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Physiological endocrine control of energy homeostasis and postprandial blood glucose levels

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Abstract. – **The aim of this review is to analyze the different components and the feedback mechanisms involved in the normal control of energy homeostasis and postprandial blood glucose levels. Such control involves exogenous and endogenous factors: while the former include quantity and quality of food intake, the latter involve the balance of glucose intestinal absorption (postprandial period), glucose production and release by the liver and its consumption by peripheral tissues. Adequate secretion and peripheral metabolic effects of insulin play a key role in the control of both processes. Insulin secretion is controlled by the level of circulating substrates and by gastrointestinal hormones. The mechanism for the immediate control of blood glucose levels is modulated by energy homeostasis, with the participation of the above mentioned hormones and others produced at the classical endocrine system and adipose tissue, whose actions integrate at the central nervous system. The alteration of such delicate mechanism of control causes diseases such as diabetes; therefore, identification of the multiple components of this mechanism and comprehension of its normal function would facilitate the selection of effective strategies for diabetes prevention and treatment.**

Key Words:

Energy homeostasis, Postprandial blood glucose levels, Diabetes, Neuroendocrine control.

Introduction

Near normoglycemia is one of the main challenges in the treatment of diabetes to avoid the development and progression of diabetes complications[1,2].

Various studies have emphasized the importance of postprandial hyperglycemia, since its association with fatal and nonfatal cardiovascular events is greater than that of fasting hyperglycemia[3], and even moderate increased levels constitute a risk factor[4]. The risk increases when postprandial hyperglycemia associates with postprandial hyperlipemia; hyperglycemia increases lipid peroxidation, thus increasing lipoprotein atherogenic capacity and decreasing plasma antioxidants[5]. Presumably, the higher the glycemic increment the higher the decrease of antioxidants will be[5]. Screening is essential in people with diabetes presenting fasting glycemia and HbA_{1c} levels within normal range, and markedly increased postprandial glycemia[6].

The regulation of postprandial glycemia is complex, and the magnitude of glycemic variations depends on multiple factors, namely, food composition, the action of gastrointestinal hormones and digestive enzymes, insulin secretion, enhancement or inhibition of hepatic glucose production, and peripheral glucose uptake. These factors act during food intake, together with other factors acting during the day to keep an adequate balance between intake and total caloric consumption (energy balance) (Figure 1).

The regulation of energy balance is important because the volume and composition of each food varies considerably from day to day and person to person; the absence of energy balance could result in the uncoupling of energy intake and caloric consumption. To explain this model of energy homeostasis, Kennedy proposed that signals originated in fat deposits would act at brain level, decreasing appetite[7]. When various intestine pep-

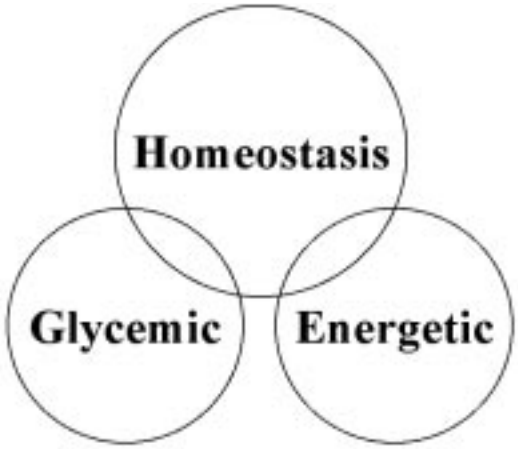

Figure 1. Integration of glycemic and energy homeostasis. Combination of signals and effectors of two systems participating in the regulation of glycemic and energy homeostasis.

tides and their receptors at the central nervous system (CNS) were identified, it was postulated that their release in response to food intake generated "immediate" satiety signals at brain level, determining the interruption of food intake[8]. The model was completed when leptin was identified as a long-term adipocyte signal released in relation to the size of body fat deposits; together with insulin, leptin acts directly at the CNS, inhibiting food intake[9].

To facilitate the understanding of postprandial glycemic regulation and energy homeostasis, we will first describe the signals and mechanisms involved, then the regulatory center, and finally how both models act.

Ghrelin

Ghrelin is a 28 aminoacid lipophilic peptide with a labile octanoic acid side chain at the serine residue, mainly expressed in enterochromaffin cells of the gastric mucosa[10]. Ghrelin has also been identified in pancreatic non-β islet cells[11]. It would circulate in plasma bound to HDL-cholesterol particles, with decreased concentration peaks after food intake, together with increased levels of insulin[12].

Ghrelin levels are low in positive energy balance conditions; they increase in people on a diet and have a negative correlation with fasting insulinemia, body weight, body mass index (BMI), and adipocyte volume[12]. On the other hand, ghrelin levels have a positive correlation with age, insulin sensitivity, total HDL-cholesterol, HDL2 and HDL3. Patients with gastric bypass present low plasma ghrelin levels, accounting for decreased appetite[13].

Ghrelin stimulates the secretion of somatotrophin, ACTH, cortisol and prolactin[14]. Its effect upon insulin and glucagon varies as a function of the dose used: a low dose inhibits insulin secretion and stimulates glucagon secretion *in vitro*, whereas high doses stimulate insulin secretion without modifying glucagon secretion[15]. However, given the circulating levels of ghrelin, it would not exert a modulatory physiological effect *in vivo* of the secretion of these hormones[15].

Ghrelin metabolic effects are opposite to those of leptin: it stimulates food intake, potentiates carbohydrate utilization, reduces fat utilization, increases gastric motility and stomach acid secretion, and would act as an adipocity signal favoring weight gain[12]. These results suggest that both the effects and the secretion of ghrelin are opposite to those of leptin.

