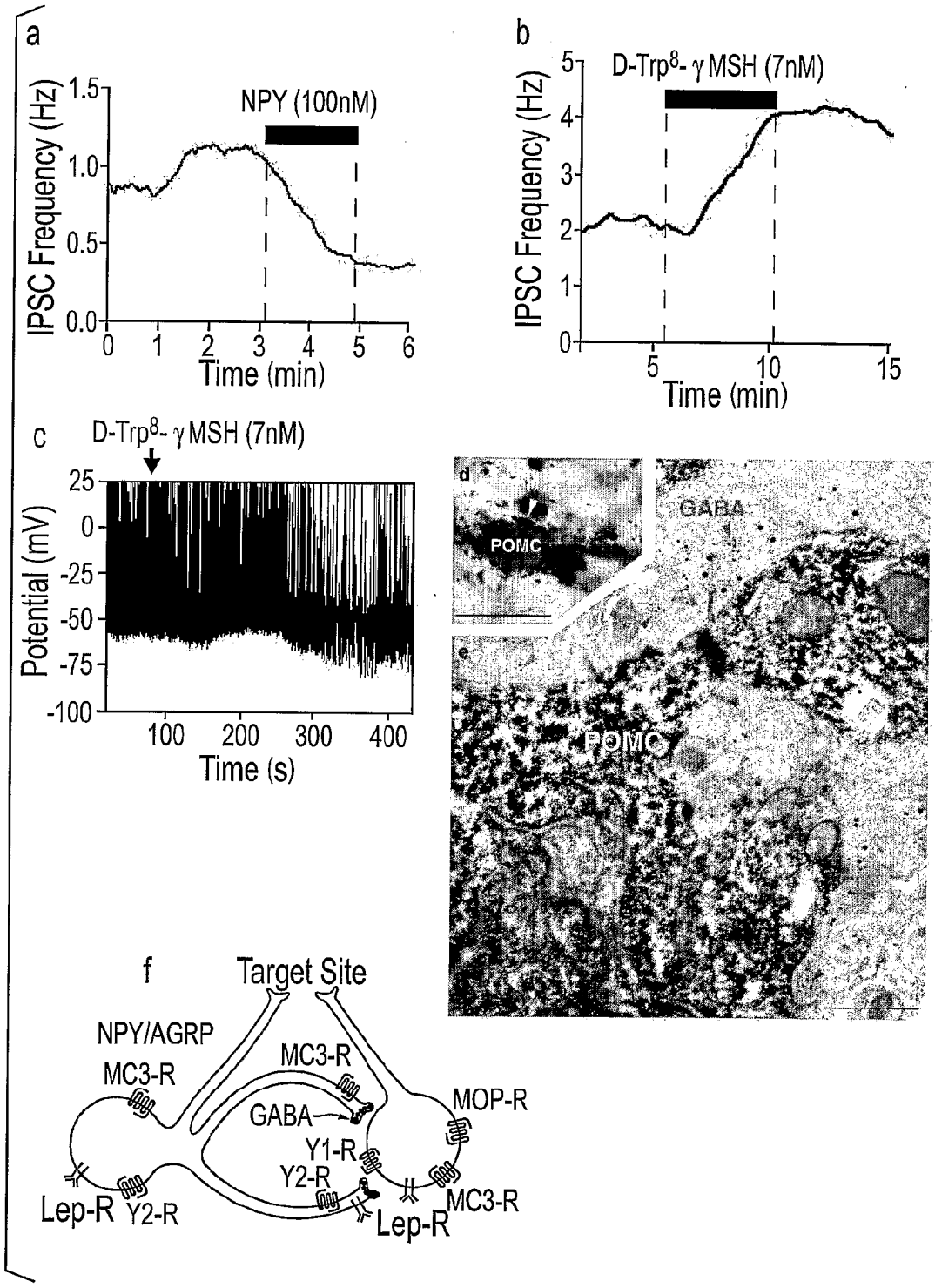

a
Leptin (100nM)
Potential (mV)
25
0
-25
-50
-75
-100
500
Times (s)
550
600
b
Depolarization (mV)
12.5
10.0
7.5
5.0
2.5
0.0
-2.5
EC50=5.9 nM
-11 -10 -9 -8 -7
log [Leptin (M)]
c
Voltage (mV)
-100 -80 -60 -40 -20 0
10
-10
-20
-30
-40
-50
-60
Net Current (pA)
d
Leptin (100nM)
IPSC Frequency (Hz)
1.5
1.0
0.5
0.0
10
20
Times (min)
e
Leptin (100nM)
Potential (mV)
25
0
-25
-50
-75
-100
500
550
600
Times (s)
f
Leptin (100nM)
Potential (mV)
20
0
-20
-40
-60
-80
-100
600
700
800
Times (s)

FIG. 4


a
NPY (100nM)
IPSC Frequency (Hz)
Time (min)
b
D-Trp8- γ MSH (7nM)
IPSC Frequency (Hz)
Time (min)
c
D-Trp8- γ MSH (7nM)
Potential (mV)
Time (s)
d
POMC
GABA
e
POMC
f
Target Site
NPY/AGRP
MC3-R
MC3-R
GABA
MOP-R
Y1-R
Y2-R
Lep-R
Y2-R
Lep-R
MC3-R

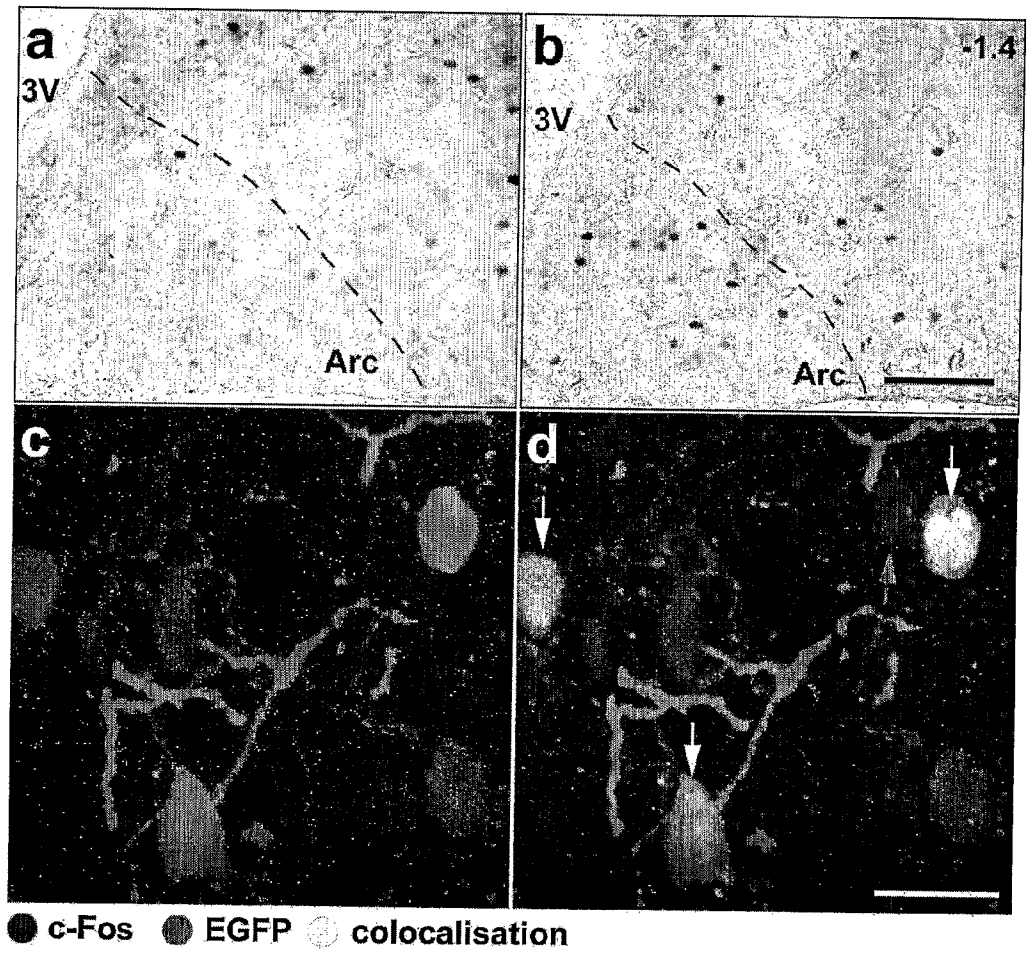
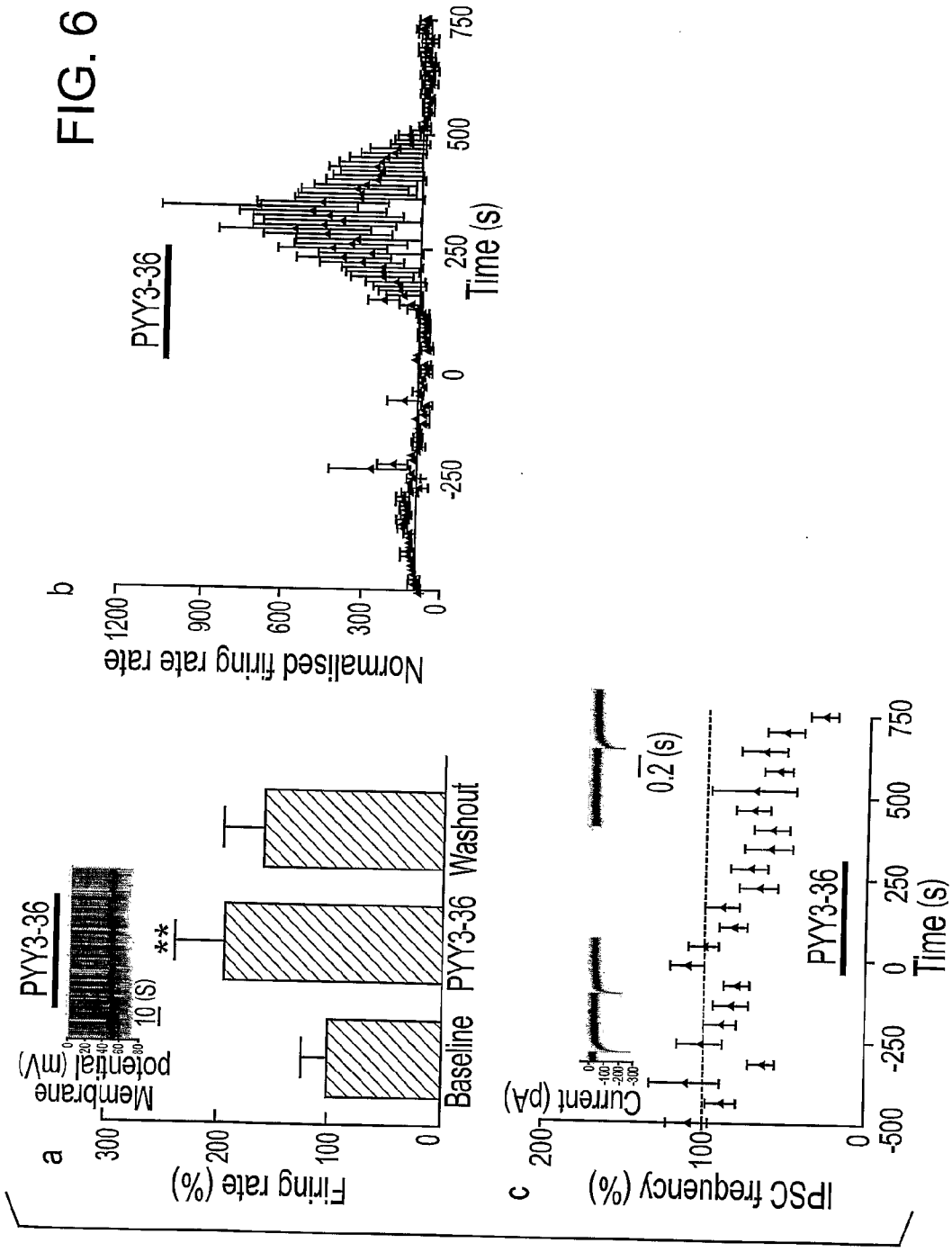

a
3V
Arc
b
-1.4
3V
Arc
c
d
c-Fos
EGFP
colocalisation

FIG. 6


a
PYY3-36
Membrane potential (mV)
10 (S)
Firing rate (%)
Baseline
PYY3-36
Washout
**
b
PYY3-36
Normalised firing rate rate
Time (s)
c
IPSC frequency (%)
Current (pA)
0.2 (s)
PYY3-36
Time (s)

