Molecular Neurobiology (2019) 56:769–796
https://doi.org/10.1007/s12035-018-1099-4

Brain Glucose-Sensing Mechanism and Energy Homeostasis

A. J. López-Gambero[1,2] · F. Martínez[1] · K. Salazar[1] · M. Cifuentes[2] · F. Nualart[1,3]

Received: 30 January 2018 / Accepted: 25 April 2018 / Published online: 24 May 2018

Abstract

The metabolic and energy state of the organism depends largely on the availability of substrates, such as glucose for ATP production, necessary for maintaining physiological functions. Deregulation in glucose levels leads to the appearance of pathological signs that result in failures in the cardiovascular system and various diseases, such as diabetes, obesity, nephropathy, and neuropathy. Particularly, the brain relies on glucose as fuel for the normal development of neuronal activity. Regions adjacent to the cerebral ventricles, such as the hypothalamus and brainstem, exercise central control in energy homeostasis. These centers house nuclei of neurons whose excitatory activity is sensitive to changes in glucose levels. Determining the different detection mechanisms, the phenotype of neurosecretion, and neural connections involving glucose-sensitive neurons is essential to understanding the response to hypoglycemia through modulation of food intake, thermogenesis, and activation of sympathetic and parasympathetic branches, inducing glucagon and epinephrine secretion and other hypothalamic-pituitary axis-dependent counterregulatory hormones, such as glucocorticoids and growth hormone. The aim of this review focuses on integrating the current understanding of various glucose-sensing mechanisms described in the brain, thereby establishing a relationship between neuroanatomy and control of physiological processes involved in both metabolic and energy balance. This will advance the understanding of increasingly prevalent diseases in the modern world, especially diabetes, and emphasize patterns that regulate and stimulate intake, thermogenesis, and the overall synergistic effect of the neuroendocrine system.

Keywords Glucose-sensing · Hypothalamus · Brain · Neurons · glia · Tanycytes · GLUT · ventricles · median eminence, astrocytes

Abbreviations

2-DG	2-Deoxyglucose
5-TG	5-Thioglucose
ACTH	Adrenocorticotropic hormone
ADP	Adenosine diphosphate
AgRP	Agouti-related Protein
AICAR	5-Aminoimidazole-4-carboxamide ribonucleotide
AMP	Adenosine monophosphate
AMPK	AMP-activated protein kinase
ANS	Autonomic nervous system
AP	Area postrema
ARC	Arcuate nucleus
ATP	Adenosine triphosphate
BAT	Brown adipose tissue
BDNF	Brain-derived neurotrophic factor
CART	Cocaine- and anfetamine-regulated transcript peptide
CFTR	Cystic fibrosis transmembrane regulator
cGMP	Guanosine monophosphate
CRH	Corticotropin-releasing hormone
CSF	Cerebrospinal fluid
Cx43-HC	Connexin 43 hemichannel
DBI	Diazepam-binding inhibitor
DMN	Dorsomedial nucleus of the hypothalamus
DMV	Dorsal motor nucleus of the Vagus Nerve
ES	Endocrine system

✉ M. Cifuentes
mcifuentes@uma.cl

✉ F. Nualart
frnualart@udec.cl

[1] Laboratory of Neurobiology and Stem Cells NeuroCellT, Department of Cellular Biology, Center for Advanced Microscopy CMA BIO BIO, Faculty of Biological Sciences, University of Concepcion, Concepcion, Chile

[2] Department of Cell Biology, Genetics and Physiology, University of Malaga, IBIMA, BIONAND, Andalusian Center for Nanomedicine and Biotechnology and Networking Research Center on Bioengineering, Biomaterials and Nanomedicine, Málaga, Spain

[3] Departamento de Biología Celular, Facultad de Ciencias Biológicas, Universidad de Concepción, Casilla 160-C, Concepción, Chile

F-2,6-P	Fructose-2,6-bisphosphate
F-6-P	Fructose 6-phosphate
G-6-P	Glucose 6-phosphate
GABA	γ-Aminobutyric acid
GE	Glucose-excited
GH	Growth hormone
GI	Glucose-inhibited
GK	Glucokinase
GLUT	Glucose transporter
ICV	Intracerebroventricular
IP3	Inositol triphosphate
IP_3R3	IP3-activated Ca^{2+} channel
K2P	Tow-pore domain K^+ channel
K_{ATP}	ATP-sensitive K^+ channel
LC	Locus coeruleus
LH	Lateral hypothalamus
MCH	Melanin-concentrating hormone
MCHR	MCH receptor
MCR	Melanocortin receptor
MCT	Monocarboxylate transporter
ME	Median eminence
MSH	Melanocyte-stimulating hormone
nNOS	Neuronal nitric oxide synthase
NO	Nitric oxide
NPY	Neuropeptide Y
NTS	Nucleus of the solitary tract
ODN	Octadecaneuropeptide
Ox	Orexin
OXT	Oxytocin
P2Y1	Purinergic receptor
PANS	Parasympathetic autonomic nervous system
PBN:	Parabrachial nucleus
PFK	Phosphofructokinase
PIP2	Phosphatidylinositol-2-phosphate
POMC	Proopiomelanocortin
PVN	Paraventricular nucleus of the hypothalamus
ROS	Reactive oxygen species
RVLM	Rostral ventrolateral region of the medulla oblongata
SANS	Sympathetic autonomic nervous system
SF1	Steroidogenic factor 1
sGC	Soluble guanylate cyclase
SGLT	Sodium-glucose cotransporter
SUR1	Sulfonyurea receptor 1
SVCT	Sodium-dependent vitamin C transporter
T1R	Taste receptor subunit
T_3	Triiodothyronine
T_4	Tetraiodothyronine
TCA	Tricarboxylic acid cycle
TRH	Thyrotropin-releasing hormone
Trpm5	Transient receptor potential cation channel subfamily M member 5
TSH	Thyroid-stimulating hormone
UCP	Uncoupling protein
VDCC	Voltage-dependent calcium channel
VGKC	Voltage-gated K^+ channels
vGLUT	Vesicular glutamate transporter
VMN	Ventromedial nucleus of the hypothalamus
Y	Neuropeptide Y receptor

Introduction

Glucose is a monosaccharide aldose carbohydrate, which corresponds to the molecular formula $C_6H_{12}O_6$, in a cyclic structure. Animals store glucose as glycogen, the major metabolite of glycolysis, gluconeogenesis and glycogenolysis metabolic pathways, for storage and production of energy as ATP.

In humans, the brain represents approximately 2% of the total body weight. Human brain cortex, the major region of mammalian brain, oxidizes and consumes glucose at a rate of approximately 0.4 μmol/min/g of tissue as determined by in vivo ^{13}C nuclear magnetic resonance (NMR) [1]. Brain glucose is necessary for the maintenance of cell activity, neurotransmitter synthesis, and nerve synapses [2, 3]. Complications, such as damage to the blood vessels, eyeballs, kidneys, and nervous system, may appear as a consequence of acutely or permanently low glucose levels (hypoglycemia) [4, 5]. Conversely, under excessively high glucose levels (hyperglycemia), microvascular complications arise, including retinopathy, nephropathy, and many forms of neuropathy, as well as macrovascular issues [6].

In order to regulate levels of blood glucose, the endocrine system (ES) is aided by the autonomic nervous system, particularly neuronal nuclei sensitive to metabolic changes. Such changes modulate complex processes, such as eating behavior and hormonal secretion of insulin and glucagon (produced in β- and α-pancreatic cells), adrenaline and cortisol (produced in the adrenal medulla and the adrenal cortex, respectively) and other hormones derived from the hypothalamic-pituitary axis, including growth hormone and thyroid hormones, T_3 y T_4, that control blood glucose levels and energy expenditure [7, 8].

Glucose-Sensing Mechanisms in the Brain

As a physiological homeostasis determinant of energy, glucose directly induces changes in the activity of pancreatic cells to modulate secretion and self-regulate plasma levels as well as in the intestinal tract, producing signals that are indicative of the metabolic state. However, most physiological processes regulated by glucose levels have a central control driven by nutrient-sensing cells [9].

The presence of glucose-sensitive neurons in the brain was first described in the 1960s, differentiating two populations of neurons: “glucose-excited” (GE) neurons and “glucose-

inhibited" (GI) neurons [10, 11], found mainly in the brain regions of the hypothalamus [12, 13] and the brainstem [14, 15]. These neurons are essential in the onset of counterregulatory response to hypoglycemia, thereby controlling activation of the sympathetic autonomic nervous system (SANS) and parasympathetic autonomic nervous system (PANS) [16]. The involvement of both regions in glycemic control and energy homeostasis will be further described in the "Energy Homeostasis and Neuroendocrine Control" section.

Glucose-Excited (GE) Neurons

Elevated glucose levels activate GE neurons, producing a seemingly inhibitory effect on the response systems to hypoglycemia [17] and activating the response to hyperglycemia primarily by stimulating the release of γ-aminobutyric acid (GABA) neurotransmitter in the ventromedial nucleus of the hypothalamus (VMN) [18] and nucleus of the solitary tract (NTS) [19] or anorexigenic neuropeptides in the arcuate nucleus (ARC) of the hypothalamus [20].

Most GE neurons display a trigger mechanism associated with metabolism and energy levels due to the entry of glucose through the glucose transporter 2 (GLUT2), initiating a signaling cascade that results in increased intracellular ATP, similar to the response of pancreatic β-cells (β-cells) in stimulating insulin secretion (at least in the canonical pathway), with which they share pathway components [21, 22]. However, glucose is also able to induce changes in the excitatory activity of GE neurons independent of intracellular metabolism through various identified mechanisms depending on sodium-dependent glucose transporters (SGLT) [23] or sweet taste receptor binding [24].

Metabolism-Dependent Glucose Sensing

GLUT2/3-GK-K_{ATP} Mechanism The main glucose-sensing mechanism described in the brain is similar to that employed by β-cells when inducing insulin secretion in response to hyperglycemia. It relies on three main components: (1) low-affinity GLUT2; (2) glucokinase (GK), which catalyzes a limiting step of glycolysis through glucose to glucose 6-phosphate (G-6-P) phosphorylation; and (3) octameric ATP-sensitive K^+ channels (KATP) formed by four sulfonylurea receptor subunits (SUR1 or SUR2) and four Kir6.1 or Kir6.2 subunits.

GLUT2 expression has been identified in several nuclei of the hypothalamus and brainstem, mostly associated with astrocytes [25] and concrete neurons in the brainstem [26], the hypothalamus [18, 22], as well as ependymal-glial α- and β-tanycytes surrounding the median eminence (ME) [27]. Of the glucose-sensing neurons in the VMN, few express GLUT2; thus, a shared role of GLUT3, with a lower Km, is assumed in glucose sensing [18]. Furthermore, GK expression is distributed in hypothalamic nuclei and brainstem neurons [28, 29], and SUR1 and Kir6.1 subunits of K_{ATP} channels are detected in glucose-sensing neurons in the VMN [30] along with SUR1 and Kir6.2 subunits of K_{ATP} channels in glucose-sensing neurons of the dorsal vagal region of the brainstem [31].

The glucose-sensing mechanism consists of glucose entry through the low-affinity GLUT2. Then, glucose is rapidly phosphorylated by GK to G-6-P, the substrate for glycolysis and subsequent tricarboxylic acid cycle (TCA), resulting in an increased ATP production. Binding of ATP to K_{ATP} channels promotes channel closure and prevents the transfer of K^+ from the cell interior to the cell exterior [21]. The intracellular accumulation of K^+ causes membrane depolarization, which triggers the opening of voltage-dependent calcium channels (VDCC), resulting in Ca^{2+} entry into the cell (Fig. 1a) [18, 29, 32].