Oxyntomodulin (OXM)

Oxyntomodulin is a 37 aminoacid peptide resulting from the processing of proglucagon (Figure 2) in intestine L cells[16], which inhibits the secretion of oxyntic glands in the stomach[17].

OXM is released in the postprandial period by distal intestine endocrine cells that produce peptides such as PYY[18] and GLP-1[19], and its circulating levels remain elevated for several hours after food intake[20]. OXM levels are markedly high in people with morbid obesity and jejuno-ileal bypass[21] and are associated with nervous anorexy and weight loss[22]. Since OXM inhibits appetite at hypothalamic level, it would be part of a negative feedback mechanism.

OXM binds to GLP-1 receptor, but with lower affinity[23]. In rats, however, the inhibitory effect of OXM on appetite is reduced by blocking GLP-1R receptors[24].

OXM infusion for 90 min reduces the calorie content of food (-20%); this effect lasts for 12 h (-11%), without modifying significantly calorie intake in the 24 h-period[25]. OXM also decreases significantly the concentration of plasma ghrelin (appetite-stimulating) before food intake, being such inhibition one of OXM´s satiety mechanisms.

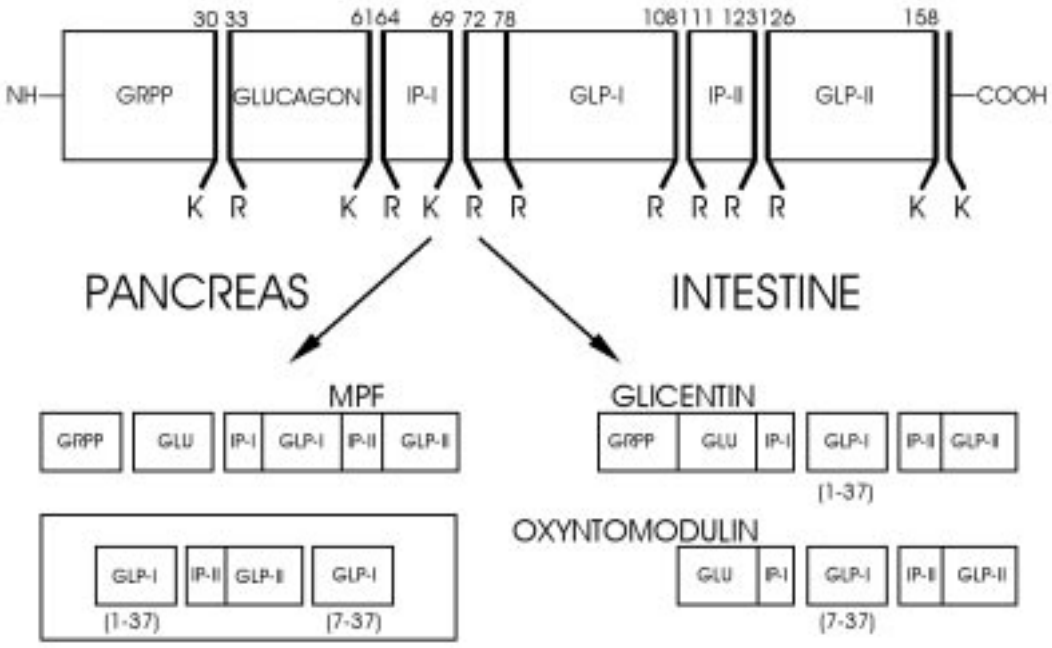

Figure 2. Proglucagon and processing systems. Different processing of proglucagon molecule in islet α-cells and intestine L-cells. Numbers and letters indicate peptidase cleavage sites.

OXM would act in the arcuate nucleus (AN) of the hypothalamus, where energy homeostasis signals are monitored and integrated[24]. It would also act at in the vagus nerve, transmitting afferent signals to the brain via ghrelin[26].

Peptide YY (PYY)

This is a gastrointestinal peptide of 36 aminoacids mainly produced by intestinal L cells[27] which belongs to the pancreatic polypeptide family together with neuropeptide Y (NPY)[28]. PYY becomes PYY_{3-36} through the action of dipeptidyl peptidase IV (DPP IV); circulating levels increase by 33% 15 min after food intake and keep elevated for approximately 90 min. The peak of PYY is proportional to the amount of calorie intake; PYY decreases appetite and consequently food intake through a negative feedback mechanism[28,29]. The early release of PYY (15 min) is initially produced by nerve stimulation and then as a function of the intestine nutrient content.

PYY_{3-36} has a 70% homology with NPY and interacts with NPY receptors of different subtypes (Y1, 2, 4 and 5)[30]. PYY_{1-36} binds to receptor Y1, 2 and 5, while PYY_{3-36} is more selective for Y2 receptor. Y2R is an inhibitory presynaptic receptor highly expressed in NPY neurons of the AN.

Intraperitoneal injection of PYY_{3-36} inhibits the daily food intake in a dose-dependent manner acting on Y2 receptors, as demonstrated by its inactivity in mice with KO of that receptor.

In man, PYY perfusion in amounts capable of reproducing the levels obtained in the postprandial period, reduce appetite and food intake by 33% for 24 h, and decrease significantly fasting and preprandial ghrelin levels[28]. Such prolongued effect suggests that, opposed to other intestinal peptides, PYY_{3-36} is a long-term appetite regulator[31].

In the AN, PYY_{3-36} would diminish the GABAergic tone through which NPY inhibits proopiomelanocortin (POMC) neurons, allowing the expression of their inhibitory effect upon appetite[32].

There is a negative correlation between PYY levels and BMI, so that basal and postprandial PYY levels are lower in obese people. Since PYY is a potent appetite inhibitor[33], decreased PYY levels would favour obesity. Opposite to what occurs with other peptides, there is no PYY_{3-36} resistance in obesity, reason why it has been proposed for its treatment. On the other hand, PYY

levels are high in patients with jejuno-ileal bypass, thus explaining their decreased appetite[34].

