

The anorectic gut hormones: GLP-1 and co-secreted peptides

The anorectic gut hormones GLP-1 (glucagon-like peptide 1) and co-secreted peptides such as oxyntomodulin and PYY are among the prime candidates for manipulation in the development of new therapies for obesity. Here their secretion and actions in pathways regulating food intake are briefly outlined.

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The regulation of food intake or energy balance is a huge and complex topic that has achieved additional importance in recent years, not only because of major advances in the field, but also because of the increased prevalence of obesity in western countries, which has been called an “epidemic” by the WHO and is clearly a major public health problem. In a brief article it would be impossible to give much more than a list of the endocrine and neurological, peripheral and central short-term and long-term mechanisms currently known to be involved, each being the subject of quite intensive research both in the academic sector and the pharmaceutical industry. It is evident that an effective and safe “slimming pill” with few and mild side effects, if such could be found, would be a commercial success on an unprecedented scale.

The whole area of nutrition, obesity and metabolism has recently been the subject of a special issue of Gastroenterology [1], which contains many excellent and detailed reviews, both of aspects mentioned here and many more besides. Another review of signalling molecules in appetite control [2] also provides a wide-ranging survey.

In this article I want to focus on GLP-1 (glucagon-like peptide 1) and the peptides with which it is co-secreted in the intestine, these being among the principal gut hormones involved in the suppression of food

Anorexigenic	Orexigenic
GLP-1	Ghrelin
Oxyntomodulin	
PYY	
CCK	

Table 1. Principal gut hormones* involved in the regulation of food intake.

*Defined restrictively as those that are secreted by endocrine cells in the gastrointestinal mucosa and act at least in part via the circulation, thus excluding hormones that are chiefly pancreatic (e.g. insulin, pancreatic polypeptide) and also neuropeptides that do not have this origin and action.

intake. Hormonal and neurological effects on energy homeostasis can be classified into the stimulation or suppression of appetite, satiety signals, and the regulation of metabolism or energy expenditure. Sometimes a particular experimental design does not permit a sharp distinction between such modes of action. The wider terms “orexigenic”, in the sense of increasing appetite or food intake, and “anorexigenic” in the opposite sense, thus have their use, just as measurement of weight gain or weight loss may be the experimental endpoint, leaving the mechanisms by which it might have occurred to be inferred from other observations. The gut hormones involved in the regulation of food intake are restrictively defined and listed in Table 1, which hence excludes a number of other hormones, neuropeptides, neurotransmitters and factors that act intracellularly.

One reason for focussing on these gut hormones is that it may prove technically simpler to modify their processing and action on food intake, at least peripherally, while keeping the side effects of such manipulations within acceptable limits. Drugs acting on neuropeptide or classical neurotransmitter pathways will almost certainly have more widespread central effects than just the desired action.

GLP-1

GLP-1 is in fact both a gut hormone and a cerebral neuropeptide with a very limited site of production in the brain.

It is produced by tissue-specific post-translational processing from the glucagon precursor, which is expressed not only by the β -cells of the pancreatic islets, but also by the endocrine L cells of the ileal and colonic mucosa, as well as by neurons chiefly located in the caudal section of the nucleus of the solitary tract (NTS). While the action of prohormone convertase 2 produces glucagon and a series of largely inactive precursor fragments in the β -cells, the action of prohormone convertase 1/3 produces GLP-1 and a second glucagon-like peptide, GLP-2, in the L-cells and the neurons, together with extended forms of glucagon that have little or no glucagon-like metabolic action. One of these forms is a C-terminally extended form of glucagon, called oxyntomodulin because of its capacity (like GLP-1) to suppress gastric acid secretion. While GLP-1 is perhaps best known through its incretin role as the most

potent gut hormone to stimulate glucose-dependent insulin secretion in response to food, both GLP-1 and its co-synthesised partner, oxyntomodulin, have proved to be quite potent anorexigenic peptides through both peripheral and central actions.