Unlike β-cells, increased neuronal activity by K_{ATP} closure and Ca^{2+} entry does not lead to the release of insulin, but neurotransmitter secretion, mainly stimulating the secretion of GABA as seen in the VMN, which decreases the secretion of glucagon and epinephrine [33, 34]. Nevertheless, GABA is not the only neuromodulator found in GLUT2/3-GK-K_{ATP} GE neurons [20, 35].

In individuals suffering from obesity and type 2 diabetes mellitus, there is an increase in the mRNA expression of mitochondrial uncoupling protein 2 (UCP2) protein in β-cells, as well as in proopiomelanocortin (POMC)- or melatonin-concentrating hormone (MCH)-producing GE neurons in the hypothalamus [20, 35]. Overexpression of UCP2 uncouples and reduces mitochondrial ATP production [36], decreasing the rate of neuronal depolarization in response to the presence of glucose. This leads to a direct and parasympathetic-mediated loss of insulin secretion by β-cells [20, 35, 37, 38]. This effect was demonstrated through treatment of POMC neurons with the UCP2 inhibitor, genipin, producing an increase in the depolarization rate in response to glucose [20].

In addition, reactive oxygen species (ROS) may have a role in neuronal excitatory activity, given that they are able to regulate and inhibit voltage-gated K^+ channels (VGKC) [39] and K_{ATP} channels in dopaminergic neurons of the striatum [40]. UCP2 activity reduces production of ROS during mitochondrial ATP synthesis. Intracarotid injection of antimycin and rotenone, both inducers of ROS formation, produces an effect similar to hyperglycemia in hypothalamic GE neurons, causing the closure of K_{ATP} channels and subsequent parasympathetic insulin release [41].

Metabolism-Independent Glucose Sensing

Sodium-Glucose Cotransporter (SGLT) Mechanism The mere presence of glucose can produce an excitatory activity in certain neuronal populations in an ATP-independent manner.

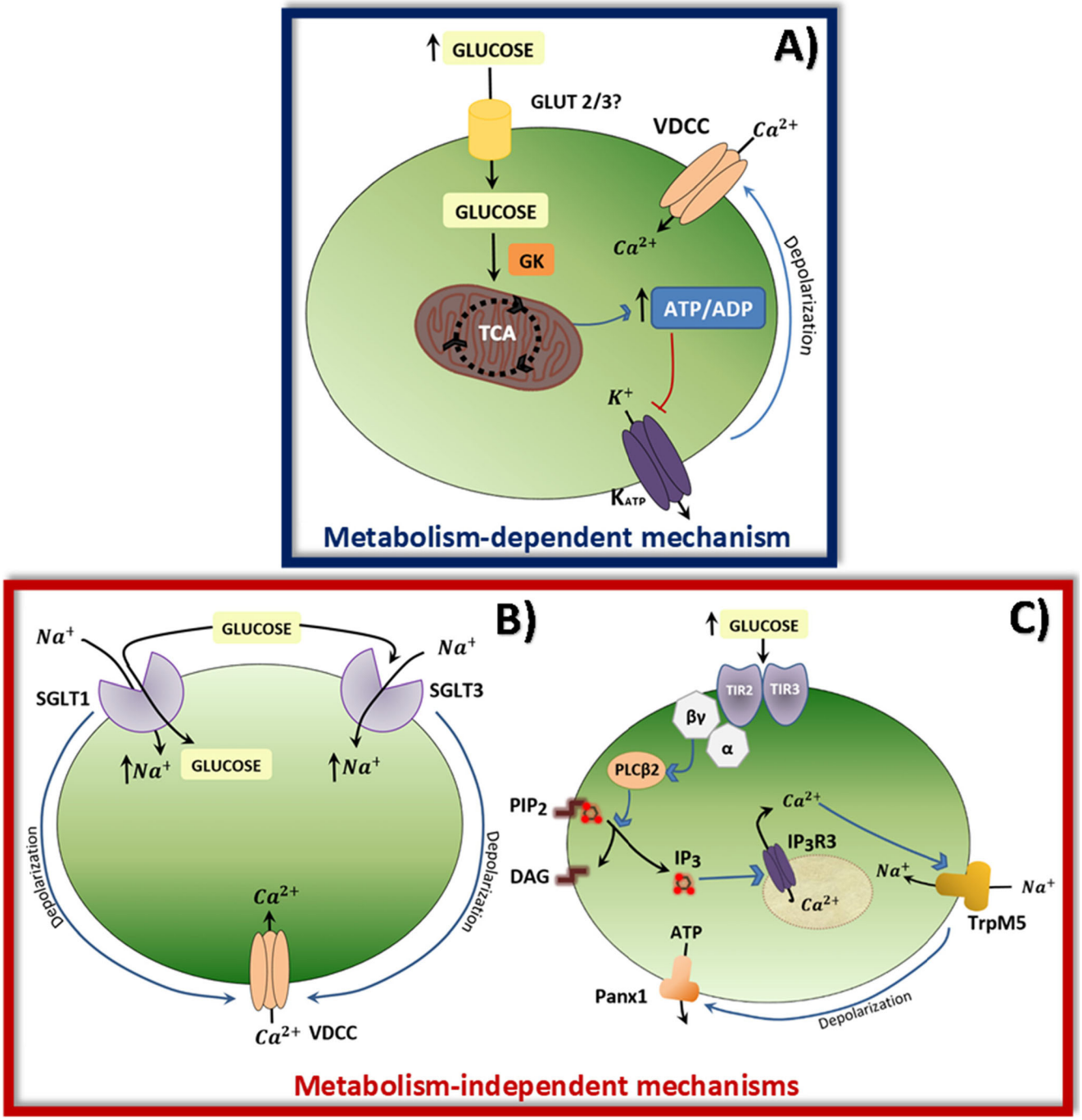

Fig. 1 Glucose-sensing mechanisms in GE neurons. **a** GLUT2/3-GK-K_{ATP} glucose-sensing mechanism. Glucose enters the cell through GLUT and is converted by GK to G-6-P (not shown), the substrate for glycolysis. The increase in ATP synthesis results in closure of K_{ATP} channels, inducing a depolarization of the membrane that allows VDCC opening, resulting in the entry of Ca^{2+} required for neurotransmitter release. **b** SGLT glucose-sensing mechanism. Glucose induces Na^+ entry into the cell through co-transport by SGLT1 or binding in the active site of SGLT3. Increased intracellular Na^+ induces membrane depolarization, activating the VDCC. **c** T1R2/3 glucose-sensing mechanism. Binding of glucose to T1R2/3 actives a G protein-coupled receptor; its βγ subunit activates PLCβ2, cleaving PIP2 membrane phospholipids into DAG and IP3, which activates the IP3R3 in the ER, inducing Ca^{2+} output into the cytoplasm. Increased Ca^{2+} levels activate the TRPM5 channel, inducing Na^+ entry. Consequently, depolarization of the membrane causes opening of Panx1 and the output of ATP, which acts as a neuromodulator. DAG diacylglycerol, GE glucose-excited, GK glucokinase, GLUT2/3 glucose transporters, IP_3 inositol triphosphate, IP_3R3 inositol triphosphate receptor, K_{ATP} ATP-sensitive K^+ channels, PanX1 ATP channel, PIP_2 phosphatidylinositol-2-phosphate, SGLT1 sodium-glucose cotransporter 1, SGLT3 sodium-glucose cotransporter 3, T1R2/3 sweet taste receptors, TCA tricarboxylic acid cycle, TrpM5 transient receptor potential cation channel subfamily M member 5, VDCC voltage-dependent Ca^{2+} channel, Bγ-α G-protein subunits

This mechanism requires the recognition of glucose by a secondary glucose transporter, which depends on the entry of one or two molecules of Na^+ through SGLT1 [42], which is present in the hypothalamus, amygdala, the cerebellum, the cortex, and the striatum [43], and SGLT3 [42], which is present in the hypothalamus and peripheral cholinergic neurons [23] (Fig. 1b).

SGLT1 couples the transport of Na^+ and glucose. Since glucose is electroneutral, Na^+ entry mediated by SGLT1 is enough to produce depolarization and increase neuronal activity. Binding of glucose (with less affinity for SGLT1) and other carbohydrates to SGLT3 results in the entry of Na^+ and membrane depolarization without cotransport of glucose, suggesting that SGLT3 acts not as a carrier but as a specialized glucose sensor.

The different affinities between SGLT3 and SGLT1 for glucose are due to a glutamine-to-glutamate change at amino acid 457 of SGLT3, directly altering the binding site for glucose [44] .

The role of SGLT in neuronal glucose sensing was demonstrated experimentally by exposing rat hypothalamic neuronal cultures to α-methyl-D-glucose pyranoside (α-MDG), a nonmetabolizable glucose analogue. Specifically, α-MDG increased the excitability of neurons that were also excited by glucose by up to 67%, producing greater neuronal excitability than glucose itself. Both responses to glucose and α-MDG were blunted by phlordizin, a competitive SGLT inhibitor. Moreover, 3-O-methyl-D-glucopyranose (3-O-MD), a nonmetabolizable glucose analogue transported by SGLT1 but not SGLT3, produced a response in almost half of the GE neurons tested, suggesting a role for SGLT1, either separately or together with SGLT3 in activation of GE neurons. However, the presence of lactate decreased neuronal response to α-MDG, possibly by a yet unknown feedback mechanism that depends on energy production, reducing SGLT transporter activity [42].

Sweet Taste Receptor (T1R2/3) Mechanism The heterodimeric sweet receptor, T1R2/3, senses the sweet taste stimulated by glucose, sucrose, and other carbohydrates. The expression of these proteins is ubiquitous in various hypothalamic nuclei, the brainstem, the hippocampus, the habenula, and choroid plexus epithelium [45, 46]. T1R2/3 is a metabotropic (G protein-coupled) receptor that can modulate neuronal activity in the presence of glucose [47] in a manner that is similar to T1R2/3 located in taste buds through Gβγ and Gα-gustducin subunits [48].

The βγ subunit induces a signaling cascade that comes together in the release of Ca^{2+} out of the endoplasmic reticulum. Released Ca^{2+} activates the Na^+ channel, TrpM5 (transient receptor potential cation channel subfamily M member 5), and promotes secretion of ATP, which interacts with purinergic receptors in neurons and glia, inducing intracellular changes [49] and activating an afferent signaling pathway [50, 51] (Fig. 1c). The α subunit promotes the expression and activity of SGLT1 in a fashion similar to gastrointestinal T1R2/3, thus enhancing glucose transport [52].

q-PCR analysis has shown that nutritional status can also induce transcriptional changes in T1R2/3 mRNA. In addition, ischemia caused by glucose restriction induces receptor expression [53]. Likewise, inhibition of the receptor results in increased T1R2/3 gene expression [46], suggesting that the presence of T1R2/3 may be essential for permanent nutritional information sensing.

Glucose-Inhibited (GI) Neurons

The activity of GI neurons increases when glucose levels fall, and is mostly associated with secretion of glutamate and noradrenaline neurotransmitters [54, 55], mainly in ventral regions of the hypothalamus, where neuronal populations with higher levels of expression of the vesicular glutamate transporter (vGLUT2) are found [55]. GI neurons are most likely shut in hyperglycemic and hyperinsulinemic conditions, modulating sympathetic and hormonal activity as counterregulatory responses to hyperglycemia [56].