Cholecystokinin (CCK)

This peptide is produced by cells from the high portion of the small intestine and at the same time in nerve terminals of the central and peripheral nervous system[35].

CCK is released to peripheral circulation after food intake; it stimulates postprandial secretion of the exocrine pancreas, the contraction of the gall bladder, gastric emptying, and intestine motility[35].

CCK also stimulates insulin secretion *in vivo* and *in vitro* in different species[36], and specific antagonists of its receptor inhibit postprandial insulin secretion[37]. Since basal insulin secretion is not affected, increased glycemia should be previous to CCK action.

Circulating CCK levels measured *in vivo* in the postprandial period would no affect glucose-induced insulin secretion, which is not inhibited by blockade of its receptor[38]. However, since it is also expressed in pancreatic nerve terminals[39], CCK could regulate insulin secretion through nerve stimulation. Alternatively, it could increase insulin secretion indirectly by stimulating GIP and GLP-1 release[40].

CCK administration to people with type 2 diabetes reduces postprandial hyperglycemia, thus suggesting increased insulin secretion[41]. Although such effect is less marked than that produced by GLP-1, it has been suggested that CCK would be a potential agent for the treatment of type 2 diabetes[36].

Glucagon-like Peptide 1 (GLP-1)

GLP-1 is produced and secreted by L cells in the distal ileum and colon[42]. These cells contain proglucagon and their processing by preprotein convertase produces GLP-1 and other bioactive peptides (Figure 2).

Preprotein convertases are intracellular serine endoproteases[43,44] and expression of proconvertases 1 and 2 (PC1 y PC2) is restricted to regulated nervous and endocrine secretion[44]. These preproteins play a key role in the posttranscriptional processing of hormone precursors such as proinsulin, POMC, and proglucagon[45,46]. Some of them are organ-specific; intestine L cells process proglucagon to glicentin, OXM, GLP-1 and 2 via PC1, while α-pancreatic cells process proglucagon to glucagon through PC2[47] (Figure 2).

The lack of expression of these enzymes manifests clinically not only as a deficit in glucagon and GLP-1 production, but is also accompanied by severe obesity, altered processing of POMC (to ACTH), proinsulin, hypocortisolemia, hypoglycemia, and deficit in intestine absorption of sugars and other nutrients, forcing life-long parenteral feeding[48].

L-cells rapidly release GLP-1 in response to nutrients, especially fats and carbohydrates[42,49], in a biphasic way. The first phase is regulated in a complex manner: GIP secreted by intestinal K-cells would stimulate acetylcholine release in celiac plexus terminals[50], which interact with type M1 muscarinic receptors[51]. Atropin (non-selective antagonist of muscarinic receptors) reduces GLP-1 integrated response during oral glucose load and after food intake[52], showing the importance of the vagus nerve to mediate its secretion. The second phase of secretion is the consequence of the direct action of nutrients on intestinal L-cells. The control of GLP-1 secretion would be complete with a self-regulating negative feedback mechanism[53].

Once GLP-1 is released, it is degraded by the action of dipeptidyl-dipeptidase IV (DPP-IV)[54,55], which would also degrade other peptides, such as hypophyseal peptide (adenylate-cyclase activator), bradykinin, and GIP[45]. The administration of DPP-IV inhibitors to people with type 2 diabetes decreased significantly HbA1c levels and the amplitude of postprandial glycemic oscillations[56].

GLP-1 binds to specific receptors located at different tissues: islet cells, stomach, and CNS. In pancreatic β-cells, it couples to a specific G-protein; when this protein binds to the hormone, it activates adenylate cyclase, producing an increase of adenosine-3',5'-cyclic monophosphate (cAMP)[57] which in turn activates protein kinase A (PKA)[58]. Through this pathway, GLP-1 promotes the phosphorilation of GLUT2 and of K^-_{ATP} and Ca^{2+} channels[58-60]. cAPM would also act independently of PKA, interacting with GEF2 or Epac2 (cAMP sensor) forming a complex with Rim2, and activating Rab3 (a component of the exocytotic cell machinery)[61].

The affinity of cAMP with PKA (Kd) is 100 nM[62], being 10 µM with GEF2. Since the basal concentration of cAMP in β-cells is in the micromolar range[62], PKA substrates would be maximally phosphorilated[58]. Therefore, the role of PKA and GEF2 would be different: GEF2 would act only when cAMP increases in response to a stimulus[61].

GLP-1 stimulates insulin secretion and inhibits glucagon secretion acting on islet β- and α-cell receptors[63]. The inhibitory effect on glucagon secretion is maintained even in people with diabetes of different etiology[64].

The insulinotropic effect of GLP-1 has extrapancreatic components as well. Outside the islet, GLP-1 acts as a hepatic portal glucose "sensor" which is activated whenever a glucose gradient is established between the portal and peripheral region, as it occurs after food intake. Such hepatic and portal sensor is the first to contact the ingested glucose, and would modulate insulin secretion through a neurohumoral pathway. This hypothesis is supported by the existence of neurons from the enteric nervous system in the pancreas which express $K-_{ATP}$ channels that would send signals when they get in contact with glucose[65]. The glucose portal sensor promotes first-phase insulin secretion, which is absent in double-KO mice for GLP-1 and GIP receptors (DIRKO mice)[65]. In these mice there is also a marked decrease of second-phase insulin secretion. Such insulinotropic effect is responsible for approximately 50% of the insulin secreted after food intake and disappears progressively during the development of type 2 diabetes due to the decrease of GLP-1 and GIP production and the lower response of β-cells, particularly to GIP[66].