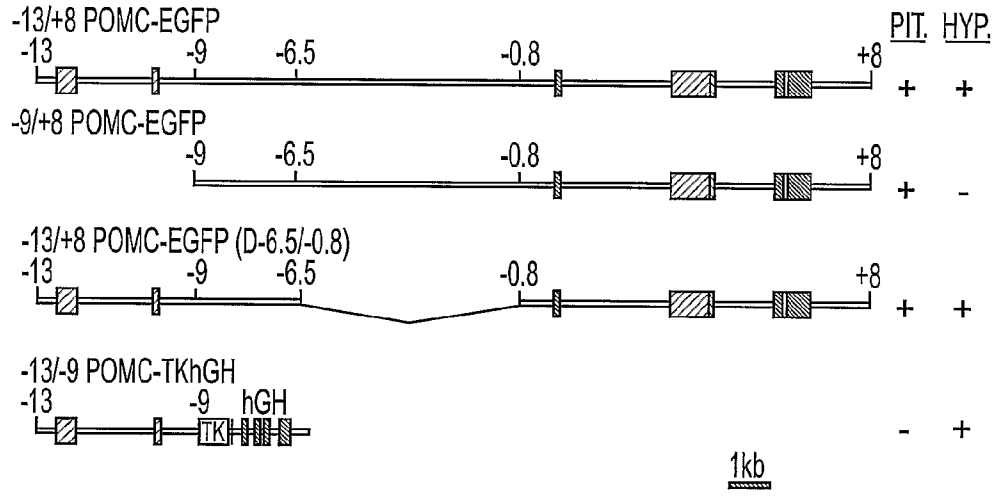

-13/+8 POMC-EGFP
-13
-9
-6.5
-0.8
+8
PIT.
HYP.
+
+
-9/+8 POMC-EGFP
-9
-6.5
-0.8
+8
+
-
-13/+8 POMC-EGFP (D-6.5/-0.8)
-13
-9
-6.5
-0.8
+8
+
+
-13/-9 POMC-TKhGH
-13
-9
TK
hGH
-
+
1kb

FIG. 7

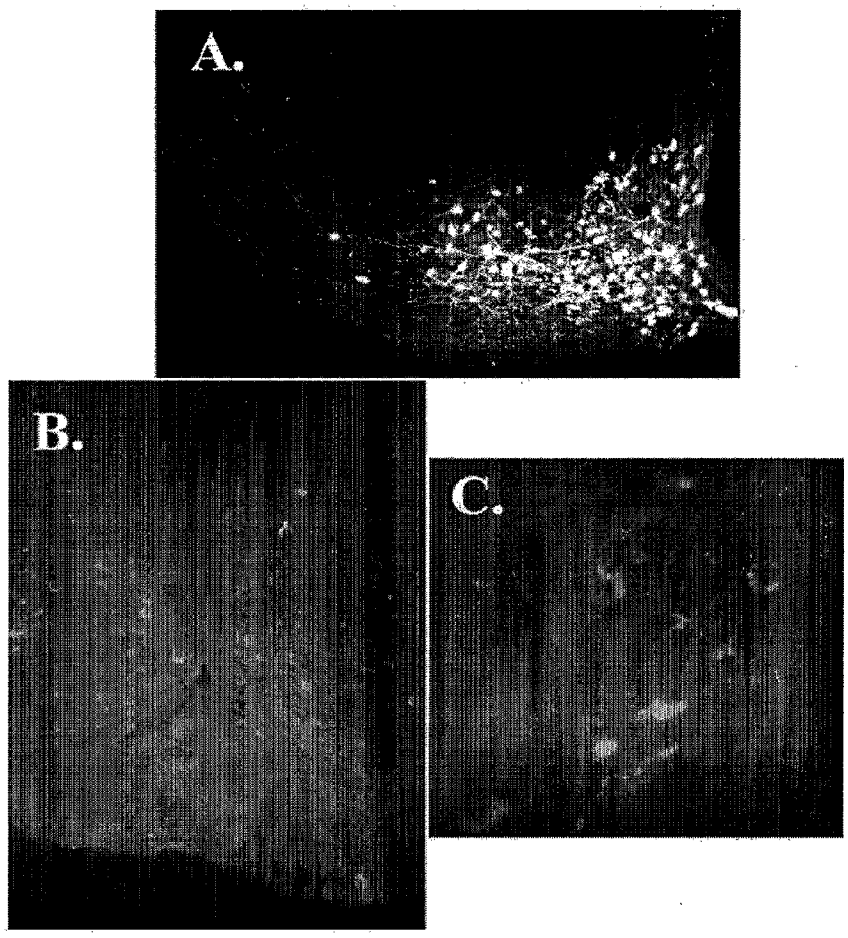


FIG. 8

FIG. 9

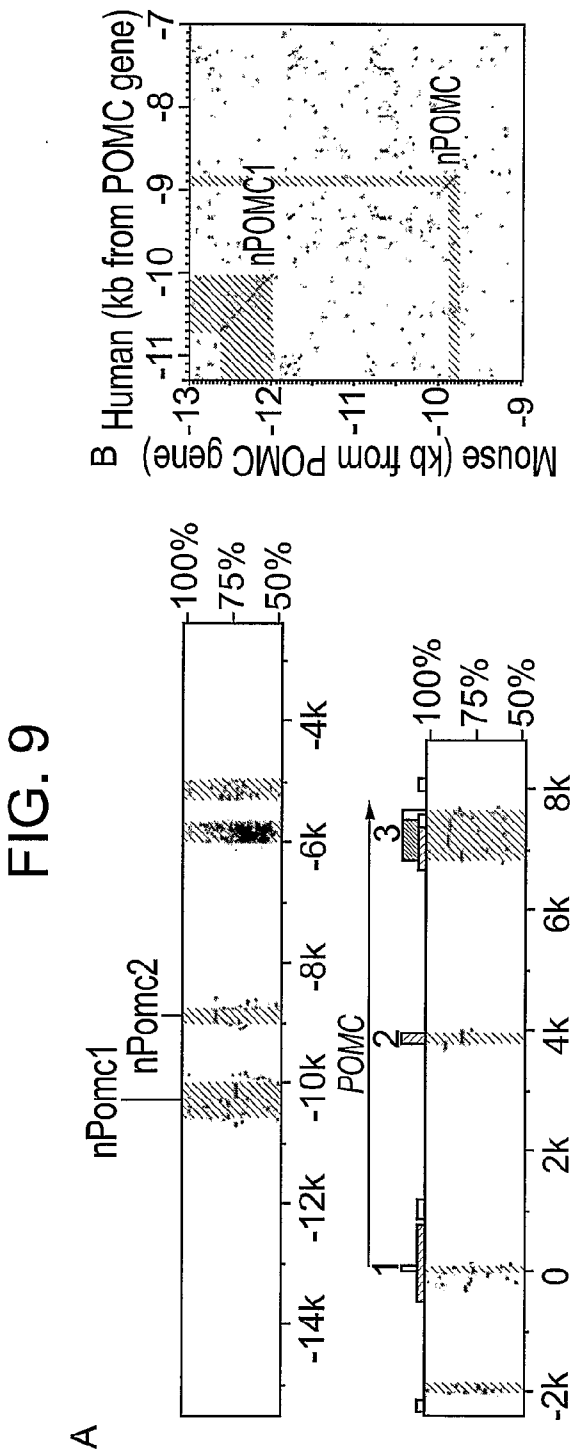
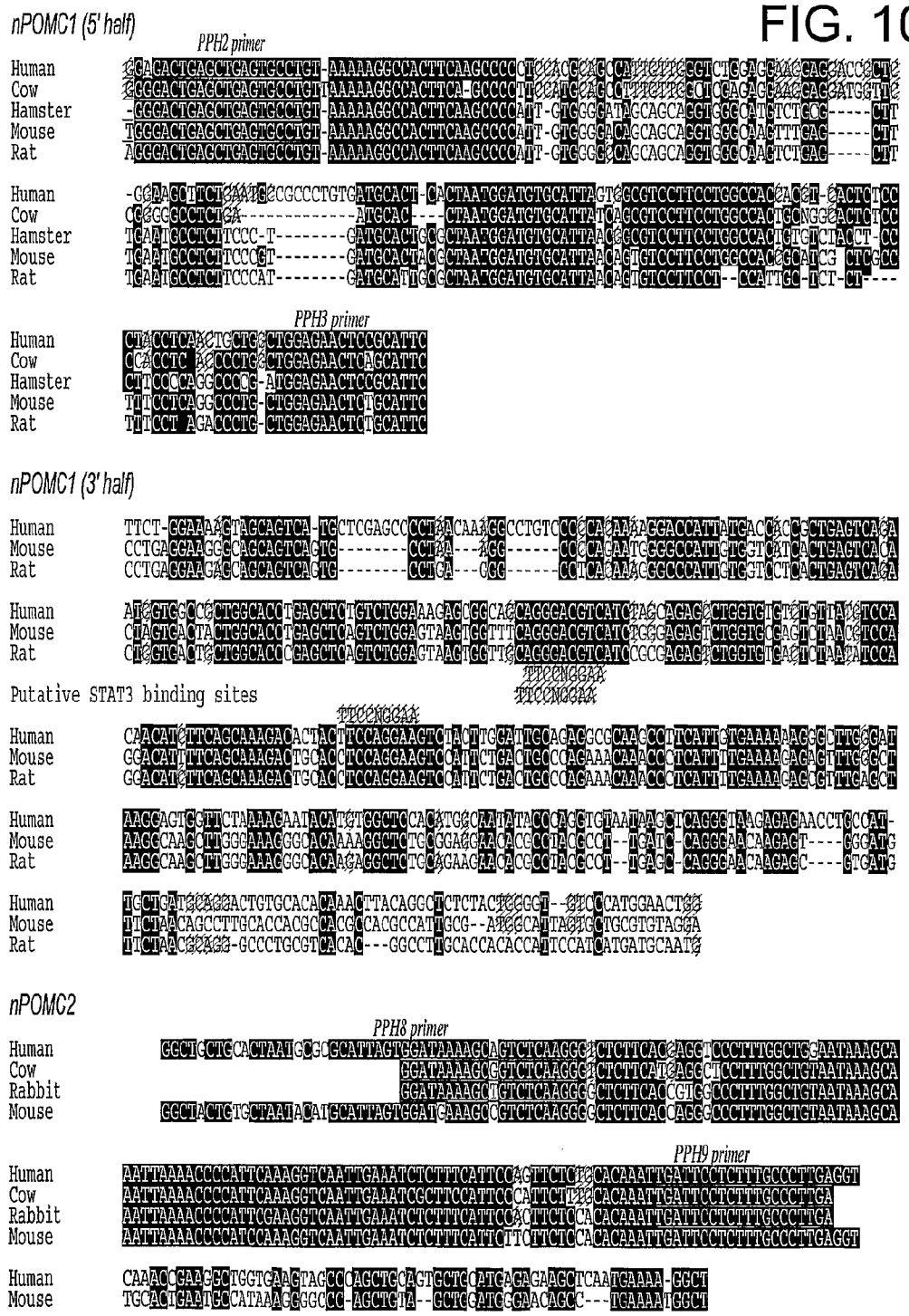