Metabolism-Dependent Glucose Sensing

Na^+/K^+_{ATPase} Mechanism This pathway is based on the detection of energy products in the glycolytic cycle, such as ATP. Na^+/K^+ ATP_{ase} GI neurons have certain similarities with GLUT2-GK-K_{ATP} GE neurons, sharing the expression of GK and GLUT2 [18, 28, 29].

A decrease in extracellular glucose leads to reduced availability of ATP generated in the glycolytic cycle, necessary for maintaining the activity of the Na^+/K^+ ATP_{ase} pump [57–59]. Deactivating the pump generates an intracellular accumulation of Na^+, resulting in depolarization of the plasma membrane [60] (Fig. 2a).

Neurons, which exhibit the Na^+/K^+_{ATPase} mechanism, are distributed throughout the lateral hypothalamic region (LH) and ARC, where reduced blood glucose concentrations induces the intracellular accumulation of Na^+ and a decrease in cytoplasmic K^+ levels, likely due to inhibition of Na^+/K^+ ATP_{ase} [60, 61]. This ionic imbalance produces a net stimulatory effect in neuronal activity. The same effect is observed with the addition of ouabain, an inhibitor of Na^+/K^+ ATP_{ase}, which increased intracellular Ca^{2+} in rat ARC GI neurons, also inducing feeding behavior [61].

GLUT-AMPK-nNOS-sGC-CFTR Mechanism This mechanism is widely described in GI neurons of the VMN [62]; it depends on the entry of glucose through GLUT, ATP production in glycolysis mediated by GK and the presence of AMP-activated protein kinase (AMPK), neuronal nitric oxide

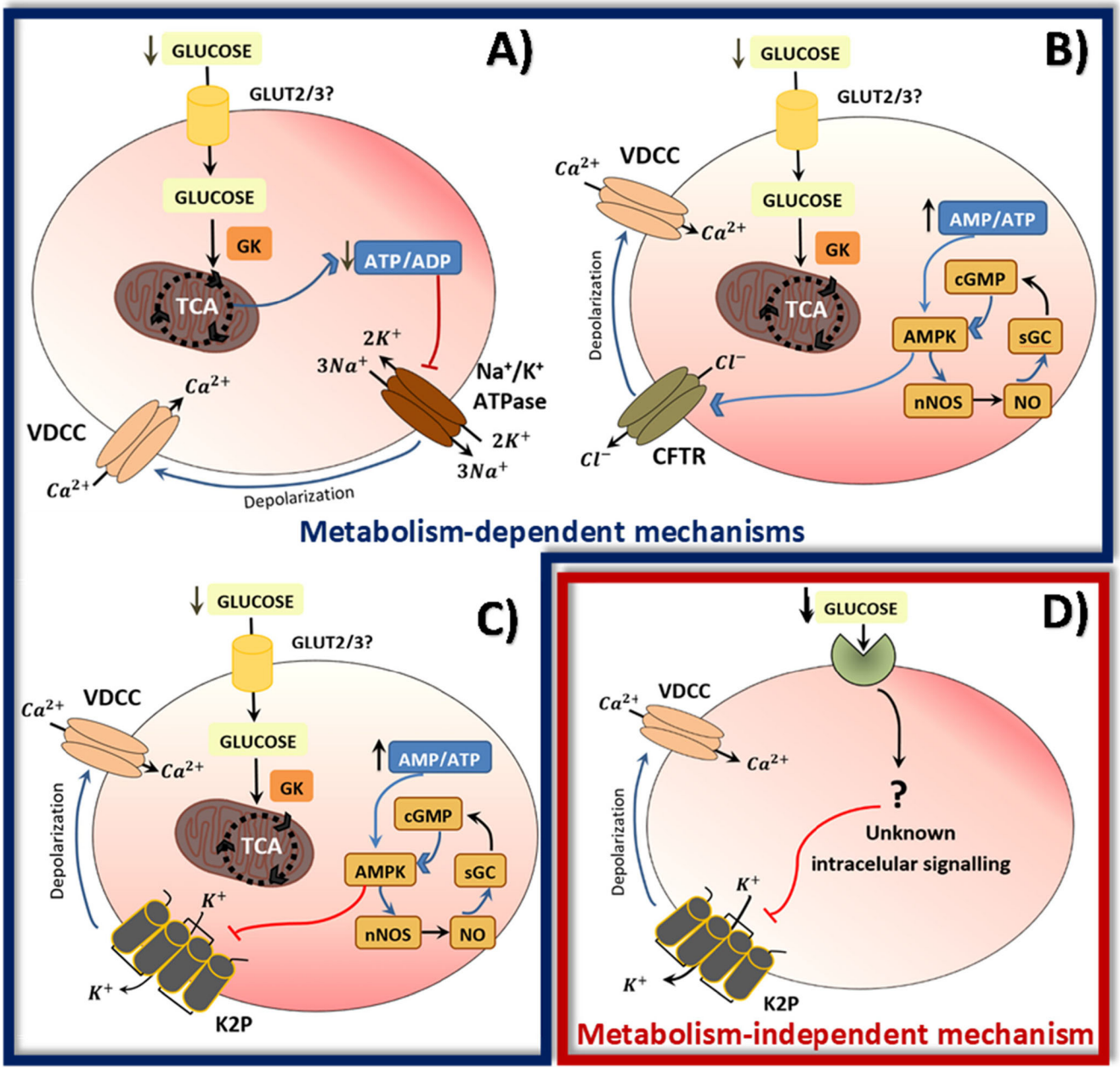

Fig. 2 Glucose-sensing mechanisms in GI neurons. **a** Na^+/K^+ ATPase glucose-sensing mechanism. A decrease in glucose leads to lower ATP production, thus inhibiting the activity of the Na^+/K^+ ATPase and resulting in membrane hyperpolarization and closure of VDCC. **b** GLUT-AMPK-nNOS-sGC-CFTR glucose-sensing mechanism. Low glucose causes an increase in the AMP/ATP ratio, which partially activates AMPK. AMPK phosphorylates nNOS, which produces NO, inducing the formation of cGMP required for full activation of AMPK. AMPK phosphorylates and induces the opening of CFTR, thus producing membrane depolarization and inducing the VDCC opening. **c** GLUT-AMPK-nNOS-sGC-CFTR glucose-sensing mechanism. Similar to the GLUT-AMPK-nNOS-sGC-CFTR pathway, reduced glucose levels induces AMPK activation, which in this case leads to the closure of K2P channels, resulting in membrane depolarization and activation of VDCCs. **d** K2P glucose-sensing mechanism. Low glucose is detected by a membrane receptor, which induces the closure of K2P channels through a metabolism-independent signal transduction cascade, increasing the membrane depolarization rate and thus, opening VDCCs. AMPK AMP-activated protein kinase, CFTR cystic fibrosis transmembrane conductance regulator (Cl^- channel), cGMP cyclic GMP, GI glucose-inhibited, GK glucokinase, GLUT2/3 glucose transporters, K2P two-pore domain K^+ channel, Na^+/K^+ ATPase ATP-dependent Na^+/K^+ pump, nNOS neuronal nitric oxide synthase, NO nitric oxide, sGC soluble guanylate cyclase, TCA tricarboxylic acid cycle, VDCC voltage-dependent Ca^{2+} channel

synthase (nNOS), soluble guanylate cyclase (sGC), and the Cl^- channel, cystic fibrosis transmembrane regulator channel (CFTR).

Glucoprivation decreases ATP synthesis, leading to an imbalance in the AMP/ATP intracellular ratio, so that the increased levels of AMP in proportion to ATP cause partial

activation of AMPK. AMP binding to the AMPKγ subunit predisposes the AMPKα subunit for phosphorylation by several kinases and later activation [63–65]. When partially activated, AMPK phosphorylates nNOS, increasing NO levels [66–68]. NO binds to sGC receptors, boosting the synthesis rate of cyclic GMP (cGMP). Higher levels of cGMP promote AMPK phosphorylation through positive feedback. When fully activated, AMPK phosphorylates and blocks CFTR channels, causing accumulation of intracellular Cl^- and, ultimately, depolarization of plasma membrane and neurotransmitter release [67].

Exposure of rat VMN GI neuronal cultures to 5-aminoimidazole-4-carboxamide ribonucleotide (AICAR), an AMPK activator, mimicked the effects of low glucose conditions, enhancing neuronal activity and NO production [69]. Moreover, the presence and activation of CFTR by AMPK has also been determined in the hypothalamus [67].

However, hypoglycemia also increases ROS levels. The combination of NO and ROS production may provoke S-nitrosylation of sGC, decreasing its activity. Recurrent hypoglycemia causes the formation of ROS, leading to a decreased sensitivity of VMN GI neurons to hypoglycemic stimulus [62, 70] (Fig. 2b).

GLUT2-AMPK-nNOS-sGC-K_{2P} Mechanism This mechanism has been described in a population of neurons found in the NTS of the brainstem also with a described role for GLUT2 [26]. A decrease in glucose is followed by AMPK activation, leading to the closure of two-pore-domain potassium channels (K_{2P}) [26, 71], encoded by genes of the KCNK subfamily, which consist of heterodimers of TWIK-related acid-sensitive K^+ channel (TASK) subunits sensitive to acidity with a TASK3 subunit [72].

As with GLUT/AMPK/nNOS/sGC/CFTR neurons, a reduction in glucose levels further decreases ATP levels, increases the AMP/ATP ratio, and activates AMPK. K_{2P} channel closure is likely induced by AMPK activity as shown in other TASK3 channels inhibited by AMPK, thereby inducing membrane depolarization [73] (Fig. 2c).

After the addition of AICAR, an AMPK activator, and oligomycin, a mitochondrial ATPase inhibitor, the same effects are observed in NTS neurons during hypoglycemia, inducing an increase in depolarization rate [74]. Characterization of K_{2P} channels as determined by current and voltage tests at different glucose concentrations was indicative of activation kinetics similar to that shown by K^+ channel leakage [26].

Metabolism-Independent Glucose Sensing

Extracellular Receptor-K_{2P} Mechanism This metabolism-independent glucose-sensing mechanism was determined within the population of orexin neurons found in the lateral hypothalamus [75]. As in GLUT2-AMPK-nNOS-sGC-K_{2P} neurons, this mechanism involves closure of K_{2P} channels (Fig. 2d). When glucose levels rise, K_{2P} remains open, maintaining a low rate of depolarization.

Exposing these neurons to halothane (1%), a specific K_{2P} activator, reproduced the effects of glucose on channels and membrane excitability, resulting in hyperpolarization [75]. Furthermore, application of glucose and ATP intracellularly did not inhibit orexin GI cells, suggesting that the activity of these neurons is dependent on extracellular glucose, but independent of the metabolic rate [75].

Channel composition was demonstrated by IC50 (inhibition of 50% of channels) analyses at a given pH, which varied between the different acid-sensitive TASK subunits. IC50 reached a pH value of 6.9, similar to that shown by TASK3 and homodimers and heterodimers containing at least one subunit of TASK3 (pH ~ 6.7) [72, 75, 76]. Channels were found to be TASK3 heterodimers, showing no effect after addition of red ruthenium, a TASK3 homodimer-specific inhibitor [72, 75]. No response of the channel to higher intracellular ATP suggests that this K2P might be unaffected by AMPK, as is also seen for TASK1, TASK3 and heteromeric TASK1/3 channels [77]. We suggest that the differences in K_{2P} sensitivity between GLUT2-AMPK-nNOS-sGC-K2P neurons and extracellular receptor-K_{2P} neurons could be due to different subunit conformation or lack of a yet-unknown intermediate between AMPK and K_{2P} activation, since phosphorylation of the channel has not been demonstrated.