The activation of the glucose sensor also increases glucose utilization by a mechanism independent of insulin action, requiring the presence of Glut2 and GLP-1 receptor to act[67].

GLP-1 inhibits gastric emptying – through the activation of its receptors in the stomach and at the hypothalamus – and food intake[68], so that GLP-1 prolongued administration decreases body weight[42].

The importance of GLP-1 in glycemic homeostasis was verified using mice with KO of GLP-1 receptors: these mice did not develop severe diabetes, but they presented defective insulin secretion during oral glucose tolerance test (OGTT)[65]. On the other hand, GLP-1 secretion after food intake decreased in type 2 diabetes[69].

Chronic administration of GLP-1 to rodents activates the transcription of genes involved in β-cell differentiation and function, and in islet neogenesis: Pdx-1, Glut2, glucokinase and insulin[70,71].

GLP-1 perfusion nomalizes fasting glycemia in people with type 2 diabetes[72] and decreases glycemic fluctuations after food intake[53]. These effects are due to the combined effect of enhanced insulin secretion, inhibited glucagon secretion, and gastric emptying.

The effect of a single GLP-1 injection is short because of its rapid metabolization, thus preventing its use in the treatment of type 2 diabetes. GLP-1 analogues such as exendin-4 and liraglutide administered to people with type 2 diabetes could significantly reduce HbA1c, representing a valid alternative for the treatment of this type of diabetes[73].

Glucose-Dependent Insulinotropic Polypeptide (GIP)

GIP is a 42 aminoacid peptide synthetized by enteroendocrine K-cells of the proximal intestine[74]. Although it was formerly called gastric inhibitory peptide (GIP), its main effect is insulinotropic; therefore, it was renamed glucose-dependent insulinotropic polypeptide. GIP belongs to the family of secretins, presenting homology with some of its members: secretin, glucagon, GLP-1 and 2, VIP, and GRRH. As most of them, GIP has a synthesis precursor of higher molecular weight[75].

The passing of food to the intestine stimulates the release of GIP, and the magnitude of the stimulus is proportional to the amount of food ingested[76]; in humans, the stimulatory effect of fat is higher than that of carbohydrates[77]. Chronic exercise increases GIP levels in children and adolescents[78].

Once released, GIP is rapidly degraded by DPP-IV to an inactive truncated derivative, especially in kidney[79]. Inhibition of DPP-IV activity decreases glycemia in people with type 2 diabetes[80] and delays the appearance of diabetes in Zucker rats[81].

GIP receptor is a glycoprotein associated to a G protein which activates adenilate cyclase with the subsequent increase of cAMP, through which it exerts its insulinotropic effect[82]. The GIP-induced increase in cAMP

acts through a PKA-dependent and a PKA-independent pathway. In the latter, GEF2-Rim2 acts as a cAMP mediator, as it occurs with GLP-1[61]. It also acts opening voltage-dependent Ca^{2+} channels, thus increasing cytosolic Ca^{2+} and activating phosphatidyl inositol 3-kinase (PI3-K) and MAP kinases[83].

GIP stimulates insulin secretion, proinsulin[84], Pdx-1, GLUT2 and glucokinase gene expression[85]. It also stimulates differentiation, replication, growth and proliferation of pancreatic β-cells[86], inhibiting their apoptosis[87].

GIP presents functional extrapancreatic receptors in liver, muscle, adipose tissue, intestine, and sympathetic nervous system (SNS). Therefore, GIP inhibits hepatic glucose production[88], glucose uptake by muscle[89] and glucose transport in adipose tissue[90], fatty acid synthesis[91] and lipoprotein lipase activity in adipose tissue[92]. Local infusion of GIP (intestine) increases GLP-1 and somatostatin secretion[66].

The physiological importance of GIP activity was confirmed using mice with KO of its receptor gene. These mice develop glucose intolerance, decreased insulin secretion and are resistent to the development of obesity when they are fed a fat-rich diet[94]. On the other hand, KO of GIP receptors in ob/ob mice causes weight loss, with improved adiposity and glucose tolerance[55]. Based on this evidence, it has been postulated that people with increased GIP response are prone to develop obesity and hyperinsulinism[83].

People with type 2 diabetes develop GIP resistance; therefore, insulin secretion decreases in response to oral glucose, which affects primarily second-phase insulin secretion[64]. In view of the therapeutic use of GIP in people with type 2 diabetes and considering its short half-life, analogues with higher activity than that of the native molecule have been developed[95].

Amylin

Amylin is a 37 aminoacid peptide produced by β-cells, stored in their secretory granules together with insulin, and cosecreted in response to glucose[96]. In supraphysiological levels it promotes the development of insulin resistance[97].

Amylin participates in glucose homeostasis by two mechanisms, retarding gastric emptying in a dose-response manner[98], and suppressing glucagon secretion[99].

The deficit of amylin in diabetic patients results in an accelerated absorption of nutrients and loss of suppression of hepatic postprandial glucose production. Amylin´s analogue (pramilentide) reduces hyperglycemia after oral – but not intravenous – glucose administration, showing that its action is exerted at gastrointestinal level[100].

Leptin

Leptin is a glycosilated protein of 16 kDa and 146 aminoacids produced predominantly in adipose tissue, although low levels of expression have also been detected in hypothalamus, hypophysis, placenta, skeletal muscle, stomach epithelium, and breast[101].

Leptin circulates bound to a carrier protein[102] and its level increases as a function of the fat mass. It interacts with specific receptors[103] located in the AN, as demonstrated by the anorexigenic effect achieved by its local injection[104] and its lack of effectiveness when the AN is disrupted[105].