Neither the release nor the mechanism of action of these gut hormones is as simple as might at first be supposed. Far from just being released in response to the

direct action on the L cells of nutrients in the intestinal lumen, which may be regarded as a fail-safe mechanism relevant for prolonged responses, their release is probably in the first instance governed by the autonomic nervous system, being stimulated by intrinsic cholinergic nerves and the neuropeptide GRP (gastrin-releasing peptide), while being inhibited by sympathetic nerves. Secondly, there is a possible influence from other gut hormones, secretion being stimulated by duodenally produced hormones such as CCK and the other incretin, GIP (glucose-dependent insulinotropic polypeptide), and inhibited by the locally acting somatostatin. These hormonal effects have been demonstrated by pharmacological experiments in different animals with varying results according to species and protocol, and their relevance to human physiological responses is unconfirmed. Interestingly, there is evidence that GLP-1 secretion from both L-cells and cerebral neurons is stimulated by the anorexigenic adipokine, leptin.

While GLP-1 certainly acts via its class-B 7-transmembrane-domain, G-protein-linked receptor (GLP-1R) on β -cells and neurons, it is by no means clear that such direct actions are the only mechanism involved. Even its initial action on the insulin

secretory response may be mediated more by nervous reflexes initiated by its action on vagal afferents than by direct action on the β -cells [3], and the hepatic vagal receptor mechanism appears to differ from that of the well-described GLP-1R in not being inhibited by the GLP-1 antagonist exendin (9-39) [4]. Although GLP-1Rs are present in gastric glands [5], the potent effects of GLP-1 to inhibit gastric acid secretion and gastric emptying are nullified by vagal afferent denervation, and it appears that the inhibitory effect is vagally mediated through central pathways involving GLP-1Rs both in the brain stem and on vagal afferents. These pathways also mediate the anorectic effect of peripheral GLP-1. The slowing of gastric emptying by GLP-1 may in fact be at least as important a factor in reducing postprandial glycaemia as its direct stimulation of insulin secretion.

The vagal pathways mediating the anorectic effect of peripheral GLP-1 also mediate the satiety effect of sulphated CCK, released from the upper small intestine, which acts on CCK-1 receptors present on vagal afferents. The satiety signal from peripheral CCK is, however, very short-lived.

When GLP-1Rs were first located in the brain [6], it was immedi-

ately apparent that the NTS and the arcuate nucleus were among the regions showing the highest level of receptor expression. These are respectively the sites of synthesis and action of GLP-1 in the brain. Subsequent, more detailed studies of receptor distribution and neuronal activation by centrally administered GLP-1 agonists showed that the hypothalamic dorsomedial and paraventricular nuclei are also sites of GLP-1 action, particularly the parvocellular region of the paraventricular nucleus containing neurons expressing the anorexigenic peptide, corticotrophin releasing hormone (CRH). In the arcuate nucleus, GLP-1 activates neurons containing proopiomelanocortin (POMC), the precursor of other anorexigenic peptides [7]. These hypothalamic nuclei are seen as key areas in the integrative regulation of food intake. GLP-1 also activates cells in the NTS and the area postrema, these forming part of the dorsal vagal complex [8]. In addition, the arcuate nucleus and area postrema are sites at which circulating signal molecules have access to the central nervous system. The fact that the NTS both synthesises and responds to GLP-1 suggests that it is an important relay and amplification site for GLP-1 signals, capable of responding to circulating inputs to the area postrema from both leptin and GLP-1. In passing, it is interesting that