In summary, the brain has developed mechanisms to detect rises and falls in glucose levels. Since it depends on glucose as fuel, it is foreseeable that more than one mechanism has been developed, either with specific functions or as redundant or overlapping systems that permit the precise control of glucose levels in the internal environment. The relationship of these mechanisms is observed in the repetition of common elements, such as APMK, GLUT, GK, or K2P. Its importance in the neuronal glucose-sensing depends on the physiological context, where glial cells may play an essential role in the metabolic coupling of glucose. These issues are discussed in the "Glial Cells as Energy Supporters," "Involvement of Metabolic State in Glucose-Sensing," "Involvement of GLUT in Glucose Sensing and Transport Kinetics," and "Possible Mechanisms of Glucose Sensing in Tanycytes" sections.

Glial Cells as Energy Supporters

Astrocytes

Astrocytes are the most abundant glial cells in the brain, present in the blood-brain barrier (BBB). Immunohistochemical analysis has identified some likely astrocyte cell bodies and processes labeled with GFAP and GLUT2 in the ARC of rat brain

[78]. In addition, a great number of gomori-positive astrocytes, a subset of astrocytes specifically abundant in the ARC, express GLUT2 as determined by immunocytochemistry [79]. Gomori staining is biased to mitochondrial-derived cytoplasmic granules, which are generated under metabolic stress conditions, likely due to high-capacity transport of large amounts of glucose via GLUT2. Immunocytochemistry and electron microscopic observations have also showed GLUT2 localization in a restricted population of astrocytes in the NTS [25].

GLUT2 knockout (glut2$^{-/-}$) mice reexpressing GLUT1 or GLUT2 in β-cells (Ripglut1; glut2$^{-/-}$ or Ripglut2; glut2$^{-/-}$) show almost normal insulin secretion but defective glucagon secretion [80]. However, reexpression of GLUT2 in astrocytes, but not neurons restored glucagon secretion in Ripglut1; glut2$^{-/-}$ mice [81]. In addition, injection of antisense oligodeoxynucleotide to GLUT2 mRNA into the ARC of Wister rats altered feeding pattern and abolished insulin response to glucose load [82]. These findings suggest that, at least certain subpopulations of astrocytes expressing GLUT2 could be involved in glucose sensing, likely along with neuronal activity either in hyperglycemic or hypoglycemic responses.

Generally, astrocytes are considered metabolic and signaling supporters of neuronal activity. Glycogen is stored mainly in astrocytes in the brain, and hypoglycemia is known to lead to glycogenolysis and astrocytic lactate production [83], which is essential to maintain neuronal activity under low-glucose conditions [84, 85].

The "astrocyte-neuron lactate shuttle" hypothesis states that under conditions of energy demand due to high neuronal activity or hypoxia, astrocytes boost nonoxidative metabolism of glucose to lactate, which is released and taken up by neurons to meet substrate requirements for aerobic glycolysis [86, 87]. Supporting this role, astrocytes express mainly lactate dehydrogenase isoform 5 (LDH5) mRNA, which favors pyruvate-to-lactate conversion, whereas expression of LDH1 mRNA, which favors pyruvate-to-lactate reaction, has been determined in neurons and in hypothalamic nuclei among other regions [88, 89]. Lactate would then be released into the extracellular space likely through monocarboxylate transporter isoform 1 (MCT1) present in astrocytes [90–93], after which it would be transferred to neurons through neuronal-specific MCT2 [91, 92, 94]. However, MCT4 is also expressed in astrocytes in the paraventricular nucleus of the hypothalamus (PVN) [92] and in the ARC [93]. Interestingly, immunocytochemistry analysis detected MCT4 in astrocytes, a different region apart from astrocytic MCT1 in the ARC [93]. MCT1 has also been detected in cultures of hypothalamic GE neurons, in which lactate induced K_{ATP} channel closure and membrane depolarization [95]. Since the ARC is a glucose-sensitive region, distribution of different isoforms of astrocytic or neuronal MCT could be linked to different metabolic coupling and sensing mechanisms.

Lactate release in vitro by astrocytes but not by neurons has been shown to be increased in higher glucose concentrations [96]. The importance of lactate supply in glucose sensing has been shown in that it induces membrane depolarization of GE but not non-GE neurons in the VMN of rat brain slices [97]; the counterregulatory response against hypoglycemia in vivo is also blunted after perfusion of lactate in the VMN [98]. Intraperitoneal injection of methionine sulfoximine (MSO), a xenobiotic amino acid that interferes with the metabolism of carbohydrates in astrocytes, leads to loss of hypothalamic GE neuronal excitability during hyperglycemia and decreased pancreatic insulin secretion [99]. This suggests that at least certain subpopulations of astroglia could be involved in glucose regulation and sensing in the brain through internalization of glucose and lactate production as a metabolic supply.

Bolanos et al. [100] described the importance of nitric oxide (NO) signaling in astrocytic lactate production in 2006. Fioramonti et al. [62] later reported NOS activity during hypoglycemia in VMN GI neurons [101]. NO increases expression of GLUT1 and GLUT3 in astrocytes and neurons, respectively [102]. Moreover, NO affects astrocyte mitochondrial respiration by inhibiting the cytochrome C chain in the electron transport chain [103], initially decreasing the formation of ATP and increasing the AMP/ATP ratio and partial activation of glial AMPK much like GI neurons with a GLUT/AMPK/sGC/NO/CFTR mechanism [66]. AMPK phosphorylates and activates phosphofructokinase 2 (PFK2), which, in turn, catalyzes the conversion of fructose-6-phosphate (F-6-P) to fructose-2,6-bisphosphate (F-2,6-P), the allosteric activator of phosphofructokinase 1 (PFK1), and a limiting step in glycolysis [66]. Anaerobic glycolysis is thus enhanced, generating ATP and lactate. This process is specific to astrocytes, whereas neurons have low expression of PFK-2 and cannot induce the formation of ATP and NO-mediated lactate [66].

The hypothesis of enhanced lactate production and release in astrocytes by VMN GI neurons proposed by Fioramonti et al. [62] could be linked to recurrent hypoglycemia and increased diabetes-associated GABAergic tone in the VMN, contributing to a defective counterregulatory response against hypoglycemia after several courses of decreased glucose [104]. The role of GABA, lactate, and K_{ATP} sensing mechanism has been assessed in vivo in the VMN of rats [104, 105], while infusion of lactate increased VMN GABA levels, which was accompanied by suppression of glucagon and epinephrine release. In addition, the MCT2 inhibitor, 4CIN, and the LDH inhibitor, OX, decreased VMN GABA levels, restoring the counterregulatory response. Moreover, the counterregulatory response was also restored following injection of the $GABA_A$ receptor antagonist, bicuculline methiodide, which decreases the postsynaptic effect of GABA, and the K_{ATP} channel opener, diazoxide, which decreased VMN GABA levels. This was linked to higher VMN GABA and lactate concentrations in recurrent hypoglycemia

and diabetic rats, both of which were decreased after OX and 4CIN treatment, restoring previously seen defective counterregulatory responses to normal [104].

Although lactate levels have not been measured during acute hypoglycemia linked to increased NO production, increased lactate levels in vivo associated with recurrent hypoglycemia in the VMN could explain defective counterregulatory responses. This would likely sustain ATP production in GE and GI neurons, thereby increasing and decreasing neuronal responses to low-glucose concentrations, respectively.

Fioramonti et al. [62] observed increased constitutive NOS activity following acute but not recurrent hypoglycemia, suggesting a link between the absence of NO production and impairment of counterregulatory responses in recurrent hypoglycemia. NO inhibition of mitochondrial activity has been described as either reversible or irreversible. Irreversible inhibition of astrocytic mitochondria could explain the lactate-enhanced production even with decreased VMN GI neuron NO production after recurrent hypoglycemia as mentioned by the group. Correlations have been reported between recurrent hypoglycemia decreasing nNOS activity, which is essential for VMN GI neurons via a GLUT/AMPK/sGC/NO/CFTR mechanism, and enhanced astrocytic lactate production, which increases activity of VMN GABAergic GE neurons via a GLUT2/3-GK-K_{ATP} mechanism**.** It has been reported that NO inhibits cytochrome c oxidase rapidly and reversibly while competing with O_2 in astrocytes [103]. However, a possible conversion of NO to peroxynitrite ($ONOO^-$) exists for irreversible damage of cytochrome c oxidase [106]. While astrocytic metabolism is robust against NO and its derivatives, neurons could suffer from damage under high concentrations of NO [106].

In vitro studies have indicated that under hypoglycemic conditions, cultured neurons have a higher survival rate when co-cultured with glial cells although it is dependent on the glycogen content, suggesting that it likely supports neuronal activity by glycolysis-derived lactate supply [107]. Along with glycolysis in astrocytes during hypoglycemia, lactate transport across the BBB is also enhanced during recurrent hypoglycemia as a result of the varying lactate concentration gradient between the plasma and brain [108]. Contribution of either astrocyte-to-neuron lactate shuttle or brain lactate uptake could maintain neuronal activity but impair the counterregulatory responses during recurrent hypoglycemia and diabetes.

A proposed model for ventromedial hypothalamus glucose sensing in recurrent hypoglycemia is shown (Fig. 3), encompassing GI neuron-lactate-GE neuron metabolic coupling and brain lactate uptake. However, the interplay of NO with increased lactate and reduced GABAergic tone and counterregulatory responses should still be evaluated.

Tanycytes

Tanycytes are specialized ependymal glia located in the lateral and lower areas of the third ventricle; their apical parts face the ventricular lumen. Similar to astrocytes, tanycytes express MCT1, MCT4 [93], GLUT2 [27, 78], and GK [109, 110].

There is a proportional relationship between plasma and cerebrospinal fluid (CSF) glucose levels [111–113]. CSF is the only cerebral fluid that shows elevated glucose concentrations of about 15 mM in hyperglycemic conditions. Due to their location, tanycytes may play a key role in signal transduction related to changes in glucose concentration in CSF, forming a link between CSF and glucose-sensitive neurons located in the hypothalamus, where there is no direct access to the CSF. Tanycytes are classified as α tanycytes or β tanycytes according to their position around the endothelial wall of the third ventricle [114]. α Tanycytes form an interface between the CSF and hypothalamic nuclei and come in two types: (1) α1 tanycytes send projections to the VMN and dorsomedial nucleus (DMN), and (2) α2 tanycytes send projections into the ARC. β Tnycytes are located on the floor of the third ventricle, and there are also two types: (1) β1 tanycytes, which send projections to the ARC and the ME of the hypothalamus, and (2) β2 tanycytes, which send their projections closely in the ME, accessing fenestrated capillaries of the hypothalamic-pituitary axis [114, 115].

Both α and β tanycytes possess functional GLUT1 and GLUT2 [27]. Interestingly, GLUT2 localization is polarized to the apical membrane in tanycytes in close contact to the CSF in a similar manner as GLUT1 specifically in β tanycytes; GLUT1 is also localized to cellular processes [27]. Polarization is also observed for MCT1 and MCT4 expression in tanycytes [93]. Multilabeling immunohistochemistry showed MCT1 in the ventricular cellular membranes of α tanycytes and, more interestingly, in end-feet processes contacting the endothelial cells of blood vessels. In contrast, predominantly β1 tanycytes showed MCT1 polarization to the apical membrane and cellular processes contacting ARC neurons, blood vessels, and external region of the brain [93]. However, MCT4 expression was predominantly detected in a subpopulation of β tanycytes, associated with long cellular processes contacting the lateral region of the ARC and with anti-GFAP staining (β1 dorsal or β1d tanycytes) and less significantly associated with short cellular processes contacting the periventricular region of the ARC and with anti-vimentin staining (termed β1 ventricular or β1v tanycytes) [93].