Despite leptin participates in diverse physiological processes, the main action is related to energy homeostasis and satiety; leptin provides information to the hypothalamus about the amount of energy stored in adipose tissue, arrests appetite, and modifies calorie consumption[106]. Ob/ob mice, which do not produce active leptin, have a 4-fold weight increase when they have free access to food intake. At clinical level, children with leptin defficiency modify their eating behaviour and develop marked obesity. Leptin administration to ob/ob mice and children[107,108] reverts weight gain, suggesting the importance of leptin´s regulatory role of food intake through satiety. However, leptin administration does not reduce adiposity in most cases of human obesity, thus suggesting the existence of leptin resistance.

Long-term weight loss programs frequently fail due to rapid weight regain. This has been partly attributed to the decreased leptin circulating levels consecutive to fat mass loss, with the subsequent decrease of satiety. In a small group of people, leptin – in amounts sufficient to achieve circulating levels of the peptide similar to those before weight loss – prevented weight recovery and preserved lean tissue mass[109]. In this context, leptin would act as a critical bond between adipose tissue and hypothalamic centers regulating energy homeostasis.

Adiponectin

Adiponectin is also known as gelatin-binding protein-28, apM1, AdipoQ, and Acrp30. It is a 244 aminoacid protein exclusively expressed in and secreted by white adipose tissue[110]. High circulating levels of the protein are present in human plasma as a polymer of 18 monomers.

Adiponectin acts as insulin sensitizer; its plasma concentration decreases in obesity and in type 2 diabetes[111].

Administration of recombinant adiponectin to rodents increases glucose uptake and fat oxidation in muscle, reduces fat acid uptake and glucose production in liver, and decreases insulin resistance[112]. In rhesus monkey, decreased circulating levels of adiponectin are associated with the development of insulin resistance and type 2 diabetes[111].

In mice, thiazolidinediones not only increase insulin sensitivity but also plasma levels and mRNA production of adiponectin[110]. At clinical level, there is a negative correlation between adiponectin and body weight, and body fat mass and insulin levels[113].

Resistin

Resistin is also known as "adipose-tissue specific factor", it is a 114 aminoacid polypeptide synthetized by adipocytes and secreted as a dimerized 94 aminoacid polypeptide[114].

Resistin levels are increased in mice with genetic obesity or diet-induced obesity, and decreased after troglitazone administration[115]. In these animals, administration of anti-resistin antibodies improved glycemia and insulin sensitivity, while administration of recombinant resistin to normal mice altered glucose tolerance and insulin action. Since these observations have not been confirmed by other researchers, the role of resistin in mice is controversial. Studies performed in human beings have not confirmed its role as insulin resistance regulator[116].

Neuropeptide Y (NPY)

This peptide is produced by neurons located in the floor of the third ventricle and acts directly at the level of the paraventricular nucleus (PVN) stimulating appetite (Figure 3). Neurons producing NPY coexpress NPY and AGRP (Agouti Gen Receptor Peptide)[117].

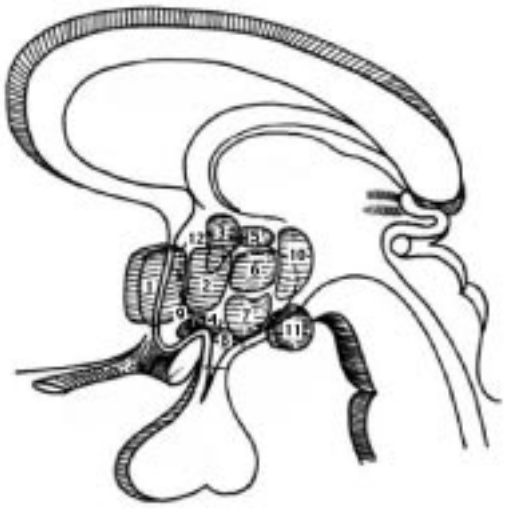

Figure 3. Hypothalamic areas and nuclei: 1. Medial and lateral preoptic area; 2. Anterior hypothalamic area; 3. Paraventricular nucleus; 4. Supraoptic nucleus; 5. Dorsal hypothalamic area; 6. Dorsal-medial nucleus; 7. Ventral medial nucleus; 8. Arcuate nucleus; 9. Suprachiasmatic nucleus; 10. Posterior hypothalamic area; 11. Mamillary bodies; 12. Lateral hypothalamic area.

NPY injection at ventricular level stimulates food intake[118], decreasing energy consumption and inducing the activity of lipogenic enzymes in liver and adipose tissue[119]. Because of these actions, continued NPY administration rapidly produces obesity[120].

During depletion of body fat deposits, there is an increased expression of NPY gene in the hypothalamus[121], thus reducing the appetite inhibitory signal of brain leptin/insulin[122]. Leptin inhibits NPY expression in the AN[123] and NPY KO reduces hyperphagia and obesity in ob/ob mice[124]. Most NPY/AGRP neurons have leptin receptors[125] which exert an inhibitory effect on these neurons[123].

During hyperphagia of insulin-deprived diabetes, there is an increase in NPY expression and secretion[126] which is blocked by insulin administration either *i.v.* or directly injected in the brain[127], suggesting a negative feedback mechanism between both hormones.

NPY receptors are coupled to a G protein and therefore their action is exerted by increasing cAMP and stimulating PKA activity[128].

Melanocortins

These are peptides derived from the hypothalamic processing of POMC, such as β-MSH[129], CRH[130] and TRH[131]. Melanocortins would act through brain MC3 and MC4 receptors[132]. Whereas the agonists of these receptors inhibit appetite, the antagonists stimulate it[133]. Mice with KO of MC4 receptors are hyperphagic and very obese[134], showing that these receptors exert a tonic signal that limits food intake and body fat mass expansion. These results have also been described in man[135].