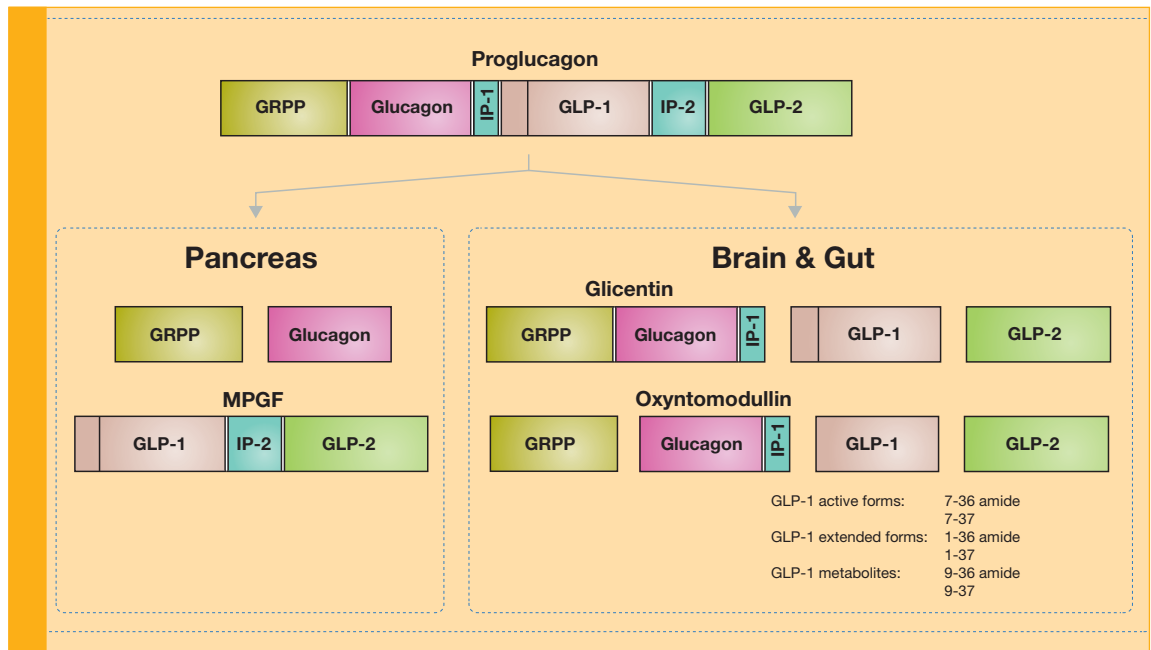


Figure 1. Tissue-specific processing of proglucagon in pancreas vs. brain and gut. MPGF, major proglucagon fragment; GRPP, glicentin-related pancreatic polypeptide; IP, intermediate peptide.

by far the highest brain levels of GLP-1 are found in the hypothalamus, where it is present in nerve endings, while the small number of GLP-1-synthesising cell bodies in the NTS make little contribution to the low brain stem level.

Oxyntomodulin

Co-synthesised with GLP-1 from the same precursor in L cells and neurons, oxyntomodulin should in principle be formed in equimolar amounts and released at the same sites and in response to the same stimuli as GLP-1. However, post-translational processing of a peptide from its precursor is not a fully efficient process, for example, there is evidence that the amount of the principal active form of GLP-1, GLP-1 (7-36) amide, elaborated in the rat intestine, may correspond to less than half of the total amount of precursor synthesised, the rest appearing as inactive extended forms or being broken down intracellularly [9]. Therefore it should not be assumed that GLP-1 and oxyntomodulin levels parallel each other at all release sites. Oxyntomodulin is quite potently anorexigenic when administered peripherally or centrally to rodents and intravenous infusion produces a pronounced anorectic effect in humans [10]. This effect may in part be due to its suppression of circulating levels of the gastric orexigenic hormone ghrelin. Oxyntomodulin has little incretin effect, however. This fact alone suggests that it may act on a specific receptor different from GLP-1R, on which it shows approximately 1% agonist activity. GLP-1R is, however, the only receptor for oxyntomodulin which has yet been identified. A manganese-enhanced magnetic resonance study of neuronal activation following peripheral injection of GLP-1 and oxyntomodulin in mice showed a substantial difference in the patterns of neuronal activity induced in the arcuate, paraventricular, supraoptic and ventromedial hypothalamic nuclei in response to the two peptides [11]. This difference is difficult to explain from a dogma of co-equal synthesis and release with GLP-1, combined with action on the same receptor. A further feature of oxyntomodulin is that its administration to humans increases voluntary activ-

ity [12], thus adding an increase in energy expenditure to its potential for inducing weight loss.