Tanycytic GLUT localization at the vasculature suggests its participation in incorporating glucose from the CSF. MCT2 expression has been observed in neuropeptide Y (NPY)/agouti-related protein (AgRP) GI neurons in the same region as MCT1-expressing β1v tanycytes [93, 116] and also in POMC/cocaine-amphetamine related transcript (CART) GE

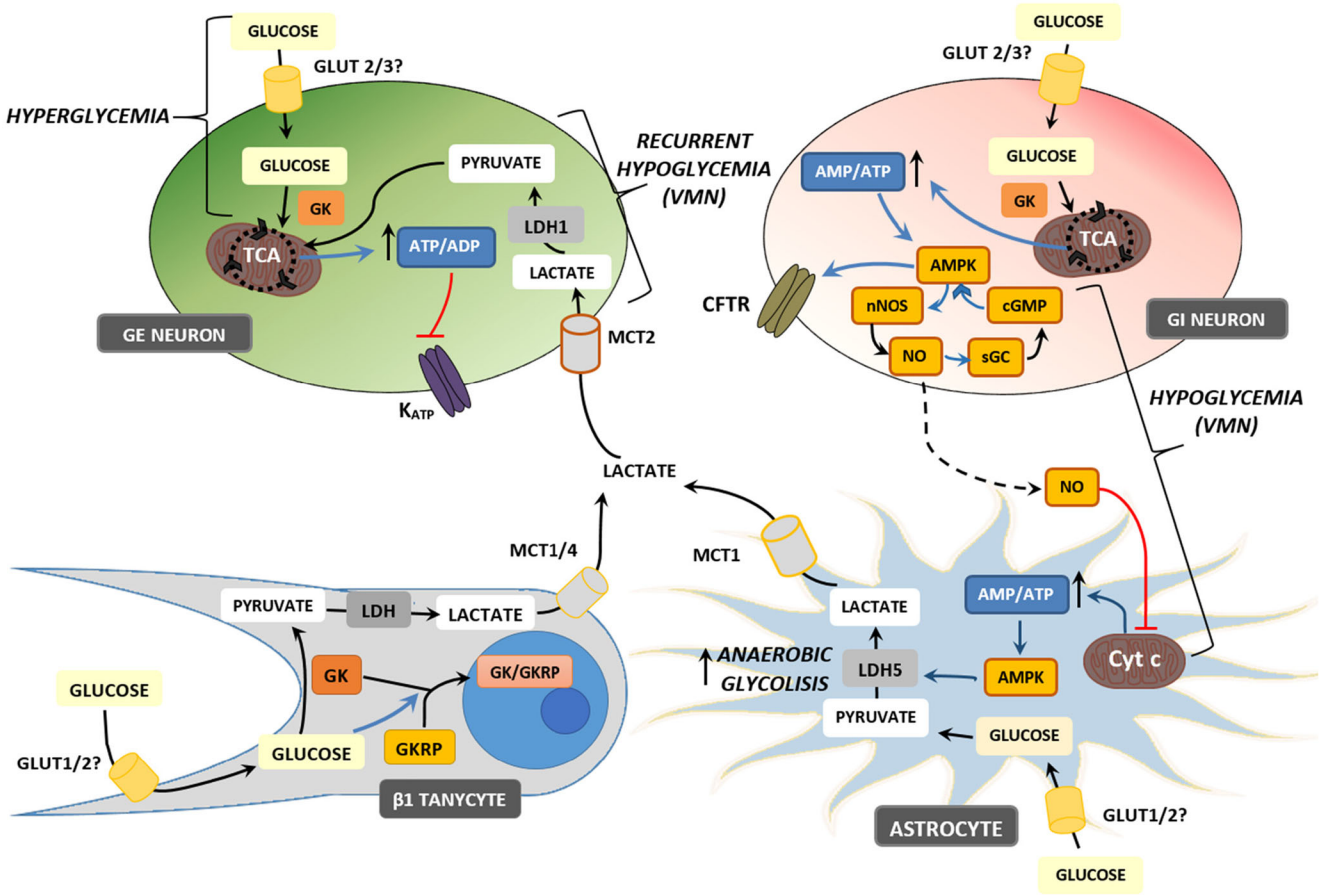

Fig. 3 Representation of astrocyte and tanycyte lactate shuttle to neurons. Both astrocytes and tanycytes uptake and metabolize glucose to lactate, which is released by MCT1 or MCT4, and internalized by ventromedial and arcuate nuclei neurons through MCT2. In high-glucose conditions, GK is internalized by GKRP in the nuclei of tanycytes. Hypoglycemia leads to NO production in GI neurons in the ventromedial hypothalamus. NO inhibits Cyt c in astrocytes, producing an increase in AMP and activating AMPK, which leads to higher anaerobic glycolysis and lactate production and release through MCT1. In recurrent hypoglycemia, high accumulation of lactate increases ATP synthesis as in hyperglycemia, enhancing GE neuronal activity and consequent GABA release, inhibiting the counterregulatory response to hypoglycemia. AMPK AMP-activated protein kinase, CFTR cystic fibrosis transmembrane conductance regulator (Cl^- channel), cGMP cyclic GMP, Cyt c cytochrome c, GE glucose-excited, GI glucose-inhibited, GK glucokinase, GKRP glucokinase regulatory protein, GLUT2/3 glucose transporters, LDH lactate dehydrogenase, MCT1/4 monocarboxylate transporter, nNOS neuronal nitric oxide synthase, NO nitric oxide, sGC soluble guanylate cyclase, and TCA tricarboxylic acid cycle. Scheme based on "Fioramonti et al. (2011). Antioxidants and signaling, 14 (3), 505-517"

neurons close to MCT4-expressing β1d tanycytes [93, 116]. Thus, it is likely that a tanycyte-neuron lactate supply occurs. Hence, glucose may be directly incorporated by tanycytes and released as lactate, which could also induce either activation or inhibition of GE and GI neurons through metabolism-dependent glucose-sensing mechanisms (Fig. 3).

The role of tanycytes as metabolic support in the response of glucose-sensitive neurons has been reproduced by injection of lactate into the third ventricle, a region with the greatest presence of tanycytes. Lactate mimicked the effects of glucose supply by decreasing feeding and other hyperglycemia response mechanisms [117].

Specific expression of GK in tanycytes has also been evaluated. Confocal microscopy, immunocytochemical detection and quantitative immunoreaction analysis revealed that GK is expressed in both the nuclei and cytoplasm of β1 tanycytes [109]. In another study, immunocytochemistry revealed the presence of GK in tanycyte cultures, showing no expression of GFAP, a marker expressed by astrocytes and α tanycytes [110, 118]. RT-PCR and Western blot analyses also confirmed the presence of liver-isoform of glucokinase regulatory protein (GKRP) in cultured tanycytes [110]. Immunocytochemistry and immunoblot analysis of nuclear extracts also showed higher GK and GKRP nuclear localization after increasing doses of glucose [110]. Therefore, it has been proposed that β1 tanycytes can regulate its response to increased glucose concentrations through the nuclear translocation of GK, mediated by GKRP, which could lead to a decrease in glucose metabolism and lactate production in an opposite manner to the hepatic GK and GKRP switch, whose translocation from the cytoplasm to the nucleus is induced in hypoglycemia [110].

Tanycytes also express the deiodinase II enzyme (D2), which converts T_4 into T_3. D2 activity is enhanced during fasting, increasing T3 production. T3 increases hypothalamic UCP2 mRNA in vivo. Interestingly, T3 induces mitochondrial uncoupling in NPY/AgRP neurons in the ARC, and lacking of UCP2 blunts the response of NPY/AgRP neurons to fasting. Also, electron microscopy showed that D2-positive tanycytes make contact with NPY/AgRP neurons, suggesting that tanycytes could control GI neuronal activation during hypoglycemia by producing T3 [119]. Since UCP2 mRNA was elevated in the hypothalamus, it is likely that D2-expressing tanycytes could also contact POMC GE neurons in the ARC, and expression of UCP2 diminishes ROS production, likely decreasing their activity [20].

Involvement of Metabolic State in Glucose-Sensing

The fact that there are two mechanisms to sense blood glucose variations (one based on glucose internalization and metabolism by altering the level of ATP and the other mechanism capable of modulating excitatory activity solely due to the presence of glucose as detected by extracellular proteins) suggests that distinct populations of glucose-sensitive neurons may have a different and synergistic role in controlling glycemia.

Taking account of the above, metabolism-dependent neurons may be the main cell metabolic state sensors, while their depolarization rate varies gradually according to an increase or progressive decrease in intracellular ATP or AMP levels. These cells have the energy support of glial cells, capable of generating lactate from glycogen reserves in situations of glucoprivation. Lactate is internalized by neurons as an energy source by holding mitochondrial ATP production, at least in a short period of time. As suggested by González et al. [120], it is likely that the primary role of these neurons is to act as an emergency response system, detecting major changes in the level of energy substrates available, preferably glucose as main neuronal fuel. Importantly, these neurons are capable of responding to small changes that compromise the cellular metabolic state, such as reduced expression of glucose transporters, which require larger amounts of glucose for cellular demand. It has also been observed that inactivation of *Glut2* gene expression in the nervous system was associated with hypoinsulinemia, lower β-cell proliferation rates, reduced sympathetic and parasympathetic nerve response to glucose and reduced parasympathetic nerve activity in the basal state during postnatal development in mice. Thus, GLUT2 metabolism-dependent glucose sensing in the nervous system is necessary to establish parasympathetic nervous stimulation, which is essential for normal pancreatic activity and β-cell mass [121]. However, as previously stated, recurrent hypoglycemia has also been linked to increased brain lactate, which could serve as a metabolic supply for hypothalamic GE neurons with a metabolism-dependent glucose-sensing mechanism, therefore increasing their activity and decreasing the counterregulatory response [104]. In the same way, GK mRNA is increased in the VMN after insulin-induced hypoglycemia, decreasing responsiveness of ARC neurons to low-glucose conditions [122]. Inhibition of GK also decreases and increases VMN GE and GI neurons responsiveness, respectively [29]. This issue will be discussed further in the "Energy Homeostasis and Neuroendocrine Control" section.

On the other hand, metabolism-independent neurons, especially those not requiring glucose transport of GK activity, could be postulated as the main sensors of extracellular glucose fluctuations, while they are able to generate a response against a milder hypoglycemia, maintaining normal cell activity by using lactate as an energy source. They would also have an advantageous position with respect to the metabolism-dependent neurons in certain pathological conditions, being able to respond to recurrent hypoglycemia (likely in diabetic patients) as mentioned before. Moreover, González et al. [120] has described a certain subpopulation of GI orexin neurons that have an adaptive non-metabolic threshold for glucose, which could allow the brain to sense changes in glucose levels and avoid slight changes in glucose to induce neuronal firing [123].

Involvement of GLUT in Glucose Sensing and Transport Kinetics

Despite the current study to determine the composition, dynamics, and kinetics of brain glucose transport, there are still several issues that need to be resolved. One of them is the aforementioned possible dual role between GLUT2 and GLUT3 in neuronal glucose-sensing mechanisms.

GLUT2 expression has been determined in different hypothalamic regions, being present along with other components of the glucose-sensing mechanisms proposed (i.e., GK or K_{ATP}). Nevertheless, GLUT2 possesses high capacity kinetics for glucose transport, but with low affinity; a Km value between 11 and 17 mM as determined with 2-DG has been observed in *Xenopus laevis* oocytes [124, 125].