Insulin

This hormone is produced by pancreatic β-cells from a precursor within the secretory granule by the action of proprotein convertases activated by acidification of the granule interior[43]. Both the regulatory mechanism of insulin secretion and its metabolic effects have been widely described in other reviews[136,137]; therefore we will only mention the most prominent aspects.

Insulin is a polypeptide formed by two aminoacid chains (chains A and B); both insulin synthesis and secretion are stimulated by glucose and aminoacids, but not by drugs such as sulfonilureas, which only stimulate insulin secretion[138]. Therefore, while postprandial serum insulin levels increase, they are low between meals.

As already mentioned, glucose-induced insulin secretion is higher when glucose is administered orally rather than i.v., due to the release of hormones or incretins by the bowel[139].

Gastrointestinal hormones stimulate insulin secretion acting directly at pancreatic β-cell level or indirectly through a glucose portal sensor: GLP-1[140] and GIP[84] are the main responsible for intestinal incretin effect. Conversely, other hormones such as leptin and catecholamines inhibit insulin secretion.

The interaction of insulin with specific receptors promotes a cascade of phosphorylation and dephosphorilation processes, starting with the phosphorylation of the receptor´s β-chain and followed by the insulin receptor substrate (IRS)[136]. Activation of this cascade produces a series of effects, as shown in Figure 4. All these effects convert insulin into an anabolic hormone which promotes removal of glucose and other metabolic substrates

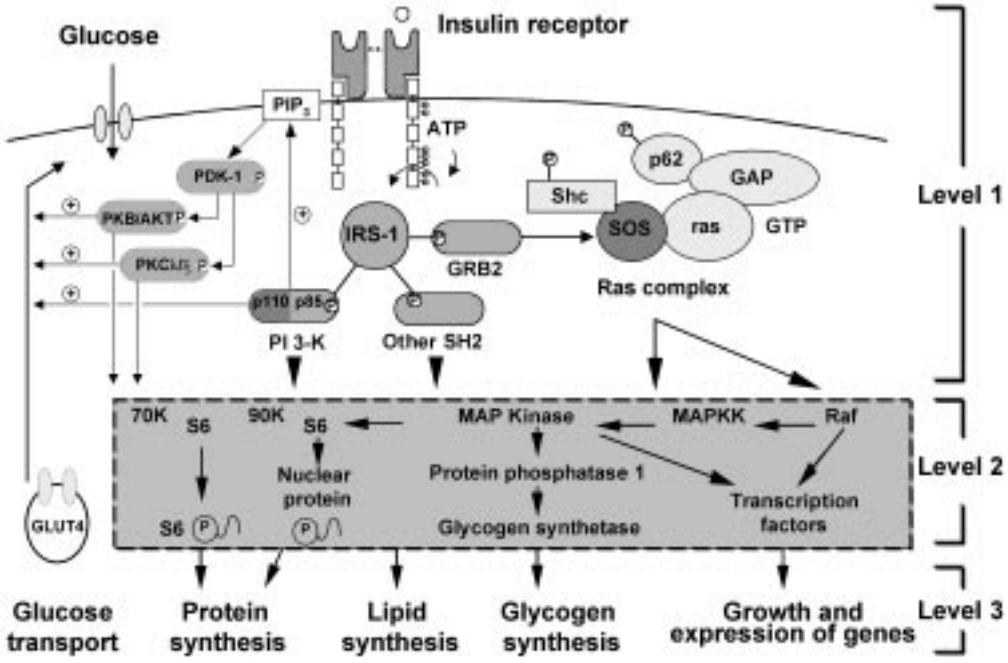

Figure 4. Intracellular insulin mediators. Interaction of insulin with its receptor, cascade of intracellular signals, and metabolic effects of insulin.

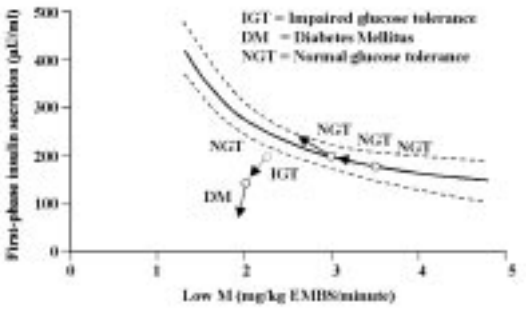

Figure 5. Relation among first peak of insulin secretion, insulin sensitivity and diabetes. Hyperbolic curve showing the relation between insulin sensitivity and insulin secretion. (Adapted from Weyer et al. 1999). Note that diabetes manifests with significantly decreased insulin secretion, with a slight change in insulin sensitivity.

from plasma and their metabolization/ deposit in liver, muscle and adipose tissue, as well as cell growth. Therefore, insulin is the main responsible for decreased postprandial glycemia[141].

Insulin secretion in response to glucose and other metabolites is biphasic, a characteristic which should be maintained to warrant insulin effective action[142].

On the other hand, β-cells couple precisely the amount of insulin released in response to a stimulus to the threshold response (sensitivity) of peripheral tissues to the hormone[143] (Figure 5). In this way, a decreased response of peripheral tissues to insulin would promote a greater release of the hormone to keep glycemia within normal range. This would suggest that glucose homeostasis will be normal if β-cells can release a sufficient amount of insulin: failure of such an adaptative capacity would cause the decrease of glucose tolerance and finally diabetes[144].

The response of peripheral tissues to insulin is modulated by different hormones: adiponectin, GLP-1 and GIP increase such response, while leptin, resistin, corticoids and somatotrofin decrease it[31,138]. Since some of these hormones are produced in adipose tissue, plasma insulin and leptin concentrations are proportional to adiposity[145]. On the other hand, both hormones interact with specific receptors located in the AN, reducing food intake and body weight in a dose-dependent manner[125] (Table I).