A
nPomc1
nPomc2
100%
75%
50%
-4k
-6k
-8k
-10k
-12k
-14k
POMC
1
2
3
100%
75%
50%
8k
6k
4k
2k
0
-2k
B
Human (kb from POMC gene)
-7
-8
-9
-10
-11
-12
-13
nPOMC1
nPOMC
Mouse (kb from POMC gene)
-9
-10
-11
-12
-13

FIG. 10


nPOMC1 (5' half)
PPH2 primer
Human
Cow
Hamster
Mouse
Rat
PPH3 primer
nPOMC1 (3' half)
Putative STAT3 binding sites
nPOMC2
PPH8 primer
Human
Cow
Rabbit
Mouse
PPH9 primer

"nPOMC 1 element"

FIG. 11

Mouse Chromosome 12 nucleotides 3,808,013 - 3,808,447

GACTGAGCTGAGTGCCTGTAAAAAGGCCACTTCAAGCCCCATTGTGGGGACAGCAGCAGGTGGGCA
AGTCTGAGCTTTGAATGCCTCTTCCCGTGATGCACTACGCTAATGGATGTGCATTAACAGTGTCCT
TCCTGGCCACCGCATCGCTCGCCTTTCCTCAGGCCCTGCTGGAGAACTCTGCATTCCTGAGGAAGG
GCAGCAGTCAGTGCCTAAAGGCCCCAGAATGGGGCCATTGTGGTCATCACTGAGTCACACTAGTGA
CTACTGGCACCTGAGCTCAGTCTGGAGTAAGTGGTTTCAGGGACGTCATCTGGGAGAGTCTGGTGC
GAGTCTAACGTCCAGGACATTTTCAGCAAAGACTGCACCTCCAGGAAGTCCATTCTGACTGCCCAG
AAACAAACCCTCATTTTGAAAAGAGAGTTTGGGCTAAGG

"nPOMC 1 element"

Human Chromosome 2 nucleotides 2,324,416 - 2,323,942 (REVERSE COMPLEMENT STRAND)

GACTGAGCTGAGTGCCTGTAAAAAGGCCACTTCAAGCCCCCTCCACGCAGCCATTGTTGGGTCTGG
AGGAAGGAGGACCGCTCGGAAGCTTCTGAATGCCGCCCTGTGATGCACTCACTAATGGATGTGCAT
TAGTGGCGTCCTTCCTGGCCACCACGTCACTCTCCCTACCTCAACTGCTGGCTGGAGAACTCCGCA
TTCTTCTGGAAAAGTAGCAGTCATGCTCGAGCCCCTAACAAAGGCCTGTCCCCCACAAAAGGACCA
TTATGACCACCGCTGAGTCAGAATGGTGGCCGCTGGCACCTGAGCTCTGTCTGGAAAGAGCGGCAG
CAGGGACGTCATCTAGCAGAGCCTGGTGTGTCTGTTATGTCCACAACATCTTCAGCAAAGACACTA
CTTCCAGGAAGTCTACTTGGATTGCAGAGGCGCAAGCCTTCATTGTGAAAAAAGGGCTTGGGATAA
GG

"nPOMC 2 element"

Mouse Chromosome 12 nucleotides 3,810,489 - 3,810,724

GGCTGGGGTGGGCTACTGTGCTAATACATGCATTAGTGGATGAAAGCCGTCTCAAGGGGCTCTTCA
CCAGGGCCCTTTGGCTGTAATAAAGCAAATTAAAACCCCATCCAAAGGTCAATTGAAATCTCTTTC
ATTCTTCTTCTCCACACAAATTGATTCCTCTTTGCCCTTGAGGTTGCACTGAATGCCATAAAGGGG
CCCAGCTGTAGCTGGATGGGAACAGCCTGAAAATGGCT

"nPOMC 2 element"

Human Chromosome 2 nucleotides 2,322,890 - 2,322,659 (REVERSE COMPLEMENT
STRAND) GGCTGCTGCACTAATGCGCGCATTAGTGGATAAAAGCAGTCTCAAGGGTCTCTTCACGA
GGTCCCTTTGGCTGGAATAAAGCAAATTAAAACCCCATTCAAAGGTCAATTGAAATCTCTTTCATT
CCAGTTCTCTGCACAAATTGATTCCTCTTTGCCCTTGAGGTCAAACCGAAGGCTGGTGAAGTAGCC
CAGCTGCAGTGCTGCATGAGAGAAGCTCAATGAAAAGGCT

FIG. 12

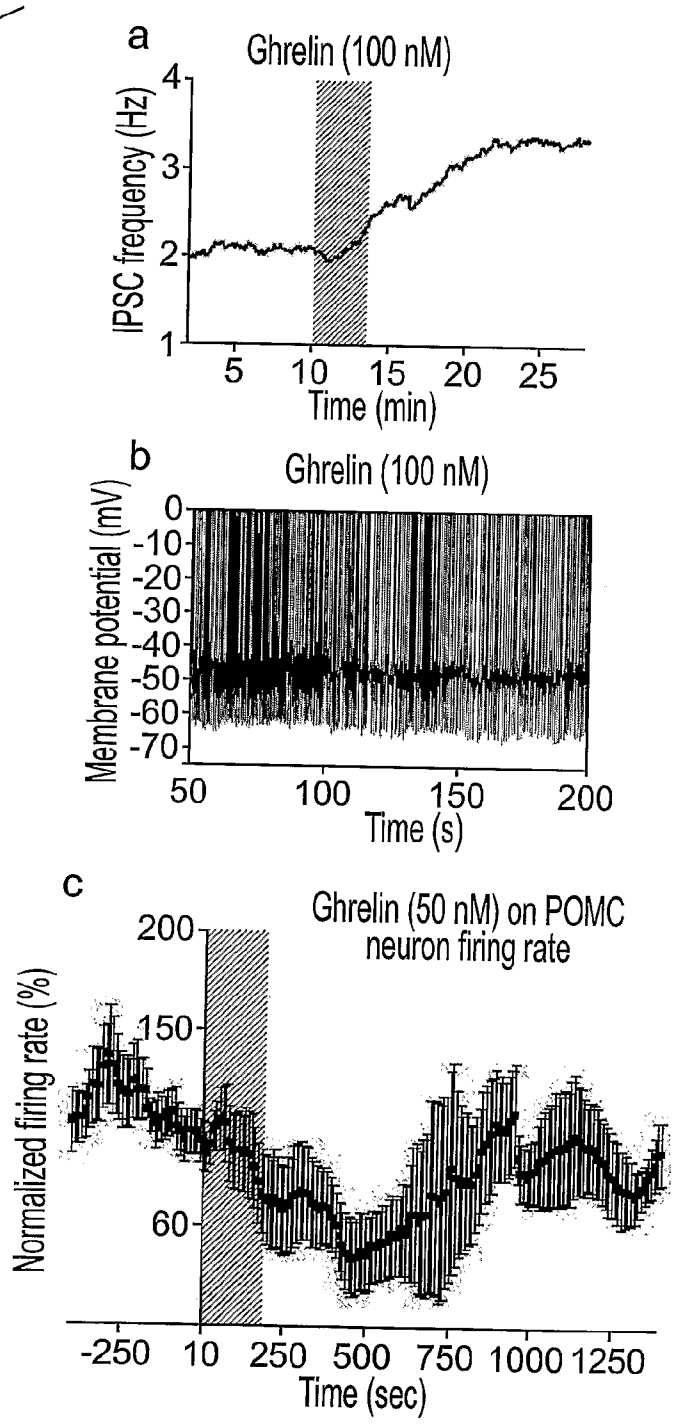
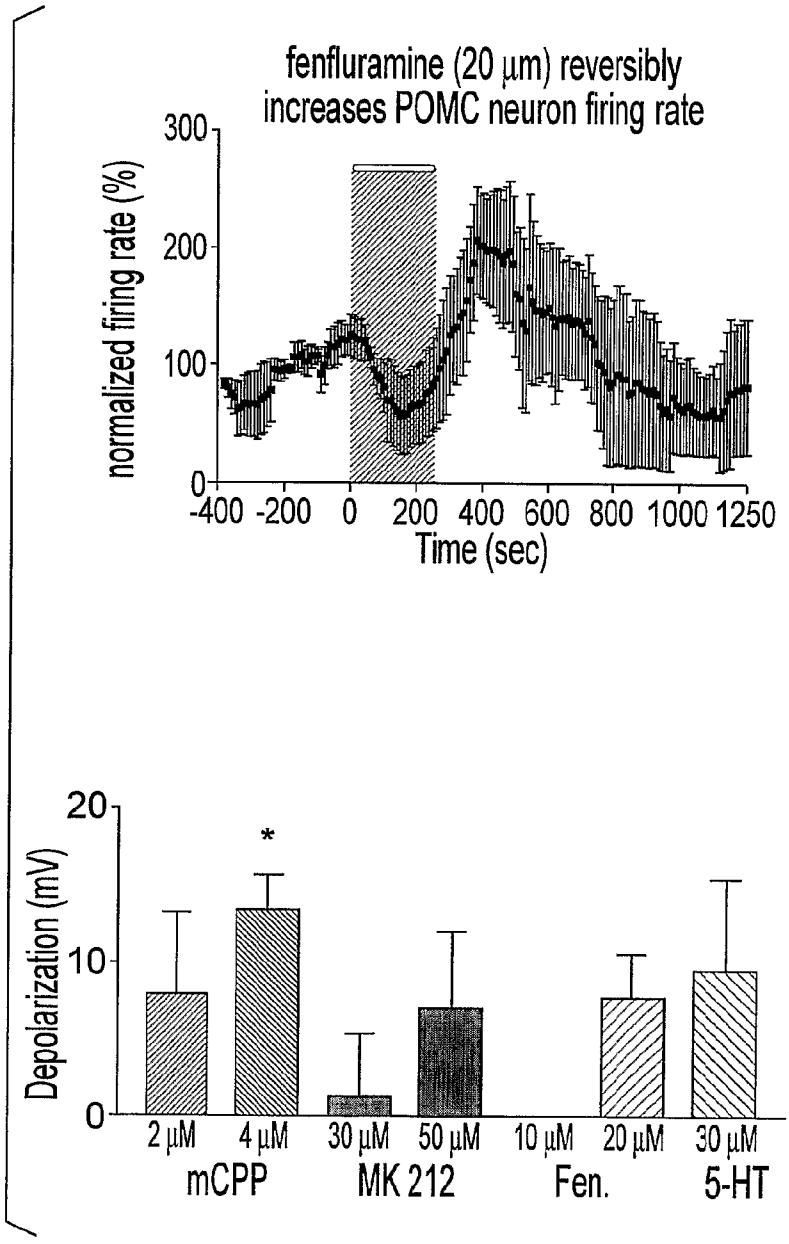

a
Ghrelin (100 nM)
IPSC frequency (Hz)
4
3
2
1
5
10
15
20
25
Time (min)
b
Ghrelin (100 nM)
Membrane potential (mV)
0
-10
-20
-30
-40
-50
-60
-70
50
100
150
200
Time (s)
c
Ghrelin (50 nM) on POMC neuron firing rate
Normalized firing rate (%)
200
150
60
-250
10
250
500
750
1000
1250
Time (sec)