As a component of the pancreatic glucose-sensing mechanism, it is plausible to consider that the neuronal system behaves similarly to promote insulin secretion, preceded by a reduction in glucose levels.

Even so, it should be noted that interstitial glucose levels in the brain correspond with approximately 30% of blood glucose concentration. Glucose-sensing neurons are often exposed to interstitial glucose levels, varying between 0.7 mM in fasting conditions and 1.5–2.5 mM following feeding in rats, although differences in threshold glucose levels could be due to the use of different strains of rats (Sprague-Dawley vs. Wistar) or anesthesia [126, 127]. Nevertheless, interstitial glucose reaches values of between 0.16 mM in insulin-

induced hypoglycemia and 4.6 mM following intraperitoneal glucose injection reproducing hyperglycemia [127].

The main transporters in the brain include GLUT1, expressed mostly in microvascular endothelial and glial cells [93, 128–130], and GLUT3, found almost exclusively in neurons [129–131]. *Xenopus* oocytes expressing GLUT3 and GLUT1 show Km values of 1.4–1.8 mM and 6.9 mM, respectively, for 2-DG, confirming that GLUT1 has a more favorable kinetic profile for glucose entry [124, 125].

The role of GLUT2 expression in glucose-sensitive neurons in the hypothalamus is currently unclear, keeping in mind that glucose levels in the CSF would never be high enough to be transported by GLUT2 to the interior of the cell.

One option that would shed light on this issue could be that these neurons respond in cases where the derivative hyperglycemia incurred with increased permeability of the BBB, allowing diffusion of higher levels of glucose. Most experiments associated with central detection of glucose in vitro or glucose injections in vivo, as well as analogs thereof, are produced with glucose concentrations higher than those presupposed in the CSF, waiting for a response of increased activity. GLUT3 is, therefore, the likely glucose transporter involved in glucose-sensing mechanisms, since its Km value is approximately similar to glucose concentration in the CSF. At present, the role of GLUT2 in these neurons remains unknown. Also, it remains unknown how glucose-sensing neurons could exert a gradual neuroendocrine control on the energy reservoir and organism behavior under physiological conditions.

This question opens another theory to develop, giving more importance to glial cells as the main glucose sensors. Both astrocytes and tanycytes can incorporate glucose and metabolize it to lactate. As previously mentioned, tanycytes express GLUT2 [27]. Since tanycytes are surrounding the ME, glucose levels may be sufficient and suitable to be transported through GLUT2. Thereby, the tanycytes would be the first glucose sensors initiating a response. In fact, it has been determined that tanycytes are able to respond themselves to changes in glucose with intracellular increases of Ca^{2+}, which propagate as waves through other tanycytes, giving them the ability to respond independently and synergistically with neuronal responses (releasing gliotransmitters as ATP), which might modulate the behavior of the organism in response to glucose levels [132, 133]. This issue will be discussed further in the "Energy Homeostasis and Neuroendocrine Control" section.

Another possible role for GLUT2 in glucose sensing could be due to its possible role as both transporter and receptor. GLUT2 presents an intracellular loop located between transmembrane domains 6 and 7 that is implicated in intracellular signaling [134]. Blocking the intracellular loop properties of GLUT2 in mice generated defects in glucose homeostasis, such as increased feeding pattern or lower c-Fos activation of the ARC in response to glucose [135, 136]. Cell-specific inhibition of GLUT2 "transceptor" properties in the brain should be assessed to acknowledge its role in glucose-sensing mechanisms as well as determine possible intermediates interacting with its intracellular loop.

Possible Mechanisms of Glucose Sensing in Tanycytes

Like glucose-sensitive neurons, tanycytes have recently become more important as possible glucose sensors possessing their own sensing mechanisms, in addition to their role of suppliers of lactate to neurons. In fact, tanycytes have been suggested to have both metabolism-dependent and metabolism-independent glucose-sensing mechanisms. However, the components of the latter have yet to be elucidated.

In addition to expressing GLUT2, the presence of Kir6.1 subunit of the K_{ATP} channel has been shown in tanycyte cultures by immunocytochemistry. Also, detection of GK, MCT1 and MCT4 as well as low detection of GFAP suggests that the tanycyte culture was enriched with β1 tanycytes, indicating that the Kir6.1 subunit was present, at least in β1 tanycytes [132]. Furthermore, it has been observed that intracellular Ca^{2+} levels arise in α and β1 tanycytes in response to increased glucose and ATP levels, which is probably related to activation of hemichannels composed of six oligomerized connexin 43 (Cx43) subunits [132, 133].

In the presence of diazoxide, an activator of K_{ATP}, intracellular Ca^{2+} was reduced significantly in β1 tanycytes [132]. GLUT, GK, and K_{ATP} blockers reduced Cx43 hemichannel activity, and tanycytes bathed in a divalent cation-free solution (DCFS), known to induce hemichannel opening, exhibited higher glucose kinetics of transport. Since Cx43 hemichannels have been reported to mediate ATP release [137], the response of tanycytes to increased ATP concentrations was determined; it exhibited a similar response to increased glucose concentration, inducing a rise in Ca^{2+} signal [132]. This suggests that a K_{ATP}-mediated mechanism is necessary for hemichannel activity, which, in turn, would allow the entry of glucose into the cell together with GLUT transporters, as well as the release of ATP.

These elements compose a possible glucose-sensing mechanism in tanycytes, involving Ca^{2+} waves capable of traveling and transmitting to other tanycytes. Based on the model described by Orellana et al. (2012) [132], higher glucose concentrations accessed by tanycytes lining the third ventricle are incorporated into the cell via GLUT2 and Cx43-hemichannels, being phosphorylated by GK, leading to increased intracellular ATP through anaerobic glycolysis, which induces the closure of K_{ATP} channels. This creates, through a yet-unknown mechanism, an increase in Cx43 hemichannel expression. Intracellular ATP is transferred out of the cell through hemichannels, binding to P2Y1 purinergic receptors.

Both $P2Y_1$ and inositol triphosphate (IP3) receptor blockers inhibited the rise in Ca^{2+} signal, suggesting that P2Y1 activation could be involved in IP3 activity, which is also likely to be necessary for intracellular Ca^{2+} release [132].

The intracellular increase of Ca^{2+} propagates similar waves among other tanycytes, through P2Y1 receptor activation. An extracellular Ca^{2+}-free solution exerted almost no effect on tanycyte response to glucose on the Ca^{2+} signal, suggesting that Cx43 hemichannels (whose inhibition decreased the Ca^{2+} signal) do not participate in the influx of extracellular Ca^{2+}, hence postulating the involvement of Ca^{2+} release from intracellular reservoirs as the main molecule responsible for the Ca^{2+} signal [132] (Fig. 4a).

Although ATP is essential to trigger the activation of P2Y1 receptors, not only glucose favors this process. The addition of the glucose analogues, such as 2-DG and α-MDG, evokes a similar response to glucose in α tanycytes [133]. It is possible that the metabolism-independent mechanism in tanycytes is similar to the SGLT mechanism shown in neurons, although the lack of data about the expression of SGLT in tanycytes has hampered this to go beyond a mere hypothesis. It is also likely that glucose binds a membrane receptor producing an increase in intracellular Ca^{2+}, which may be mediated by ATP release and PY21 activation as seen in the metabolism-dependent mechanism. Supporting this idea, expression of the sweet taste receptor T1R2/3 has been identified in 58% of tanycytes in hypothalamic slices, being activated by nonnutritive sweeteners [24]. However, activation of metabolism-independent tanycytes was reduced by removing either extracellular and intracellular

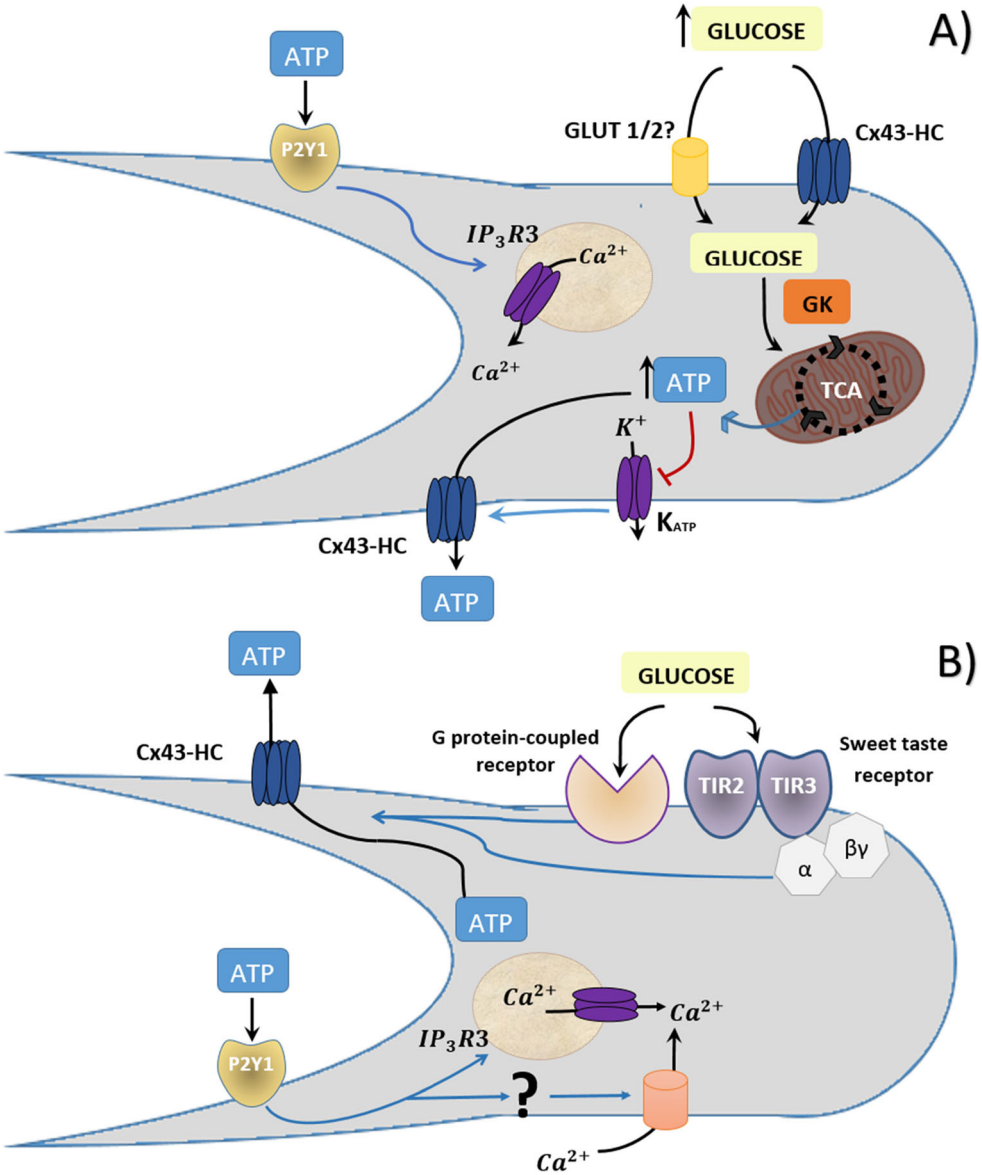

Fig. 4 Schematic representation of the proposed glucose-sensing mechanisms in tanycytes. **a** Metabolism-dependent mechanism. Glucose is internalized via GLUT and Cx43 hemichannels and enters the glycolytic cycle, producing ATP, which leads to closure of K_{ATP} channels. By a yet unknown mechanism, K_{ATP} channels enhance ATP output through Cx43 hemichannels. Binding of ATP to P2Y1 receptors induces intracellular Ca^{2+} release from the internal storage in an IP3-dependent manner. **b** Metabolism-independent mechanism. Either binding of glucose or glucose analogs to T1R2/3 or another extracellular receptor produces a Ca^{2+} signal, possibly inducing ATP output through Cx43 hemichannels. P2Y1 activation induces intracellular Ca^{2+} release from the internal storage and, possibly extracellular Ca^{2+} internalization. Cx43-HC connexin 43 hemichannels. GK glucokinase, GLUT2/3 glucose transporter, IP_3R3 inositol triphosphate receptor, K_{ATP} ATP-sensitive K^+ channel, P2Y1 purinergic receptor, SGLT sodium-glucose cotransporter, T1R2/3 sweet taste receptors, TCA tricarboxylic acid cycle. Scheme based on "Orellana et al. (2012). Glia, 60(1), 53-68" [24]

Ca^{2+}, suggesting a mechanism of activation that is different than metabolism-dependent tanycytes [24] (Fig. 4b). Differences in procedures (i.e., tanycyte cultures vs. brain slice tanycytes) could be responsible for the conflicting reports regarding the requirement for extracellular Ca^{2+}.