Regulatory Center of Appetite and Energy Balance

The hypothalamus is the main center regulating food intake and body weight, with the ventromedial nucleus (VMN) as the satiety center and the lateral nucleus (LN) as the hunger center[130]. However, the concept of specific brain centers for the control of food intake and body weight has been replaced by "discrete neuronal pathways responding in an integrated way to stimuli related to changes in fuel stores"[146].

Translation of Adipocyte Signals Into Neuronal Responses

The AN is located in the floor of the third ventricle and its neurons coexpress NPY and AGRP[117]. There are also other neurons in the AN containing POMC and CART (Cocaine- and Amphetamine-Regulated Transcript)[147] which exert an opposite action to that of NPY and AGRP on appetite, suggesting that circuits originated in this brain area play a key role on energy homeostasis.

Leptin acts through the AN as shown by the anorexigenic effect achieved by its local injection[104] and its lack of effectiveness in case of lesion on the AN[105]. Most NPY/AGRP and POMC/CART neurons have leptin receptors[125] and both cellular types are regulated by leptin in different ways: NPY/AGRP are inhibited and POMC/CART are stimulated[123,117], thus explaining their anorexigenic effect.

The AN also has insulin receptors[148]; decreased insulin and leptin levels inhibit

Table I. Peptides involved in the control of energy homeostasis.

Orexigen	*Anorexigen*
AGRP	α-MSH
Ghrelin	CART
Noradrenaline	CCK
NPY	GIP
Orexin A and B	GLP1
	Insulin
	Leptin
	OXM
	PYY
	Serotonine

Peptides were grouped according to their effect upon appetite.

POMC[149] and CART[150] expression in the AN, whereas their administration prevents it. On the other hand, a 5% increase of body weight in rats causes a 3-fold increase in POMC mRNA in the AN[151]. In conclusion, insulin and leptin messages are transformed into neuronal responses in the AN, confirming that energy homeostasis involves integrated and redundant pathways rather than a discrete group of interconnected neurons[31].

Model of Second-Order Signals

The hyopothalamus is formed by the paraventricular nucleus (PVN), the *zona incerta*, the perifornical region (PFR) and the LN, and is richly innervated by neurons from NPY/AGRP and POMC/CART cells[152]. Stimulation of the PVN inhibits food intake, whereas stimulation of the lateral nucleus (LN) stimulates it[130]. On the other hand, a PVN lesion causes hyperphagia and obesity, while a LN lesion causes anorexy and weight loss[130]. Hypocretins 1 and 2 or orexins A and B are two peptides exclusively expressed at the LN which stimulate appetite[153].

Various neurons of the PVN, PFR and LN project to the AN generating a bidirectional information flux. Therefore, these nuclei would actively modify the information received, rather than being passive receptors of information from the AN.

Mechanism of Action of the Regulatory System of Energy Homeostasis. Satiety Signals and Control of Food Intake

The frequency and amount of food must be regulated to achieve energy homeostasis. The major determinant of food size is the beginning of satiety, generated by neurohumoral stimuli promoting intake interruption. The beginning of meals is modified by external and internal factors (emotions, time of the day, availability and palatability), resulting in a biological process less controlled than satiety[154].

During the course of a meal, satiety signals are transmitted by afferent fibers from the vagus nerve and the spine coming from the high part of the gastrointestinal tract[155] to the nucleus of the solitary tract. This caudal area of the brain stem integrates sensory information coming from the gastrointestinal tract and gustatory information from the mouth[156].

Satiety signals coming to the solitary tract nucleus originate during food intake by mechanical or chemical stimuli of the stomach and small bowel, metabolites produced by the liver[157], and hormones released by neroendocrine secretory cells of the intestine in response to nutrients[158]. Therefore, the control of the end of intake involves brain areas independent of the hypothalamus influence. The leptin potentiation of the activatory effect of CCK on neurons from the solitary tract nucleus shows that the signals involved in energy homeostasis modulate the response of these neurons to satiety signals[159]. Therefore, the nucleus of the solitary tract or other areas of the brain stem as the AN, contain leptin-responsive neurons which through ascending projections towards key sites of the brain-stem contribute to adapt food intake to changes in body fat content.

Monoaminergic Neurotransmitters and Food Intake

Noradrenaline is synthetized at the dorsal nucleus of the vagus nerve and the locus ceruleus; these areas project downstream towards the stem and to the rostral hypothalamus and the brain cortex. In some of these neurons, including those projecting to the PVN, noradrenaline colocalizes with NPY. As it occurs with NPY, noradrenaline injected in the PVN increases food intake, and repeated injections may cause a significant increase in body weight[160].

Ob/ob mice present high noradrenaline levels in the PNV[161], indicating that leptin would inhibit noradrenaline release at the terminals in this brain area. Therefore, increased noradrenaline content in the PVN as well as in other hypothalamic areas would contribute to hyperphagia induced by a leptin deficit.

Dopamine

Pharmacologic[162] or gene[163] depletion of dopamine synthesis markedly modify food intake. Apparently, such decrease would contribute to the hyperphagia consecutive to leptin deficit[164].

Serotonin

Drugs such as dexfluoramine and sibutramine increase the signal of serotonin receptor and decrease appetite, while antagonists produce the opposite effect[165]. Serotonin is in-

volved in this effect and the fact that mice with KO of serotonin receptor increase food intake and body weight confirms the inhibitory effect of appetite on this monoamine[166].

Food Composition, Gastric Emptying, Digestive Enzymes and Postprandial Glycemia

Postprandial glycemic excursion depends on food composition, velocity of gastric emptying, food digestion in the intestinal lumen and removal of blood glucose: glucose is metabolized by insulin-dependent (liver, muscle, adipocytes) and independent (blood cells and nervous) tissues.