FIG. 13


fenfluramine (20 μm) reversibly
increases POMC neuron firing rate
normalized firing rate (%)
300
200
100
0
-400 -200 0 200 400 600 800 1000 1250
Time (sec)
Depolarization (mV)
20
10
0
*
2 μM 4 μM 30 μM 50 μM 10 μM 20 μM 30 μM
mCPP
MK 212
Fen.
5-HT

SEQUENCE LISTING

<110> Cowley, Michael

Cone, Roger

Low, Malcolm

Butler, Andrew

<120> Assessment of Neurons in the Arcuate Nucleus to Screen for Agents that Modify Feeding Behavior

<130> 899-63986

<140>

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<150> US 60/324,406

<151> 2001-09-24

<150> US 60/392,109

<151> 2002-06-28

<160> 352

<170> PatentIn version 3.1

<210> 1

<211> 36

<212> PRT

<213> Homo sapiens

<400> 1

Tyr Pro Ile Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu
1 5 10 15

Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 2

<211> 36

<212> PRT

<213> Homo sapiens

<400> 2

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 3

<211> 36

<212> PRT

<213> Homo sapiens

<400> 3

Ala Ser Leu Glu Pro Glu Tyr Pro Gly Asp Asn Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Ala Ala Glu Leu Arg Arg Tyr Ile Asn Met Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 4

<211> 3

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<213> Homo sapiens

<400> 4

Ala Ala Ala
1

<210> 5

<211> 36

<212> PRT

<213> Rattus sp.

<400> 5

Tyr Pro Ala Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu
1 5 10 15

Leu Ser Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 6

<211> 36

<212> PRT

<213> Sus sp.

<400> 6

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1 5 10 15

Leu Ser Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 7

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<212> PRT

<213> Cavia porcellus

<400> 7

Tyr Pro Ser Lys Pro Glu Ala Pro Gly Ser Asp Ala Ser Pro Glu Glu
1 5 10 15

Leu Ala Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 8

<211> 36

<212> PRT

<213> Rana sp.

<400> 8

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1 5 10 15

Met Thr Lys Tyr Leu Thr Ala Leu Arg His Tyr Ile Asn Leu Val Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 9

<211> 36

<212> PRT

<213> Raja sp.

<400> 9

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Tyr Pro Pro Lys Pro Glu Asn Pro Gly Asp Asp Ala Ala Pro Glu Glu
1               5                   10                  15

Leu Ala Lys Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
            20                  25                  30

Arg Gln Arg Tyr
        35
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<210> 10

<211> 36

<212> PRT

<213> Dogfish sp.

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Tyr Pro Pro Lys Pro Glu Asn Pro Gly Glu Asp Ala Pro Pro Glu Glu
1               5                   10                  15

Leu Ala Lys Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
            20                  25                  30

Arg Gln Arg Tyr
        35
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<210> 11

<211> 36

<212> PRT

<213> Lampetra sp.

<400> 11

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Phe Pro Pro Lys Pro Asp Asn Pro Gly Asp Asn Ala Ser Pro Glu Gln
1               5                   10                  15

Met Ala Arg Tyr Lys Ala Ala Val Arg His Tyr Ile Asn Leu Ile Thr
            20                  25                  30