PYY

While peptide YY, a member of the PP-fold peptide family, is produced from a different precursor from that of GLP-1, it is secreted from the same L cells that elaborate GLP-1 in the intestine. This is not to assume that all L cells express GLP-1 and PYY in the same ratio; the distribution of PYY along the intestinal tract is perhaps somewhat more distal than GLP-1, with a very high level in the rectum. Nevertheless, secretion from the same cell type does mean that PYY secretion is subject to the same stimulatory and inhibitory influences as GLP-1 secretion. Like GLP-1, PYY inhibits gastric secretion and emptying, but it has additional effects that are not shown by GLP-1. Unlike GLP-1, its major circulating anorexigenic form is the metabolite PYY(3-36), the product of digestion by the widely distributed enzyme dipeptidyl peptidase IV (DP4). DP4 is also the rapid first step in the breakdown of circulating GLP-1 and oxyntomodulin, whereby the anorectic effects of these two peptides are thought to be nullified, along with most, but not quite all, of their other actions. PYY(3-36) acts selectively on the Y2 receptor, an inhibitory presynaptic receptor on the orexigenic NPY neurons of the arcuate nucleus. A direct inhibitory action of circulating PYY(3-36) on these neurons has been demonstrated, leading to a disinhibition of the anorexigenic POMC neurons [13]. PYY(3-36) may also act in a similar manner to GLP-1 via vagal afferents and the dorsal vagal complex, and the effects of these two peptides to inhibit food intake are additive [14].

Therapeutic interventions

It is evident that even the rough outline of the systems given here suggests a number of points susceptible to therapeutic intervention for the treatment of obesity. However, the initial interventions that are just now coming to fruition have focused on the treatment of type 2 diabetes mellitus. Interest has centred on the incretin effect of GLP-1 and how this may be potentiated or prolonged. The

main drives have been i) to develop GLP-1 agonists of prolonged action, by promoting its binding to albumin, by combining it with albumin, or by otherwise increasing its resistance to rapid breakdown by DP4, and ii) to develop orally active small molecule inhibitors of DP4, thus prolonging the activity of endogenous GLP-1.

Among long-acting GLP-1 agonists, the first to be released was Exenatide, a synthetic but otherwise unmodified form of exendin-4, the potent GLP-1 agonist peptide present in Gila monster (*Heloderma suspectum*) venom. This has a glycine residue instead of an alanine residue at position 2, making it a poor substrate for DP4, so that its extended half-life makes it appear many times as active as GLP-1 *in vivo*. Exenatide has to be injected twice daily and has been followed up by a long-acting slow release preparation for weekly injection. Treatment with exenatide has been associated with a significant mean weight loss of between 1.5 and 2.8 kg in different trials, whereas insulin glargine therapy in a control group, for example, was associated with a weight gain of 1.8 kg over the six month trial period.

Liraglutide is a synthetic form of GLP-1 modified by two amino acid substitutions and a fatty acid acyl group to promote binding to albumin. Treatment with this preparation was also associated with mean weight loss of 2.9 kg. Other GLP-1 agonists under study include a recombinant GLP-1-albumin hybrid, an exendin-4-albumin conjugate and a sequence-modified and extended form of exendin-4.

Of orally active DP4 inhibitors, sitagliptin has recently been released. These agents increase postprandial plasma levels of GLP-1 and GIP and significantly improved HbA1c as an index of glycaemic control. Though significant weight loss under sitagliptin treatment has been reported [15], the overall assessment of DP4 inhibitor treatment so far is that it is weight neutral [16]. This may reflect the fact that GLP-1 and PYY(3-36) anorectic effects are additive, and DP4 inhibition would diminish the production of the latter while

prolonging the action of the former.

While the effects of GLP-1 agonist treatment on weight are in the nature of incidental findings in clinical trials directed at improving diabetic control, they are nevertheless important and promising results, as they are the first experiments in which weight loss has been induced by long-term gut hormone therapy in a large number of patients. This points the way to therapy specifically directed at obesity that mimics the natural anorectic actions of gut hormones.

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