Together, the data presented in this section clarify the importance of the environment surrounding glucosensitive neurons. Metabolism-dependent glucose-sensing seems to be closely linked to the glia, essentially to astrocytes and tanycytes. Astrocytes are the main source of lactate, which, on the one hand, could promote a more sensitive response to increased glucose levels, but also decrease the neuronal response by maintaining ATP levels in cases of recurrent hypoglycemia. Tanycytes surround the ME and ventricular walls of the hypothalamus. In addition to directly accessing glucose in the CSF and delivering it as lactate, they may have their own glucose-sensing mechanisms. To couple all this information, it is necessary to describe which regions or neuronal nuclei are involved in glucose-sensing and each individual role in the responses to glucose variations, as will be seen in the "Energy Homeostasis and Neuroendocrine Control" section.

Energy Homeostasis and Neuroendocrine Control

Two brain regions cooperate in regulating glucose levels: (1) the hypothalamus and (2) the brainstem (consisting of the midbrain, pons, and medulla oblongata) [138]. Both are involved in the counterregulatory response to hypoglycemia through activation of the SANS, hormone secretion of the anterior pituitary, and regulation of behavioral patterns, such as control of appetite and food intake, energy expenditure, thermogenesis, and circadian rhythms.

Brain Regions

The Hypothalamus

The hypothalamus is an ensemble of interconnected heterogeneous nuclei, allowing a fine control of glucose levels by integrating and scattering signals with projections to different regions, especially to the brainstem. Specifically, it has been established that various nuclei help maintain energy homeostasis, including the ARC, LH, VMN, PVN, and the dorsal medial hypothalamus (DMH), regions demonstrated to contain populations of glucose-sensitive neurons [139, 140] (Fig. 5).

The central control of homeostasis is established by a balance in the secretion of orexigenic peptides (stimulators of food intake and inhibitors of body energy expenditure) and anorexigenic peptides (inhibitors of food intake and stimulators of body energy expenditure) by different neurons responsive to glucose and other metabolic signals. Since the hypothalamus is adjacent to the third ventricle, where the BBB becomes slightly permeable, hypothalamic nuclei have access to different peptides and hormones produced by adipose tissue (leptin), the pancreas (insulin, glucagon), and the gastrointestinal tract (polypeptide YY and cholecystokinin). All of these metabolic signals access the brain through the blood supply, modulating neuronal response according to nutritional status and body energy demand [142].

The Arcuate Nucleus (ARC) The ARC is the closest structure to the hypothalamic ME. Due to its location, the ARC is considered the center for integration of peripheral metabolic information and control of the energy state of the body. Control of energy homeostasis in the ARC resides in the activity and secretion of two distinct neuronal populations: (1) GI neurons that produce NPY and AgRP to promote body mass increase and suppress appetite and (2) GE neurons that produce POMC and CART, which cause weight loss by inhibiting food intake and stimulating energy expenditure (Table 1). Balance in the secretion of these neuropeptides is essential in controlling behavior intake [162, 163].

The Lateral Hypothalamus (LH) The LH is a hypothalamic region with orexigenic function, considered a "hunger center". It is the most interconnected hypothalamic nucleus, sending projections to other hypothalamic nuclei, in addition to the thalamus, hippocampus, brainstem, and spinal cord. An injury at the LH causes a decrease in food intake, body weight loss, and increased energy expenditure [164–167].

These effects reside in the activity and secretion of two neuronal populations in the LH: GI neurons that produce orexin A and/or orexin B neuropeptides, and GE neurons that produce MCH (Table 1). Both peptides are essential in the melanocortin pathway as well as the onset of the pancreatic and adrenal sympathetic response in conditions of hypoglycemia [168].

The Ventromedial Nucleus of the Hypothalamus (VMN) As opposed to the LH, the VNM is preferably anorexigenic and considered a "satiety center". It is one of the most studied hypothalamic regions for glucose-sensing mechanisms, despite not having neuronal populations with neurochemical markers defined as NPY/AgRP neurons and POMC/CART neurons of the ARC or orexin neurons and MCH neurons of the LH. A bilateral injury in the VMN in rodents induces an increase in food intake and subsequent obesity, confirming its role as an anorexigenic regulator [164, 169] (Table 2).

The Paraventricular Nucleus of the Hypothalamus (PVN) Located at the upper portion of the third ventricle, the PVN is one of the most important centers in the regulation of glycemia through the hypothalamic-pituitary axis. It

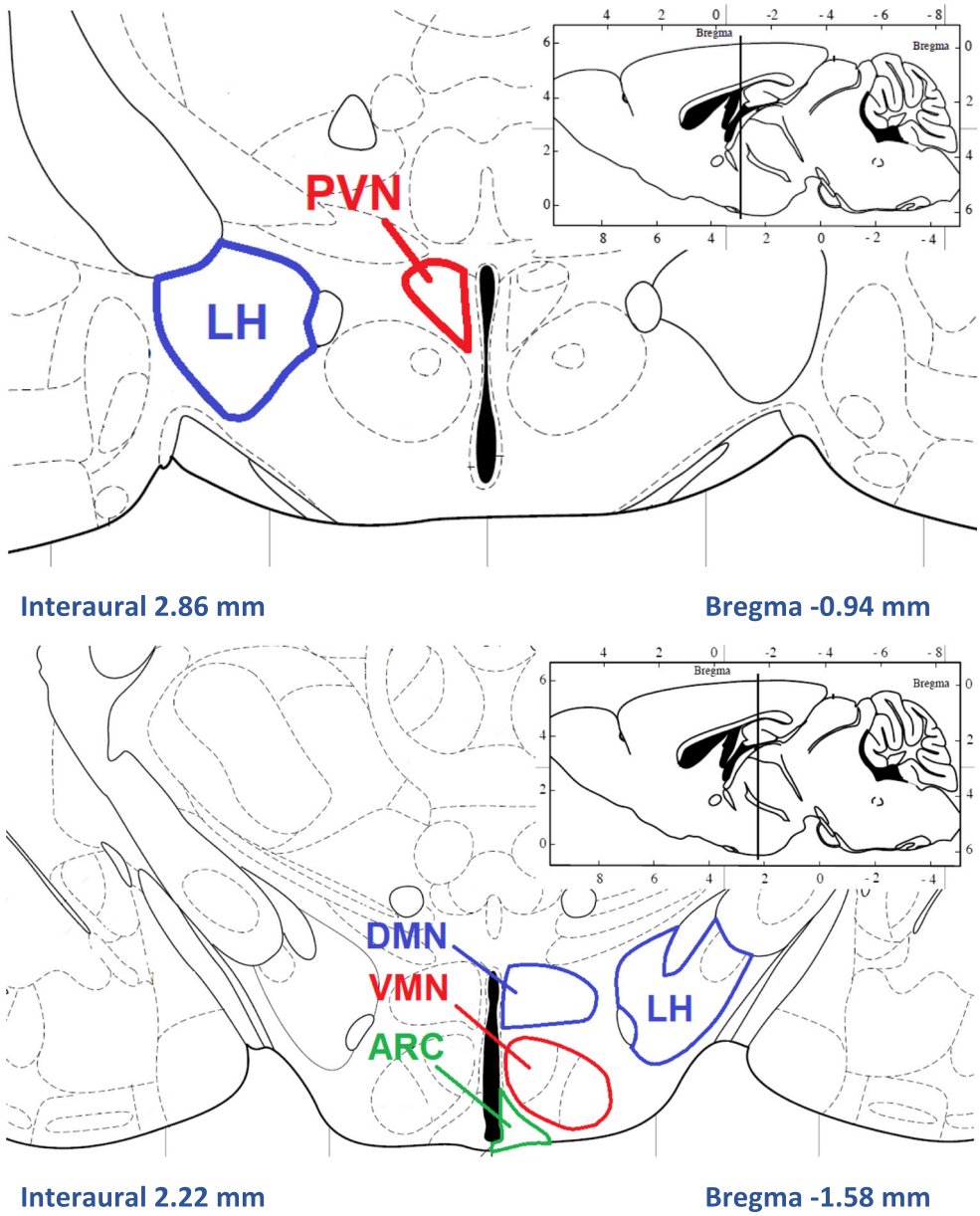

Fig. 5 Stereotactic location of hypothalamic nuclei in the mouse brain. Green: arcuate nucleus (ARC), first-order center. Blue: lateral hypothalamus (LH) and dorsomedial nucleus (DMN), orexigenic or hunger centers. Red: paraventricular nucleus (PVN) and ventromedial nucleus (VMN), anorexigenic or satiety centers. Modified from "Paxinos, G., & Franklin, K. B. (2004). The mouse brain in stereotaxic coordinates. Gulf Professional Publishing " [141]

harbors populations of parvocellular neurons secreting corticotrophin releasing hormone (CRH) [175] and thyrotropin-releasing hormone (TRH) [176], both of which suppress appetite and food intake, suggesting that the PVN is also a "satiety center" [177, 178]. In fact, bilateral lesions of the PVN in animals generated an increased dietary intake along with obesity [179].

Neurosecretory neurons are preferentially located in the medial part of the PVN and have no intrinsic glucose-sensing mechanism. Thus, they depend on afferent signals provided by other neuronal populations. The neural regulation of food intake and body energy homeostasis lies in GE and GI neurons of the PVN, mostly distributed in the dorsal area of the PVN [13] (Table 2). These neurons are connected via projections to the brainstem, as well as to ANS neurons in the spinal cord, which innervate endocrine organs, the liver or adipose tissue, regulating hormone secretion and metabolic activity associated with plasma glucose levels [180, 181].

Dorsomedial Hypothalamic Nucleus (DMN) The DMN is a hypothalamic nucleus involved in the control of circadian rhythms; it receives projections from other hypothalamic nuclei involved in glucose sensing.