Arg Gln Arg Tyr
        35
```

<210> 12

<211> 36

<212> PRT

<213> Petromyzontidae gen. sp.

<400> 12

Met Pro Pro Lys Pro Asp Asn Pro Ser Pro Asp Ala Ser Pro Glu Glu
1 5 10 15

Leu Ser Lys Tyr Met Leu Ala Val Arg Asn Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 13

<211> 36

<212> PRT

<213> Rattus sp.

<400> 13

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 14

<211> 36

<212> PRT

<213> Oryctolagus cuniculus

<400> 14

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 15

<211> 36

<212> PRT

<213> Canis familiaris

<400> 15

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 16

<211> 36

<212> PRT

<213> Sus sp.

<400> 16

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1 5 10 15

Leu Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 17

<211> 36

<212> PRT

<213> Bos taurus

<400> 17

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Leu Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 18

<211> 36

<212> PRT

<213> Ovis aries

<400> 18

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Asp Asp Ala Pro Ala Glu Asp
1 5 10 15

Leu Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 19

<211> 36

<212> PRT

<213> Cavia porcellus

<400> 19

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 20

<211> 36

<212> PRT

<213> Avian

<400> 20

Tyr Pro Ser Lys Pro Asp Ser Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 21

<211> 36

<212> PRT

<213> Rana sp.

<400> 21

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Met Ala Lys Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 22

<211> 36

<212> PRT

<213> Carassius auratus

<400> 22

Tyr Pro Thr Lys Pro Asp Asn Pro Gly Glu Gly Ala Pro Ala Glu Glu
1 5 10 15

Leu Ala Lys Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 23

<211> 36

<212> PRT

<213> Dogfish sp.

<400> 23

Tyr Pro Ser Lys Pro Asp Asn Pro Gly Glu Gly Ala Pro Ala Glu Asp
1 5 10 15

Leu Ala Lys Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 24

<211> 36

<212> PRT

<213> Lampetra sp.

<400> 24

Pro Pro Asn Lys Pro Asp Ser Pro Gly Glu Asp Ala Pro Ala Glu Asp
1 5 10 15

Leu Ala Arg Tyr Leu Ser Ala Val Arg His Tyr Ile Asn Leu Ile Thr
20 25 30

Arg Gln Arg Tyr
35

<210> 25

<211> 36

<212> PRT

<213> Ovis aries

<400> 25

Ala Pro Leu Glu Pro Val Tyr Pro Gly Asp Asn Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Ala Ala Asp Leu Arg Arg Tyr Ile Asn Met Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 26

<211> 36

<212> PRT

<213> Sus sp.

<400> 26

Ala Pro Leu Glu Pro Val Tyr Pro Gly Asp Asp Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Ala Ala Glu Leu Arg Arg Tyr Ile Asn Met Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 27

<211> 36

<212> PRT

<213> Canis familiaris

<400> 27

Ala Pro Leu Glu Pro Val Tyr Pro Gly Asp Asp Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Ala Ala Glu Leu Arg Arg Tyr Ile Asn Met Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 28

<211> 36

<212> PRT

<213> Felis catus

<400> 28

Ala Pro Leu Glu Pro Val Tyr Pro Gly Asp Asn Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Ala Ala Glu Leu Arg Arg Tyr Ile Asn Met Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 29

<211> 36

<212> PRT

<213> Bos taurus

<400> 29

Ala Pro Leu Glu Pro Glu Tyr Pro Gly Asp Asn Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Ala Ala Glu Leu Arg Arg Tyr Ile Asn Met Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 30

<211> 36

<212> PRT

<213> Rattus sp.

<400> 30

Ala Pro Leu Glu Pro Met Tyr Pro Gly Asp Tyr Ala Thr His Glu Gln
1 5 10 15

Arg Ala Gln Tyr Glu Thr Gln Leu Arg Arg Tyr Ile Asn Thr Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 31

<211> 36

<212> PRT

<213> Mus musculus

<400> 31

Ala Pro Leu Glu Pro Met Tyr Pro Gly Asp Tyr Ala Thr Pro Glu Gln
1 5 10 15

Met Ala Gln Tyr Glu Thr Gln Leu Arg Arg Tyr Ile Asn Thr Leu Thr
20 25 30

Arg Pro Arg Tyr
35

<210> 32

<211> 37

<212> PRT

<213> Cavia porcellus

<400> 32

Ala Pro Leu Glu Pro Val Tyr Pro Gly Asp Asn Ala Thr Pro Glu Gln
1 5 10 15

Gln Met Ala Gln Tyr Ala Ala Glu Met Arg Arg Tyr Ile Asn Met Leu
20 25 30

Thr Arg Pro Arg Tyr
35

<210> 33

<211> 36

<212> PRT

<213> Gallus gallus

<400> 33

Gly Pro Ser Gln Pro Thr Tyr Pro Gly Asp Asp Ala Pro Val Glu Asp
1 5 10 15

Leu Ile Arg Phe Tyr Asn Asp Leu Gln Gln Tyr Leu Asn Val Val Thr
20 25 30

Arg His Arg Tyr
35

<210> 34

<211> 36

<212> PRT

<213> Alligator sp.

<400> 34

Thr Pro Leu Gln Pro Lys Tyr Pro Gly Asp Gly Ala Pro Val Glu Asp

1 5 10 15

Leu Ile Gln Phe Tyr Asn Asp Leu Gln Gln Tyr Leu Asn Val Val Thr
20 25 30

Arg Pro Arg Phe
35

<210> 35

<211> 36

<212> PRT

<213> Rana catesbeiana

<400> 35

Ala Pro Ser Glu Pro His His Pro Gly Asp Gln Ala Thr Pro Asp Gln
1 5 10 15

Leu Ala Gln Tyr Tyr Ser Asp Leu Tyr Gln Tyr Ile Thr Phe Ile Thr
20 25 30

Arg Pro Arg Phe
35

<210> 36

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 36

Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 37

<211> 12

<212> PRT
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<220>

<223> Polypeptide variation

<400> 37

Arg His Thr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 38

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 38

Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 39

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 39

Arg His Tyr Ile Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 40

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 40

Arg His Tyr Val Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 41

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 41

Arg His Tyr Leu Gln Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 42

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 42

Arg His Tyr Leu Asn Ile Val Thr Arg Gln Arg Tyr
1 5 10

<210> 43

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 43

Arg His Tyr Leu Asn Val Val Thr Arg Gln Arg Tyr
1 5 10

<210> 44

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 44

Arg His Tyr Leu Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 45

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 45

Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 46

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 46

Arg His Tyr Leu Asn Leu Val Ser Arg Gln Arg Tyr
1 5 10

<210> 47

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 47

Arg His Tyr Leu Asn Leu Val Thr Lys Gln Arg Tyr
1 5 10

<210> 48

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 48

Arg His Tyr Leu Asn Leu Val Thr Arg Asn Arg Tyr
1 5 10

<210> 49

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 49

Arg His Tyr Leu Asn Leu Val Thr Arg Gln Lys Tyr
1 5 10

<210> 50

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 50

Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Thr
1 5 10

<210> 51

<211> 12

<212> PRT

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<220>

<223> Polypeptide variation

<400> 51

Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Phe
1 5 10

<210> 52

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 52

Lys His Thr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 53

<211> 12

<212> PRT

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<220>

<223> Polypeptide variation

<400> 53

Lys His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 54

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 54

Lys His Tyr Ile Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 55

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 55

Lys His Tyr Val Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 56

<211> 12

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<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 56

Lys His Tyr Leu Gln Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 57

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 57

Lys His Tyr Leu Asn Ile Val Thr Arg Gln Arg Tyr
1 5 10

<210> 58

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 58

Lys His Tyr Leu Asn Val Val Thr Arg Gln Arg Tyr
1 5 10

<210> 59

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 59

Lys His Tyr Leu Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 60

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 60

Lys His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 61

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 61

Lys His Tyr Leu Asn Leu Val Ser Arg Gln Arg Tyr
1 5 10

<210> 62

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 62

Lys His Tyr Leu Asn Leu Val Thr Lys Gln Arg Tyr
1 5 10

<210> 63

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 63

Lys His Tyr Leu Asn Leu Val Thr Arg Asn Arg Tyr
1 5 10

<210> 64

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 64

Lys His Tyr Leu Asn Leu Val Thr Arg Gln Lys Tyr
1 5 10

<210> 65

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 65

Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Thr
1 5 10

<210> 66

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 66

Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Phe
1 5 10

<210> 67

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 67

Arg His Thr Ile Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 68

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 68

Arg His Thr Val Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 69

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 69

Arg His Thr Leu Gln Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 70

<211> 12

<212> PRT

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<220>

<223> Polypeptide variation

<400> 70

Arg His Thr Leu Asn Ile Val Thr Arg Gln Arg Tyr
1 5 10

<210> 71

<211> 12

<212> PRT

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<220>

<223> Polypeptide variation

<400> 71

Arg His Thr Leu Asn Val Val Thr Arg Gln Arg Tyr
1 5 10

<210> 72

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 72

Arg His Thr Leu Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 73

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 73

Arg His Thr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 74

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 74

Arg His Thr Leu Asn Leu Val Ser Arg Gln Arg Tyr
1 5 10

<210> 75

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 75

Arg His Thr Leu Asn Leu Val Thr Lys Gln Arg Tyr
1 5 10

<210> 76

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 76

Arg His Thr Leu Asn Leu Val Thr Arg Asn Arg Tyr
1 5 10

<210> 77

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 77

Arg His Thr Leu Asn Leu Val Thr Arg Gln Lys Tyr
1 5 10

<210> 78

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 78

Arg His Thr Leu Asn Leu Val Thr Arg Gln Arg Thr
1 5 10

<210> 79

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 79

Arg His Thr Leu Asn Leu Val Thr Arg Gln Arg Phe
1 5 10

<210> 80

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 80

Arg His Phe Ile Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 81

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 81

Arg His Phe Val Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 82

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 82

Arg His Phe Leu Gln Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 83

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 83

Arg His Phe Leu Asn Ile Val Thr Arg Gln Arg Tyr

1 5 10

<210> 84

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 84

Arg His Phe Leu Asn Val Val Thr Arg Gln Arg Tyr
1 5 10

<210> 85

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 85

Arg His Phe Leu Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 86

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 86

Arg His Phe Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 87

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 87

Arg His Phe Leu Asn Leu Val Ser Arg Gln Arg Tyr
1 5 10

<210> 88

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 88

Arg His Phe Leu Asn Leu Val Thr Lys Gln Arg Tyr
1 5 10

<210> 89

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 89

Arg His Phe Leu Asn Leu Val Thr Arg Asn Arg Tyr
1 5 10

<210> 90

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 90

Arg His Phe Leu Asn Leu Val Thr Arg Gln Lys Tyr
1 5 10

<210> 91

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 91

Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Thr
1 5 10

<210> 92

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 92

Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Phe
1 5 10

<210> 93

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 93

Arg His Tyr Leu Gln Ile Val Thr Arg Gln Arg Tyr
1 5 10

<210> 94

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 94

Arg His Tyr Leu Gln Val Val Thr Arg Gln Arg Tyr
1 5 10

<210> 95

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 95

Arg His Tyr Leu Gln Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 96

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 96

Arg His Tyr Leu Gln Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 97

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 97

Arg His Tyr Leu Gln Leu Val Ser Arg Gln Arg Tyr
1 5 10

<210> 98

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 98

Arg His Tyr Leu Gln Leu Val Thr Lys Gln Arg Tyr
1 5 10

<210> 99

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 99

Arg His Tyr Leu Gln Leu Val Thr Arg Asn Arg Tyr
1 5 10

<210> 100

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 100

Arg His Tyr Leu Gln Leu Val Thr Arg Gln Lys Tyr
1 5 10

<210> 101

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 101

Arg His Tyr Leu Gln Leu Val Thr Arg Gln Arg Thr
1 5 10

<210> 102

<211> 12

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

<400> 102

Arg His Tyr Leu Gln Leu Val Thr Arg Gln Arg Phe
1 5 10

<210> 103

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 103

Arg His Tyr Leu Asn Ile Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 104

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 104

Arg His Tyr Leu Asn Ile Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 105

<211> 12

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<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 105

Arg His Tyr Leu Asn Ile Val Ser Arg Gln Arg Tyr
1 5 10

<210> 106

<211> 12

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<220>

<223> Polypeptide variation

<400> 106

Arg His Tyr Leu Asn Ile Val Thr Lys Gln Arg Tyr
1 5 10

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<400> 107

Arg His Tyr Leu Asn Ile Val Thr Arg Asn Arg Tyr
1 5 10

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<211> 12

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<400> 108

Arg His Tyr Leu Asn Ile Val Thr Arg Gln Lys Tyr
1 5 10

<210> 109

<211> 12

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<223> Polypeptide variation

<400> 109

Arg His Tyr Leu Asn Ile Val Thr Arg Gln Arg Thr
1 5 10

<210> 110

<211> 12

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<223> Polypeptide variation

<400> 110

Arg His Tyr Leu Asn Ile Val Thr Arg Gln Arg Phe
1 5 10

<210> 111

<211> 12

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<223> Polypeptide variation

<400> 111

Arg His Tyr Leu Asn Val Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 112

<211> 12

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<223> Polypeptide variation

<400> 112

Arg His Tyr Leu Asn Val Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 113

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<400> 113

Arg His Tyr Leu Asn Val Val Ser Arg Gln Arg Tyr
1 5 10

<210> 114

<211> 12

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<223> Polypeptide variation

<400> 114

Arg His Tyr Leu Asn Val Val Thr Lys Gln Arg Tyr
1 5 10

<210> 115

<211> 12

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<223> Polypeptide variation

<400> 115

Arg His Tyr Leu Asn Val Val Thr Arg Asn Arg Tyr
1 5 10

<210> 116

<211> 12

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<400> 116

Arg His Tyr Leu Asn Val Val Thr Arg Gln Lys Tyr
1 5 10

<210> 117

<211> 12

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<223> Polypeptide variation

<400> 117

Arg His Tyr Leu Asn Val Val Thr Arg Gln Arg Thr
1 5 10

<210> 118

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<223> Polypeptide variation

<400> 118

Arg His Tyr Leu Asn Val Val Thr Arg Gln Arg Phe
1 5 10

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<223> Polypeptide variation

<400> 119

Arg His Tyr Leu Asn Leu Ile Ser Arg Gln Arg Tyr
1 5 10