Jenkins was the first to describe variability in glycemic excursion following the intake of the same amount of carbohydrates, giving rise to the concept of glycemic index[167]. Therefore, the nature and composition of food ingested modify the amplitude of postprandial glycemic excursion.

The velocity of gastric emptying modifies glycemia, as can be seen in gastrectomized people[141]. Although it has been described that hyperglycemia retards gastric emptying in people with and without diabetes[168], its effect would be minimal in the range of glycemic variations observed in clinical practice.

Digestibility of food at intestinal level may be reduced in the presence of inhibitors such as pectin and phytates, while other substrates such as tanine reduce the action of enzymes like pancreatic amylase, thus decreasing the amplitude of glycemic excursion[141]. The inhibition of pancreatic α-amilase and intestinal α-glucosidase reduce glucose flow to blood, and could thus decrease the magnitude of postprandial hyperglycemia[169].

In the period between meals, glycemia remains within normal range because the liver produces and releases glucose as a function of the demand of peripheral tissues. Food intake causes a different situation, since intestinal absorption provides a new source of glucose: the liver supresses its production[170], keeps around 33% of portal glucose[171], and the rest is used by peripheral tissues.

The glucose absorbed and the gastrointestinal hormones released to the intestine (mainly GLP-1 and GIP – *see previous description*) in response to food intake, stimulate the biphasic secretion of insulin and inhibit glucagon secretion by 20-30%[172].

The combination of hyperglycemia, hyperinsulinemia and decreased glucagon secretion causes a 75% decrease in hepatic glucose production[173] and 25% stimulation in the uptake of glucose by splanchnic tissue (intestine and liver). In liver, glucose is stored as glycogen[141].

Glucose consumption by muscle tissue is about 25-56%[173] of the glucose entering the general circulation by the suprahepatic vein, from which 50% is oxidized, 35% is stored and 15% is metabolized and releases as lactate[174]. Adipose tissue and non-insulin-dependent tissues use the remaining glucose: one third is oxidized and the other two thirds are stored as glycogen and triglycerides[174]. These data show that the amount of glucose taken up by liver and muscle through insulin action is similar to that absorbed, preventing postprandial glycemic fluctuations far above the physiological values.

In people with type 2 diabetes, the effect of incretins is altered[69]: GLP-1 secretion is markedly decreased in the postprandial period[69] and although GIP secretion is almost normal, its effect is decreased (resistence to the insulinotropic action of GIP)[175]. The lack of response to GIP results in decreased second phase insulin secretion. Since such alteration is also present in first degree relatives of people with diabetes, it was initially suggested that it was genetically determined[176]. However, it has been recently reported that the defect manifests not only in people with type 2 diabetes but also with phenotypes of different etiology, such as type 1 diabetes, LADA, post pancreatitis diabetes, lean type 2 diabetes and MODY 3[64,177], and it would be subsequent to a GIP post-receptor defect induced by diabetes dysmetabolism.

One of the characteristics of people with impaired glucose tolerance (IGT) or diabetes is the early and progressive loss of first-phase insulin secretion[142,144,178], resulting in a decreased inhibitory effect of insulin upon glucagon secretion[141,179] and decreased free fatty acids release[178,180]. These promote insulin resistance, with the consequent excess hepatic glucose production and release, reaching 2-fold values as compared to people without diabetes[177]. Glucagon suppresion may inhibit hepatic glucose production and reduce the amplitude of postprandial gycemic excursion[178] even in people with type 2 diabetes and impaired insulin secretion.

In conclusion, the data analyzed show that the control of energy homeostasis and postpran-

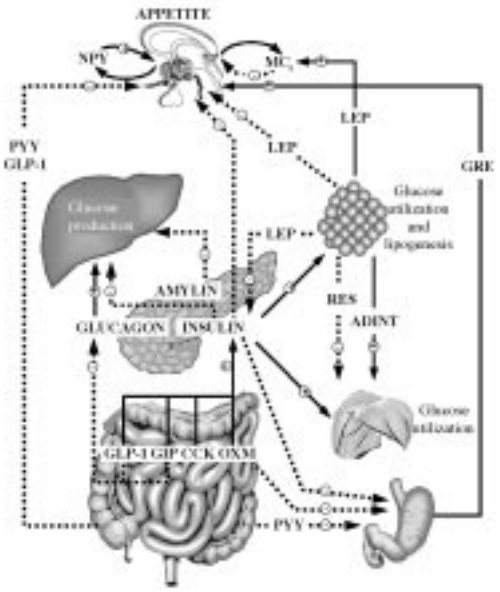

Figure 6. Interaction of messengers in the regulation of glucose homeostasis and energy balance. Interaction of the different hormones with the endocrine pancreas and the SNS to regulate glycemic homeostasis and energy balance. Food intake triggers the secretion of gastrointestinal hormones that modulate insulin secretion (immediate regulation of postprandial glycemia) and interact at the SNS to modulate appetite sensation and satiety (slow regulation of energy balance).

dial blood glucose levels is multifactorial, with the participation of exogenous and endogenous factors, as shown in Figure 6. The former include the quantity and quality of food intake while the latter involve the balance of glucose intestinal absorption (postprandial period), glucose production and release by the liver and its consumption by peripheral tissues. Adequate secretion and peripheral metabolic effects of insulin play a key role in the control of both processes. Insulin secretion is controlled by the level of circulating substrates as well as by gastrointestinal hormones. The mechanism for the immediate control of blood glucose levels is modulated by energy homeostasis, with the participation of the above mentioned hormones and others produced at the classical endocrine system and adipose tissue, whose actions integrate at the central nervous system. The alteration of such delicate mechanism of control causes diseases such as diabetes, thus, identification of the multiple components of this mechanism and comprehension of its normal function would facilitate the selection of effective strategies for diabetes prevention and treatment.

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