<210> 120

<211> 12

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<223> Polypeptide variation

<400> 120

Arg His Tyr Leu Asn Leu Ile Thr Lys Gln Arg Tyr
1 5 10

<210> 121

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<400> 121

Arg His Tyr Leu Asn Leu Ile Thr Arg Asn Arg Tyr
1 5 10

<210> 122

<211> 12

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<223> Polypeptide variation

<400> 122

Arg His Tyr Leu Asn Leu Ile Thr Arg Gln Lys Tyr
1 5 10

<210> 123

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<400> 123

Arg His Tyr Leu Asn Leu Ile Thr Arg Gln Arg Thr
1 5 10

<210> 124

<211> 12

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<223> Polypeptide variation

<400> 124

Arg His Tyr Leu Asn Leu Ile Thr Arg Gln Arg Phe
1 5 10

<210> 125

<211> 12

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<223> Polypeptide variation

<400> 125

Arg His Tyr Leu Asn Leu Leu Ser Arg Gln Arg Tyr
1 5 10

<210> 126

<211> 12

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<400> 126

Arg His Tyr Leu Asn Leu Leu Thr Lys Gln Arg Tyr
1 5 10

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<400> 127

Arg His Tyr Leu Asn Leu Leu Thr Arg Asn Arg Tyr
1 5 10

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<223> Polypeptide variation

<400> 128

Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Lys Tyr
1 5 10

<210> 129

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 129

Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Thr
1 5 10

<210> 130

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 130

Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Phe
1 5 10

<210> 131

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 131

Arg His Tyr Leu Asn Leu Val Ser Lys Gln Arg Tyr
1 5 10

<210> 132

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 132

Arg His Tyr Leu Asn Leu Val Ser Arg Asn Arg Tyr
1 5 10

<210> 133

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 133

Arg His Tyr Leu Asn Leu Val Ser Arg Gln Lys Tyr
1 5 10

<210> 134

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 134

Arg His Tyr Leu Asn Leu Val Ser Arg Gln Arg Thr
1 5 10

<210> 135

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 135

Arg His Tyr Leu Asn Leu Val Ser Arg Gln Arg Tyr
1 5 10

<210> 136

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 136

Arg His Tyr Leu Asn Leu Val Thr Lys Asn Arg Tyr
1 5 10

<210> 137

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 137

Arg His Tyr Leu Asn Leu Val Thr Lys Gln Lys Tyr
1 5 10

<210> 138

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 138

Arg His Tyr Leu Asn Leu Val Thr Lys Gln Arg Thr
1 5 10

<210> 139

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 139

Arg His Tyr Leu Asn Leu Val Thr Lys Gln Arg Phe
1 5 10

<210> 140

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 140

Arg His Tyr Leu Asn Leu Val Thr Arg Asn Lys Tyr
1 5 10

<210> 141

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 141

Arg His Tyr Leu Asn Leu Val Thr Arg Asn Arg Thr
1 5 10

<210> 142

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 142

Arg His Tyr Leu Asn Leu Val Thr Arg Asn Arg Phe
1 5 10

<210> 143

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 143

Arg His Tyr Leu Asn Leu Val Thr Arg Gln Lys Thr
1 5 10

<210> 144

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 144

Arg His Tyr Leu Asn Leu Val Thr Arg Gln Lys Phe
1 5 10

<210> 145

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 145

Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 146

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 146

Ile Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 147

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 147

Val Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr

1 5 10

<210> 148

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 148

Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 149

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 149

Thr Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 150

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 150

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 151

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 151

Ser Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 152

<211> 16

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 152

Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 153

<211> 16

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 153

Thr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 154

<211> 16

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 154

Phe Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 155

<211> 17

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 155

Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
1 5 10 15

Tyr

<210> 156

<211> 17

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 156

Thr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
1 5 10 15

Tyr

<210> 157

<211> 17

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 157

Phe Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
1 5 10 15

Tyr

<210> 158

<211> 18

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 158

Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln
1 5 10 15

Arg Tyr

<210> 159

<211> 18

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 159

Lys Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln
1 5 10 15

Arg Tyr

<210> 160

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 160

Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 161

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 161

Gln Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 162

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 162

Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 163

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 163

Ile Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 164

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 164

Val Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 165

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 165

Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val
1 5 10 15

Thr Arg Gln Arg Tyr
20

<210> 166

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 166

Asp Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val
1 5 10 15

Thr Arg Gln Arg Tyr
20

<210> 167

<211> 22

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 167

Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu
1 5 10 15

Val Thr Arg Gln Arg Tyr
20

<210> 168

<211> 22

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 168

Asp Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu
1 5 10 15

Val Thr Arg Gln Arg Tyr
20

<210> 169

<211> 23

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 169

Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn
1 5 10 15

Leu Val Thr Arg Gln Arg Tyr
20

<210> 170

<211> 24

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 170

Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu
1 5 10 15

Asn Leu Val Thr Arg Gln Arg Tyr
20

<210> 171

<211> 24

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 171

Thr Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu
1 5 10 15

Asn Leu Val Thr Arg Gln Arg Tyr
20

<210> 172

<211> 25

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 172

Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr
1 5 10 15

Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 173

<211> 25

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 173

Ser Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His Tyr
1 5 10 15

Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 174

<211> 26

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 174

Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His
1 5 10 15

Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 175

<211> 26

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 175

Glu Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg His
1 5 10 15

Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 176

<211> 27

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 176

Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg
1 5 10 15

His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 177

<211> 27

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 177

Asp Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Arg
1 5 10 15

His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 178

<211> 28

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 178

Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu
1 5 10 15

Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 179

<211> 29

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 179

Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser
1 5 10 15

Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 180

<211> 30

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 180

Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala
1 5 10 15

Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 181

<211> 30

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 181

Ser Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala
1 5 10 15

Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 182

<211> 31

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 182

Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr
1 5 10 15

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 183

<211> 31

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 183

Asp Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr
1 5 10 15

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 184

<211> 32

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 184

Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr
1 5 10 15

Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 185

<211> 33

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 185

Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg
1 5 10 15

Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
20 25 30

Tyr

<210> 186

<211> 33

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 186

Arg Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg
1 5 10 15

Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
20 25 30

Tyr

<210> 187

<211> 33

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 187

Gln Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg
1 5 10 15

Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
20 25 30

Tyr

<210> 188

<211> 33

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 188

Asn Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg
1 5 10 15

Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg
20 25 30

Tyr

<210> 189

<211> 34

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 189

Leu Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn
1 5 10 15

Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln
20 25 30

Arg Tyr

<210> 190

<211> 34

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 190

Val Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn
1 5 10 15

Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln
20 25 30

Arg Tyr

<210> 191

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 191

Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 192

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 192

Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 193

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 193

Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 194

<211> 16

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 194

Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 195

<211> 17

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 195

Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg
1 5 10 15

Tyr

<210> 196

<211> 18

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 196

Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln
1 5 10 15

Arg Tyr

<210> 197

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 197

Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 198

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 198

Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 199

<211> 21

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 199

Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val
1 5 10 15

Thr Arg Gln Arg Tyr
20

<210> 200

<211> 22

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 200

Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu
1 5 10 15

Val Thr Arg Gln Arg Tyr
20

<210> 201

<211> 23

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 201

Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn
1 5 10 15

Leu Val Thr Arg Gln Arg Tyr
20

<210> 202

<211> 23

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 202

Ser Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn
1 5 10 15

Leu Val Thr Arg Gln Arg Tyr
20

<210> 203

<211> 24

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 203

Ala Ser Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr Leu
1 5 10 15

Asn Leu Val Thr Arg Gln Arg Tyr
20

<210> 204

<211> 25

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 204

Asp Ala Ser Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His Tyr
1 5 10 15

Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 205

<211> 26

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 205

Glu Asp Ala Ser Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu Lys His
1 5 10 15

Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 206

<211> 28

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 206

Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser Leu
1 5 10 15

Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 207

<211> 29

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 207

Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr Tyr Ala Ser
1 5 10 15

Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 208

<211> 29

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 208

Ala Pro Gly Glu Asp Ala Ser Glu Glu Leu Asn Arg Tyr Tyr Ala Ser
1 5 10 15

Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25

<210> 209

<211> 30

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 209

Glu Ala Pro Gly Glu Asp Ala Ser Glu Glu Leu Asn Arg Tyr Tyr Ala
1 5 10 15

Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 210

<211> 32

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 210

Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn Arg Tyr
1 5 10 15

Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 211

<211> 32

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 211

Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Glu Glu Leu Asn Arg Tyr
1 5 10 15

Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
20 25 30

<210> 212

<211> 33

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 212

Ile Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Glu Glu Leu Asn Arg
1 5 10 15

Tyr Tyr Ala Ser Leu Lys His Tyr Leu Asn Leu Val Thr Arg Gln Arg
20 25 30

Tyr

<210> 213

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> ACETYLATION

<400> 213

Leu Arg His Tyr Ile Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 214

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> ACETYLATION

<400> 214

Leu Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 215

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 215

Leu Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 216

<211> 24

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 216

Pro Ala Glu Asp Leu Ala Gln Tyr Ala Ala Glu Leu Arg His Tyr Leu
1 5 10 15

Asn Leu Leu Thr Arg Gln Arg Tyr
20

<210> 217

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 217

Leu Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 218

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 218

Met Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 219

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 219

Ala Arg Tyr Tyr Ser Ala Leu Arg His Phe Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 220

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> D-Ala

<220>

<221> MOD_RES

<222> (1)..(1)

<223> ACETYLATION

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 220

Xaa Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 221

<211> 18

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<223> N-terminus is bonded to -H

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<222> (18)..(18)

<223> AMIDATION

<400> 221

Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg Gln
1 5 10 15

Arg Tyr

<210> 222

<211> 20

<212> PRT

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<222> (20)..(20)

<223> AMIDATION

<400> 222

Xaa Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<223> D-Ser

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<222> (19)..(19)

<223> AMIDATION

<400> 223

Xaa Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 224

<211> 20

<212> PRT

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<222> (20)..(20)

<223> AMIDATION

<400> 224

Ala Ala Arg Tyr Ser His Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 225

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<223> D-Ile

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<222> (19)..(19)

<223> AMIDATION

<400> 225

Xaa Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 226

<211> 20

<212> PRT

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<222> (20)..(20)

<223> AMIDATION

<220>

<221> MOD_RES

<222> (1)..(1)

<223> ACETYLATION

<400> 226

Arg Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 227

<211> 18

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<223> N-terminus is bonded to -H

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<221> MOD_RES

<222> (18)..(18)

<223> AMIDATION

<400> 227

Gln Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg Gln
1 5 10 15

Arg Tyr

<210> 228

<211> 19

<212> PRT

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<223> N-terminus is bonded to -H

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<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 228

Ala Arg Phe Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 229

<211> 20

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<223> N-terminus is bonded to -H

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<222> (20)..(20)

<223> AMIDATION

<400> 229

Xaa Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 230

<211> 20

<212> PRT

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<222> (20)..(20)

<223> AMIDATION

<220>

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<222> (1)..(1)

<223> METHYLATION

<400> 230

Leu Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 231

<211> 20

<212> PRT

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<223> desamino

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 231

Xaa Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 232

<211> 19

<212> PRT

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<223> AMIDATION

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<222> (1)..(1)

<223> FORMYLATION

<400> 232

Ala Arg Tyr Tyr Ser Glu Leu Arg Arg Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 233

<211> 20

<212> PRT

<213> Artificial Sequence

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<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 233

Xaa Ala Arg Tyr Ala Ser Ala Leu Arg His Tyr Leu Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 234

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 234

Ala Arg Tyr Tyr Thr Gln Leu Arg His Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 235

<211> 20

<212> PRT

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<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 235

Leu Ala Arg Tyr Tyr Ser Asn Leu Arg His Tyr Ile Asn Val Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 236

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<223> N-terminus is bonded to -H

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<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 236

Ala Arg Tyr Tyr Asp Ser Leu Arg His Tyr Ile Asn Thr Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 237

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (1)..(1)

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<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 237

Ala Arg Tyr Tyr Ser Ala Leu Gln His Tyr Ile Asn Leu Leu Thr Arg
1 5 10 15

Pro Arg Tyr

<210> 238

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 238

Leu Ala Arg Tyr Tyr Ser Ala Leu Arg Gln Tyr Arg Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Phe
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<210> 239

<211> 18

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (1)..(1)

<223> N-terminus is bonded to -H

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<221> MOD_RES

<222> (18)..(18)

<223> AMIDATION

<400> 239

Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln
1 5 10 15

Arg Phe

<210> 240

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 240

Ser Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 241

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> ACETYLATION

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 241

Ser Arg Tyr Tyr Ala Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 242

<211> 20

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 242

Xaa Ala Arg Tyr Tyr Asn Ala Leu Arg His Phe Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 243

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MISC_FEATURE

<222> (1)..(1)

<223> D isomer of Ala

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 243

Xaa Arg Tyr Glu Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

His Arg Tyr

<210> 244

<211> 21

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (21)..(21)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> Bz

<400> 244

Xaa Leu Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile
1 5 10 15

Thr Arg Pro Arg Phe
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<210> 245

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 245

Ala Leu Tyr Tyr Ser Ala Leu Arg His Phe Val Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 246

<211> 19

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> D-Ala

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 246

Xaa Arg Tyr Tyr Ser Ala Leu Arg His Tyr Val Asn Leu Ile Phe Arg
1 5 10 15

Gln Arg Tyr

<210> 247

<211> 18

<212> PRT

<213> Artificial Sequence

<220>

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<221> MISC_FEATURE

<222> (1)..(1)

<223> MeSer

<220>

<221> MOD_RES

<222> (18)..(18)

<223> AMIDATION

<400> 247

Xaa Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Met Ile Thr Arg Gln
1 5 10 15

Arg Phe

<210> 248

<211> 20

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MISC_FEATURE

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<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 248

Arg Ile Arg Tyr Tyr Ser Ala Leu Arg His Phe Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Phe
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<210> 249

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<222> (1)..(1)

<223> N-terminal is bonded to -H

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 249

Leu Ser Arg Tyr Tyr Ser Ala Leu Arg His Phe Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
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<210> 250

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

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<222> (1)..(1)

<223> Xaa is MeIle

<400> 250

Xaa Arg Tyr Tyr Ser Ala Leu Gln His Phe Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 251

<211> 19

<212> PRT

<213> Artificial Sequence

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<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 251

Xaa Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg
1 5 10 15

Gln Arg Phe

<210> 252

<211> 20

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MISC_FEATURE

<222> (1)..(1)

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<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 252

Met Ala Arg Tyr Tyr Ser Asp Leu Arg Arg Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 253

<211> 19

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MOD_RES

<222> (19)..(19)

<223> AMIDATION

<400> 253

Ala Arg Tyr Tyr Ser Glu Leu Arg His Tyr Ile Ile Leu Ile Thr Arg
1 5 10 15

Gln Arg Tyr

<210> 254

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

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<222> (1)..(1)

<223> D-Ala

<220>

<221> MOD_RES

<222> (20)..(20)

<223> AMIDATION

<400> 254

Xaa Ala Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr
1 5 10 15

Arg Gln Arg Tyr
20

<210> 255

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 255

Ala Ser Leu Arg His Trp Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 256

<211> 35

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (25)..(25)

<223> im-DNP-HIS; 2,2-diphenylalanine Hisitidine

<220>

<221> MOD_RES

<222> (35)..(35)

<223> AMIDATION

<400> 256

Tyr Pro Ala Lys Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu
1 5 10 15

Ser Thr Tyr Tyr Ala Ser Leu Arg Xaa Tyr Leu Asn Leu Val Thr Arg
20 25 30

Glx Arg Tyr
35

<210> 257

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 257

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 258

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 258

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Ala Arg Gln Arg Tyr
1 5 10 15

<210> 259

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 259

Ala Ala Leu Arg His Tyr Leu Asn Leu Val Ala Arg Gln Arg Tyr
1 5 10 15

<210> 260

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 260

Ala Ser Leu Arg His Tyr Glu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 261

<211> 15

<212> PRT

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<223> Polypeptide variation

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<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MISC_FEATURE

<222> (13)..(13)

<223> Xaa is Ornithine

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 261

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Xaa Arg Tyr
1 5 10 15

<210> 262

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (5)..(5)

<223> Xaa is p.Cl.Pro; 4-chlorophenylalanine

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 262

Ala Ser Leu Arg Xaa Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 263

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 263

Ala Ser Leu Arg His Tyr Glu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 264

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

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<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is N-Me-Tyr

<400> 264

Ala Ser Leu Arg His Phe Glu Asn Leu Val Thr Arg Gln Arg Xaa
1 5 10 15

<210> 265

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (13)..(13)

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<223> N-alpha-ACETYLATION

<220>

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<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is N-Me-Tyr

<400> 265

Ala Ser Leu Arg His Tyr Glu Asn Leu Val Thr Arg Xaa Arg Xaa
1 5 10 15

<210> 266

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<223> N-alpha-myristoyl

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 266

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 267

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<222> (1)..(1)

<223> N-alpha-naphthateneacetyl

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 267

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 268

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is N-Me-Tyr

<220>

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<222> (1)..(1)

<223> N-alpha-ACETYLATION

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<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (13)..(13)

<223> Xaa is Ornithine

<400> 268

Ala Ser Leu Arg His Phe Glu Asn Leu Val Thr Arg Xaa Arg Xaa
1 5 10 15

<210> 269

<211> 15

<212> PRT

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<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 269

Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 270

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<222> (6)..(6)

<223> Xaa is 3-benzothienyalanine

<220>

<221> MOD_RES

<222> (7)..(7)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<400> 270

Ala Ser Leu Arg His Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 271

<211> 16

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> Xaa is 4,4'-biphenylalanine

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

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<222> (16)..(16)

<223> AMIDATION

<400> 271

Xaa Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 272

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is 3-benzothienyalanine

<400> 272

Ala Ser Leu Arg His Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 273

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<221> MOD_RES

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<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is 3-benzothienyalanine

<400> 273

Ala Ser Leu Arg His Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 274

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

<220>

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<223> N-alpha-ACETYLATION

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<222> (15)..(15)

<223> AMIDATION

<400> 274

Ala Ser Leu Arg His Trp Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 275

<211> 15

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<222> (15)..(15)

<223> AMIDATION

<400> 275

Ala Ser Leu Arg His Trp Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 276

<211> 15

<212> PRT

<213> Artificial Sequence

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<223> Polypeptide variation

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<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is 2-thienylalanine

<400> 276

Ala Ser Leu Arg Asn Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 277

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

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<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is tetrahydroisoquinoline

<400> 277

Ala Ser Leu Arg His Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 278

<211> 3

<212> PRT

<213> Homo sapiens

<400> 278

Ala Ala Ala
1

<210> 279

<211> 11

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (11)..(11)

<223> AMIDATION

<400> 279

His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 280

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MOD_RES

<222> (1)..(1)

<223> ACETYLATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is 2-thienylalanine

<400> 280

Ala Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Xaa
1 5 10 15

<210> 281

<211> 16

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (16)..(16)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is 4-Thiazolylalanine

<400> 281

Ala Ser Leu Arg His Xaa Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 282

<211> 16

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (16)..(16)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is 4-Thiazolylalanine

<400> 282

Ala Ser Leu Arg His Xaa Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 283

<211> 3

<212> PRT

<213> Homo sapiens

<400> 283

Ala Ala Ala
1

<210> 284

<211> 3

<212> PRT

<213> Homo sapiens

<400> 284

Ala Ala Ala
1

<210> 285

<211> 3

<212> PRT

<213> Homo sapiens

<400> 285

Ala Ala Ala
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<210> 286

<211> 3

<212> PRT

<213> Homo sapiens

<400> 286

Ala Ala Ala
1

<210> 287

<211> 3

<212> PRT

<213> Homo sapiens

<400> 287

Ala Ala Ala
1

<210> 288

<211> 3

<212> PRT

<213> Homo sapiens

<400> 288

Ala Ala Ala
1

<210> 289

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 289

Phe Ser Leu Arg Asn Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 290

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 290

Tyr Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 291

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 291

Ala Ser Leu Arg His Tyr Trp Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 292

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 292

Ala Ser Leu Arg His Tyr Leu Asn Trp Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 293

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

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<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 293

Ala Ser Leu Arg Ala Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 294

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

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<222> (1)..(1)

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<220>

<221> MOD_RES

<222> (14)..(14)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (5)..(5)

<223> Xaa is 3'-benzothienyalanine

<400> 294

Ala Ser Leu Arg Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 295

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 295

Ala Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 296

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 296

Ala Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Phe
1 5 10 15

<210> 297

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (11)..(11)

<223> Xaa is D form of Trp

<220>

<221> MOD_RES

<222> (11)..(11)

<223> AMIDATION

<220>

<221> MOD_RES

<222> (11)..(11)

<223> N-alpha-ACETYLATION

<400> 297

Ala Ser Leu Arg His Phe Leu Asn Leu Val Xaa Arg Gln Arg Tyr
1 5 10 15

<210> 298

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (13)..(13)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to CH3CO-

<400> 298

Leu Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 299

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (13)..(13)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to CH3CO-

<400> 299

Leu Arg His Tyr Ile Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 300

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> AMIDATION

<220>

<221> MOD_RES

<222> (13)..(13)

<223> AMIDATION

<400> 300

Leu Arg His Tyr Leu Asn Leu Leu Thr Arg Gln Arg Tyr
1 5 10

<210> 301

<211> 13

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> AMIDATION

<220>

<221> MOD_RES

<222> (13)..(13)

<223> AMIDATION

<400> 301

Leu Arg His Tyr Ile Asn Leu Ile Thr Arg Gln Arg Tyr
1 5 10

<210> 302

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (14)..(14)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (10)..(10)

<223> Xaa is Norvaline

<220>

<221> MISC_FEATURE

<222> (3)..(3)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (7)..(7)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (9)..(9)

<223> Xaa is Norleucine

<400> 302

Ala Ser Xaa Arg His Trp Xaa Asn Xaa Xaa Thr Arg Gln Xaa Xaa
1 5 10 15

<210> 303

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

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<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (14)..(14)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (3)..(3)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (7)..(7)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (10)..(10)

<223> Xaa is Norvaline

<400> 303

Ala Ser Xaa Arg His Trp Xaa Asn Trp Xaa Thr Arg Gln Xaa Xaa
1 5 10 15

<210> 304

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (14)..(14)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (3)..(3)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (7)..(7)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (9)..(9)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (10)..(10)

<223> Xaa is Norvaline

<400> 304

Ala Ser Xaa Arg His Phe Xaa Asn Xaa Xaa Thr Arg Gln Xaa Xaa
1 5 10 15

<210> 305

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (14)..(14)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (3)..(3)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (7)..(7)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (10)..(10)

<223> Xaa is Norvaline

<400> 305

Ala Ser Xaa Arg His Phe Xaa Asn Trp Xaa Thr Arg Gln Xaa Xaa
1 5 10 15

<210> 306

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

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<221> MOD_RES

<222> (12)..(12)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (12)..(12)

<223> Xaa is a pseudopeptide bond consiting of --CH2--NH--

<220>

<221> MISC_FEATURE

<222> (11)..(11)

<223> Xaa is a pseudopeptide bond consiting of --CH2--NH--

<400> 306

Arg His Tyr Leu Asn Trp Val Thr Arg Gln Xaa Xaa
1 5 10

<210> 307

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

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<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (12)..(12)

<223> AMIDATION

<400> 307

Arg His Tyr Leu Asn Trp Val Thr Arg Gln Arg Tyr
1 5 10

<210> 308

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (14)..(14)

<223> Xaa is a psuedopeptide bond consisting of --CH2--NH2

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is a psuedopeptide bond consisting of --CH2--NH2

<220>

<221> MISC_FEATURE

<222> (7)..(7)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (10)..(10)

<223> Xaa is Norvaline

<400> 308

Ala Ser Leu Arg His Tyr Xaa Asn Trp Xaa Thr Arg Gln Xaa Xaa
1 5 10 15

<210> 309

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

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<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<220>

<221> MISC_FEATURE

<222> (15)..(15)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH2--

<220>

<221> MISC_FEATURE

<222> (14)..(14)

<223> Xaa is a pseudopeptide bond consisting of --CH2--NH2--

<220>

<221> MISC_FEATURE

<222> (3)..(3)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (7)..(7)

<223> Xaa is Norleucine

<220>

<221> MISC_FEATURE

<222> (10)..(10)

<223> Xaa is Norvaline

<400> 309

Ala Ser Xaa Arg His Tyr Xaa Asn Trp Xaa Thr Arg Gln Xaa Xaa
1 5 10 15

<210> 310

<211> 9

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (9)..(9)

<223> bonded to -OCH3

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<400> 310

Ile Asn Pro Ile Tyr Arg Leu Arg Tyr
1 5

<210> 311

<211> 9

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> DISULFID

<222> (4)..(4)

<223> Sequence is linked to identical sequence by a disulfide bond

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MISC_FEATURE

<222> (9)..(9)

<223> C-terminus is bonded to -NH2

<400> 311

Ile Asn Pro Cys Tyr Arg Leu Arg Tyr
1 5

<210> 312

<211> 6

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> C-terminus is bonded to -OCH3

<220>

<221> DISULFID

<222> (1)..(1)

<223> sequence is linked to an identical sequence

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<400> 312

Cys Tyr Arg Leu Arg Tyr
1 5

<210> 313

<211> 8

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MISC_FEATURE

<222> (3)..(4)

<223> Connected by --NH---CH--CO--

<220>

<221> MISC_FEATURE

<222> (3)..(4)

<223> Identical peptide chains are connected by --(CH2)4-- at the -CH o
f --NH--CH--CO--

<400> 313

Ile Asn Pro Tyr Arg Leu Arg Tyr
1 5

<210> 314

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminus is bonded to -H

<220>

<221> MISC_FEATURE

<222> (5)..(5)

<223> C-terminus is bonded to -OCH3

<400> 314

Tyr Arg Leu Arg Tyr Tyr Arg Leu Arg Tyr
1 5 10

<210> 315

<211> 34

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> DISULFID

<222> (18)..(22)

<223>

<400> 315

Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp Met Ala
1 5 10 15

Arg Cys Tyr Ser Ala Cys Arg His Tyr Ile Asn Leu Ile Thr Arg Gln
20 25 30

Arg Tyr

<210> 316

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 316

Arg His Tyr Leu Asn Leu Ile Gly Arg Gln Arg Tyr
1 5 10

<210> 317

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (3)..(7)

<223> ACETYLATION

<400> 317

Arg His Gly Leu Asn Leu Leu Gly Arg Gln Arg Tyr
1 5 10

<210> 318

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 318

Tyr Ile Asn Leu Ile Tyr Arg Leu Arg Tyr
1 5 10

<210> 319

<211> 11

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 319

His Tyr Ile Asn Leu Ile Tyr Arg Leu Arg Tyr
1 5 10

<210> 320

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 320

Arg His Tyr Ile Asn Leu Ile Tyr Arg Leu Arg Tyr
1 5 10

<210> 321

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 321

Tyr Ile Asn Leu Leu Tyr Arg Gln Arg Tyr
1 5 10

<210> 322

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (5)..(5)

<223> Xaa is 6-amino hexanoic acid

<400> 322

Tyr Pro Ser Leu Xaa Tyr Ile Asn Leu Ile Tyr Arg Leu Arg Tyr
1 5 10 15

<210> 323

<211> 9

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 323

Ile Asn Leu Ile Tyr Arg Leu Arg Tyr
1 5

<210> 324

<211> 12

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (12)..(12)

<223> AMIDATION

<400> 324

Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10

<210> 325

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 325

Ala Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 326

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MISC_FEATURE

<222> (1)..(1)

<223> N-terminal is bonded to -H

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 326

Ala Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 327

<211> 9

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (9)..(9)

<223> AMIDATION

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<400> 327

Ala Ser Leu Arg Thr Arg Gln Arg Tyr
1 5

<210> 328

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MISC_FEATURE

<222> (6)..(6)

<223> Xaa is 2-thienylalanine

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 328

Ala Ser Leu Arg His Xaa Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 329

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<220>

<221> MOD_RES

<222> (1)..(1)

<223> N-alpha-ACETYLATION

<220>

<221> MOD_RES

<222> (15)..(15)

<223> AMIDATION

<400> 329

Tyr Ser Leu Arg His Phe Leu Asn Leu Val Thr Arg Gln Arg Tyr
1 5 10 15

<210> 330

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 330

Asp Asp Asp Asp Tyr
1 5

<210> 331

<211> 3

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 331

Gly Pro Arg
1

<210> 332

<211> 3

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 332

Ala Gly Gly
1

<210> 333

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 333

His Pro Phe His Leu
1 5

<210> 334

<211> 34

<212> PRT

<213> Homo sapiens

<400> 334

Ile Lys Pro Glu Ala Pro Gly Glu Asp Ala Ser Pro Glu Glu Leu Asn
1 5 10 15

Arg Tyr Tyr Ala Ser Leu Arg His Tyr Leu Asn Leu Val Thr Arg Gln
20 25 30

Arg Tyr

<210> 335

<211> 34

<212> PRT

<213> Artificial Sequence

<220>

<223> Polypeptide variation

<400> 335

Ser Lys Pro Asp Asn Pro Gly Glu Asp Ala Pro Ala Glu Asp Met Ala
1 5 10 15

Arg Tyr Tyr Ser Ala Leu Arg His Tyr Ile Asn Leu Ile Thr Arg Gln
20 25 30

Arg Tyr

<210> 336

<211> 204

<212> DNA

<213> Homo sapiens

<400> 336

ggagactgag ctgagtgcct gtaaaaaggc cacttcaagc cccctccacg cagccattgt 60

tgggtctgga ggaaggagga ccgctcggaa gcttctgaat gccgccctgt gatgcactca 120

ctaatggatg tgcattagtg gcgtccttcc tggccaccac gtcactctcc ctacctcaac 180

tgctggctgg agaactccgc attc 204

<210> 337

<211> 190

<212> DNA

<213> Bos taurus

<220>

<221> misc_feature

<222> (146)..(146)

<223> n is any nucleotide

<400> 337

ggggactgag ctgagtgcct gttaaaaagg ccacttcagc cccttccatg cagcctttgt 60

tggctcgaga ggaaggagga tggttccggg ggcctctgaa tgcacctaat ggatgtgcat 120

tatcagcgtc cttcctggcc actgcnggca ctctccccac ctccaccccc ggctggagaa 180

ctcagcattc 190

<210> 338

<211> 189

<212> DNA

<213> Mesocricetus sp.

<400> 338

gggactgagc tgagtgcctg taaaaaggcc acttcaagcc ccattgtggg gatagcagca 60

ggtgggcatg tctgcgcttt gaatgcctct tccctgatgc actgcgctaa tggatgtgca 120

ttaacggcgt ccttcctggc cactgtgtct acctcccttc cccaggcccc gatggagaac 180

tccgcattc 189

<210> 339

<211> 191

<212> DNA

<213> Mus musculus

<400> 339

tgggactgag ctgagtgcct gtaaaaaggc cacttcaagc cccattgtgg ggacagcagc 60

aggtgggcaa gtttgagctt tgaatgcctc ttcccgtgat gcactacgct aatggatgtg 120

cattaacagt gtccttcctg gccaccgcat cgctcgcctt tcctcaggcc ctgctggaga 180

actctgcatt c 191

<210> 340

<211> 184

<212> DNA

<213> Rattus sp.

<400> 340

agggactgag ctgagtgcct gtaaaaaggc cacttcaagc cccattgtgg ggccagcagc 60

aggtgggcaa gtctgagctt tgaatgcctc ttcccatgat gcattgcgct aatggatgtg 120

cattaacagt gtccttcctc cattgctctc ttttccttag accctgctgg agaactctgc 180

attc 184

<210> 341

<211> 408

<212> DNA

<213> Homo sapiens

<400> 341

ttctggaaaa gtagcagtca tgctcgagcc cctaacaaag gcctgtcccc cacaaaagga 60

ccattatgac caccgctgag tcagaatggt ggccgctggc acctgagctc tgtctggaaa 120

gagcggcagc agggacgtca tctagcagag cctggtgtgt ctgttatgtc cacaacatct 180

tcagcaaaga cactacttcc aggaagtcta cttggattgc agaggcgcaa gccttcattg 240

tgaaaaaagg gcttgggata aggagtggtt ctaaaagaat acatgtggct ccacatggca 300

atatacccag gtgtaataag ctcagggtaa gagagaacct gccattgctg atgcaggact 360

gtgcacacaa acttacaggc tctctactgg ggtgtcccat ggaactgg 408

<210> 342

<211> 387

<212> DNA

<213> Mus musculus

<400> 342

cctgaggaag ggcagcagtc agtgcctaaa ggccccagaa tggggccatt gtggtcatca 60

ctgagtcaca ctagtgacta ctggcacctg agctcagtct ggagtaagtg gtttcaggga 120

cgtcatctgg gagagtctgg tgcgagtcta acgtccagga cattttcagc aaagactgca 180

cctccaggaa gtccattctg actgcccaga aacaaaccct cattttgaaa agagagtttg 240

ggctaaggca agcttgggaa agggcacaaa aggctctgcg gaggaacacg cctacgcctt 300

gatccaggga acaagagtgg gatgttctaa cagccttgca ccacgccacg ccacgccatt 360

gcgatggcat tagtgctgcg tgtagga 387

<210> 343

<211> 385

<212> DNA

<213> Rattus sp.

<400> 343

cctgaggaag agcagcagtc agtgcctgag ggcctcacaa agggcccatt gtggtcctca 60

ctgagtcaga ctggtgactg ctggcacccg agctcagtct ggagtaagtg gttgcaggga 120

cgtcatccgc gagagtctgg tgtgactcta atatccagga catcttcagc aaagactgca 180

cctccaggaa gtccattctg actgcccaga aacaaaccct cattttgaaa agagcgtttg 240

agctaaggca agcttgggaa agggcacaag aggctctgca gaagaacacg cctacgcctt 300

gagccaggga acaagagcgt gatgttctaa cgcagggccc tgcgtcacac ggccttgcac 360

cacaccattc catcatgatg caatg 385

<210> 344

<211> 231

<212> DNA

<213> Homo sapiens

<400> 344

ggctgctgca ctaatgcgcg cattagtgga taaaagcagt ctcaagggtc tcttcacgag 60

gtccctttgg ctggaataaa gcaaattaaa accccattca aaggtcaatt gaaatctctt 120

tcattccagt tctctgcaca aattgattcc tctttgccct tgaggtcaaa ccgaaggctg 180

gtgaagtagc ccagctgcag tgctgcatga gagaagctca atgaaaaggc t 231

<210> 345

<211> 136

<212> DNA

<213> Bos taurus

<400> 345

ggataaaagc ggtctcaagg gtctcttcat gaggctcctt tggctgtaat aaagcaaatt 60

aaaaccccat tcaaaggtca attgaaatcg cttccattcc cattctttgc acaaattgat 120

tcctctttgc ccttga 136

<210> 346

<211> 226

<212> DNA

<213> Mus musculus

<400> 346

ggctactgtg ctaatacatg cattagtgga tgaaagccgt ctcaaggggc tcttcaccag 60

ggccctttgg ctgtaataaa gcaaattaaa accccatcca aaggtcaatt gaaatctctt 120

tcattcttct tctccacaca aattgattcc tctttgccct tgaggttgca ctgaatgcca 180

taaaggggcc cagctgtagc tggatgggaa cagcctgaaa atggct 226

<210> 347

<211> 136

<212> DNA

<213> Oryctolagus cuniculus

<400> 347

ggataaaagc tgtctcaagg ggctcttcac cgtggccctt tggctgtaat aaagcaaatt 60

aaaaccccat tcgaaggtca attgaaatct ctttcattcc acttctccac acaaattgat 120

tcctctttgc ccttga 136

<210> 348

<211> 435

<212> DNA

<213> Mus musculus

<400> 348

gactgagctg agtgcctgta aaaaggccac ttcaagcccc attgtgggga cagcagcagg 60

tgggcaagtc tgagctttga atgcctcttc ccgtgatgca ctacgctaat ggatgtgcat 120

taacagtgtc cttcctggcc accgcatcgc tcgcctttcc tcaggccctg ctggagaact 180

ctgcattcct gaggaagggc agcagtcagt gcctaaaggc cccagaatgg ggccattgtg 240

gtcatcactg agtcacacta gtgactactg gcacctgagc tcagtctgga gtaagtggtt 300

tcagggacgt catctgggag agtctggtgc gagtctaacg tccaggacat tttcagcaaa 360

gactgcacct ccaggaagtc cattctgact gcccagaaac aaaccctcat tttgaaaaga 420

gagtttgggc taagg 435

<210> 349

<211> 464

<212> DNA

<213> Homo sapiens

<400> 349

gactgagctg agtgcctgta aaaaggccac ttcaagcccc ctccacgcag ccattgttgg 60

gtctggagga aggaggaccg ctcggaagct tctgaatgcc gccctgtgat gcactcacta 120

atggatgtgc attagtggcg tccttcctgg ccaccacgtc actctcccta cctcaactgc 180

tggctggaga actccgcatt cttctggaaa agtagcagtc atgctcgagc ccctaacaaa 240

ggcctgtccc ccacaaaagg accattatga ccaccgctga gtcagaatgg tggccgctgg 300

cacctgagct ctgtctggaa agagcggcag cagggacgtc atctagcaga gcctggtgtg 360

tctgttatgt ccacaacatc ttcagcaaag acactacttc caggaagtct acttggattg 420

cagaggcgca agccttcatt gtgaaaaaag ggcttgggat aagg 464

<210> 350

<211> 236

<212> DNA

<213> Mus musculus

<400> 350

ggctggggtg ggctactgtg ctaatacatg cattagtgga tgaaagccgt ctcaaggggc 60

tcttcaccag ggccctttgg ctgtaataaa gcaaattaaa accccatcca aaggtcaatt 120

gaaatctctt tcattcttct tctccacaca aattgattcc tctttgccct tgaggttgca 180

ctgaatgcca taaaggggcc cagctgtagc tggatgggaa cagcctgaaa atggct 236

<210> 351

<211> 231

<212> DNA

<213> Homo sapiens

<400> 351

ggctgctgca ctaatgcgcg cattagtgga taaaagcagt ctcaagggtc tcttcacgag 60

gtccctttgg ctggaataaa gcaaattaaa accccattca aaggtcaatt gaaatctctt 120

tcattccagt tctctgcaca aattgattcc tctttgccct tgaggtcaaa ccgaaggctg 180

gtgaagtagc ccagctgcag tgctgcatga gagaagctca atgaaaaggc t 231

<210> 352

<211> 17

<212> DNA

<213> Unknown

<220>

<223> Homologous among Homo sapiens, Bos taurus, Oryctolagus cuniculus, Mus musculus, and Mesocricetus sp.

<400> 352

ctaatggatg tgcatta 17