Table 1 Neurosecretory phenotype of glucose-sensitive neurons in the arcuate nucleus (ARC) and lateral hypothalamus (LH) and effects on energy homeostasis

Nucleus and phenotype	Glucose-sensing mechanism	Metabolic effects	Receptors	Experiments supporting and other issues	Refs.
ARC: NPY/AgRP Nt: GABA	Glucose-inhibited (GI) AMPK/nNOS/sGC/CFTR Mechanism	Orexigenic ↑Food intake Thermogenesis modulation	NPY: Y_1, Y_2, Y_4, Y_5, Y_6 G protein-coupled (G_i inhibitory subunit) AgRP: MC3R, MC4R (antagonist)	Inhibition of 40% of isolated NPY neurons by glucose. c-Fos expression and increased depolarization rate in hypoglycemia. ICV injection of NPY and AgRP stimulates food intake. GABA vesicular transporter (*vgat*) knockout mice in AgRP neurons induces a lean phenotype.	[143] [144, 145] [146, 147]
ARC: POMC/CART (release of α-MSH) Nt: glutamate and GABA	Glucose-excited (GE) GLUT3/GK/K_{ATP} mechanism	Anorexigenic ↓Food intake Thermogenesis modulation	α-MSH: MC3R, MC4R (agonist)	Lower depolarization rate in hypoglycemia. Biphasic phenotype discovery: decrease and later increase of depolarization rate in acute hypoglycemia. Expression of K_{ATP} subunits SUR1 and Kir6.2 and glycemia-insensitive Kir6.2 knockout mice. Inhibition of UCP2 with genipin and higher response to glucose. Suppression of MC4R, leading to hyperphagia and obese phenotype.	[148] [149] [150] [20] [151, 152]
LH: Orexin A and/or Orexin B Nt: acetilcholine	Glucose-inhibited (GI) K2P mechanism	Orexigenic ↑Food intake ↑Gastric motility Wakefulness	Orexin A, orexin B: OxR1, OxR2	Depolarization rate was decreased in 80% of orexigenic neurons with >5 mM glucose. Depolarization rate decreased in 95% of LH orexigenic neurons with 0.2–5 mM glucose. Food intake was stimulated after ICV injection of orexin. Orexin knockout mice and decreased food intake.	[153] [154] [155] [156]
LH: MCH Nt not determined	Glucose-Excited (GE) GLUT3/GK/K_{ATP} mechanism	Orexigenic ↑Food intake (only caloric nutrients) Anabolic processes Sleepness	MCH: MCH-R1, MCH-R2	Depolarization rate increased in 80% of MCH neurons in normoglycemia, depending on dosis. MCH and MCH1R knockout mice and increased metabolic rate with lean phenotype. MCH1R antagonist administration and lower appetite for glucose (no changes observed for saccharin). Appetite for glucose maintained in TrpM5 (Na^+ channel) deficient mice.	[154] [157, 158] [159] [160, 161]

Table 2 Neurosecretory phenotype of glucose sensitive neurons in the ventromedial nucleus (VMN), paraventricular nucleus (PVN), and dorsomedial nucleus (DMN), and effects on energy homeostasis

Nucleus and phenotype	Glucose-sensing mechanism	Metabolic effects	Experiments supporting and other issues	Refs.
VMN: Neuropeptide not defined Nt: Glutamate	Glucose-inhibited (GI) AMPK/nNOS/sGC/CFTR mechanism	Sympathetic response activation and release of glucagon and epinephrine	SF1 neurons in the VMN necessary to set the sympathetic response. Vesicular glutamate transporter (*vglut2*) knockout mice and suppression of glucagon release. Loss of GABAergic afferent projections on BDNF1 knockout mice depending on MCH-R4 and inhibition of glucagon release.	[170] [55] [171]
VMN: Neuropeptide not defined Nt: GABA	Glucose-excited (GE) GLUT3/GK/K_{ATP} mechanism	Sympathetic response inhibition	Inhibition of $GABA_A$ receptors with the antagonist, bicuculline, and activation with the agonist, muscimol. Induction and inhibition of sympathetic response on hypoglycemia, respectively.	[172]
VMN: Neuropeptide not defined Nt not definied	Glucose-excited (GE) SGLT1 mechanism	Sympathetic response inhibition and lower release of glucagon and epinephrine	Lower expression of SGLT1 mRNA in the VMN increases the counterregulatory response against hypoglycemia with increased secretion of glucagon and epinephrine.	[173]
PVN: Neuropeptide not defined Nt not defined	Glucose-inhibited (GI) AMPK/nNOS/sGC/? mechanism		Depolarization of 26% of PVN neurons in glucopenia (0.2 mM glucose).	[13]
PVN: Neuropeptide not defined Nt not defined	Glucose-excited (GE) Metabolism-independent mechanism		Hyperpolarization of 24% of PVN neurons in glucopenia (0.2 mM glucose). Addition of K_{ATP} blockers tolbutamide and glibenclamide as well as addition of K_{ATP} activator diazoxide with no glucose-like response in PVN neurons.	[13] [13]
DMN: Neuropeptide not defined Nt not defined	Glucose-inhibited (GI) K2P (adaptative) mechanism	↑Food intake	Basal membrane potential was restored faster in DMN GI neurons than in LH GI neurons. Injury of the DMN decreases food intake and produces a lean phenotype.	[7] [174]

Like the LH, the DMN has orexinergic GI neurons. However, the DMN orexinergic neurons present an adaptive phenotype with the ability to retrieve the baseline membrane potential quickly after being inhibited in the presence of glucose [123] (Table 2).

This structure also has NPY neurons. The presence of neuronal populations secreting orexigenic peptides gives the DMN a role as a "hunger center," dependent on other hypothalamic nuclei. Indeed, bilateral injury of the DMN in animals generates a decrease of both food intake and body weight [174].

The Brainstem

The brainstem is the most caudal part of the brain, consisting of the medulla oblongata, pons, and midbrain. Several neuronal nuclei of the brainstem are involved in the control of heart rate, blood pressure and energy expenditure. The existence of glucose-sensitive neuronal populations in the medulla oblongata have been described, particularly in the rostral ventrolateral region of the medulla (RVLM) and the dorsal vagal complex (DVC), the latter being composed of the nucleus of the solitary tract (NTS), the dorsal motor nucleus of the vagus (DMV) and the area postrema (AP) (Fig. 6). These regions surround the aqueduct and the fourth ventricle, allowing diffusion of plasma glucose to the brain parenchyma [57, 123, 183].

The Nucleus of the Solitary Tract (NTS) The NTS is a set of neuronal nuclei pierced by the fiber bundle of the solitary tract, bringing together glucose-sensitive neurons among different neuronal groups. Specifically, glucose sensing is defined by a subpopulation of GI neurons in the NTS, sending GABAergic projections to DMV neurons in order to initiate the counterregulatory response in hypoglycemia [19, 26].

Acting as an integrating center, the NTS receives peripheral hypoglycemic signals detected by glucose-sensitive cell bodies in the hepatic portal vein, which project on the intermediolateral nucleus (IML) of the spinal cord, contacting the NTS [184].

The NTS also encompasses catecholaminergic A2 (noradrenergic) and C2 (adrenergic) neurons, which both express NPY and send axonal projections on the PVN, exercising control over the secretion of CRH in hypoglycemia. Reciprocally, CRH neurons project onto the A2 neurons, establishing a feedback loop [185].

Many other hypothalamic nuclei innervate the NTS, and the amygdala innervates the ARC, which sends projections on the A1 neurons and non-catecholaminergic neurons in the RVLM [186].

The Dorsal Motor Nucleus of the Vagus (DMV) The soma of the preganglionic neurons of the vagus nerve are located in the DMV, which activates the parasympathetic pathway, exercising control over pancreatic secretion. At least a subpopulation of these neurons is glucose-sensitive, decreasing their depolarization rate in conditions of hypoglycemia [57, 187].

The Area Postrema (AP) The AP is considered to be the "trigger" of the emetic response and harbors both GE and GI neurons. Ablation of the AP in rats causes an increase in food intake, demonstrating that this area has a role in glucose homeostasis [57, 188].

The Rostral Ventrolateral Medulla (RVLM) The RVLM harbors spinal metabolism-dependent GI neurons, producing a hyperglycemic response against hypoglycemia. The sympathetic-adrenal response is mediated by A1/C1 catecholaminergic neurons, inducing the secretion of epinephrine in the adrenal medulla. The destruction of C1 neurons by immunotoxins results in a decrease in the secretion of epinephrine and an increase in glucagon secretion as a compensatory mechanism [189, 190].

Given the complexity involved in glucose-sensing, it is expected that most of the regions responsible for this process surround the cerebral ventricles, such as the hypothalamus and

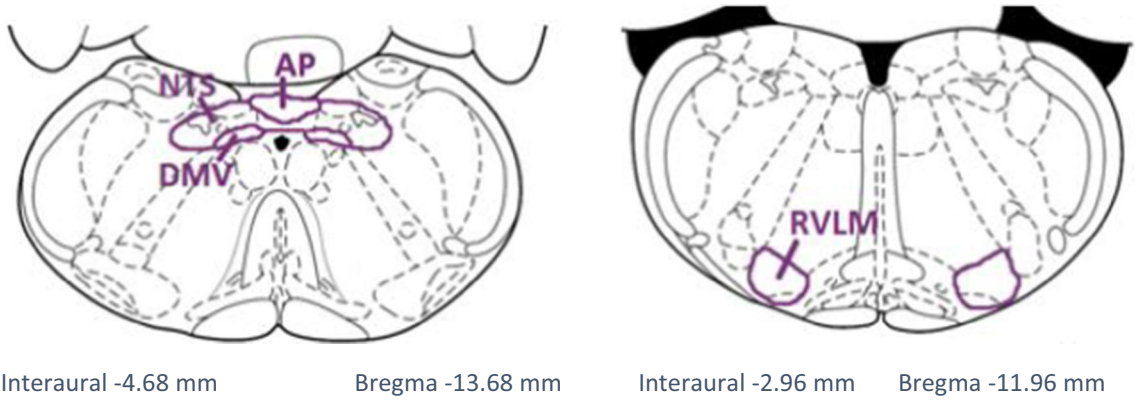

Fig. 6 Stereotactic location of the brainstem nuclei in the rat brain. Area postrema (AP), nucleus of the solitary tract (NTS), dorsal motor nucleus of the vagus (DMV), and rostral ventrolateral medulla (RVLM). Modified from "Paxinos, G., & Franklin, K. B. (1998). The rat brain in stereotaxic coordinates. Academic Press" [182]

brainstem. According to their proximity to the ME and their distribution, hypothalamic nuclei present specialized glucosensitive neuronal populations, which control homeostatic processes in the brainstem, as a link with the autonomic nervous system. Understanding the interactions of these nuclei may shed light on how they occur and the responses involved in the control of physiological processes related to both metabolic and energy balance.

The Autonomic Nervous System and the Response to Hypoglycemia

During hypoglycemia, the nervous system initiates three main mechanisms that mediate the hormonal release of glucagon followed by epinephrine, arising systematically and increasing in magnitude as blood glucose decreases.

The first of the mechanisms described sets up the activation of the parasympathetic branch of the pancreas, mediating glucagon secretion by pancreatic α-cells when glucose levels fall to between 85 and 75 mg/dL (4.7 and 4.2 mM). The second mechanism, which starts before a more pronounced glucose drop to between 75 and 65 mg/dL (4.2 and 3.6 mM), involves a sympatho-adrenal response and the release of epinephrine from the adrenal medulla, which, in turn, stimulates the release of glucagon in the pancreas. The third mechanism arises after the appearance of the first clinical symptoms of hypoglycemia, about 50 mg/dL (2.8 mM), and involves the activation of the sympathetic branch innervating the β-cells [191].

Central Glucose Sensing and Pancreatic and Adrenal Secretions

The pancreas is an essential organ in glucose regulation, widely innervated by the ANS. Parasympathetic activation induces insulin secretion by β-cells and glucagon by α-cells. Conversely, sympathetic activation inhibits insulin secretion and promotes glucagon and epinephrine secretion, the latter being in the adrenal medulla [191].

Anterograde virus neuronal tracing with pseudorabies virus (PSV) has shown that pancreatic parasympathetic innervation is established by DMV neurons in the brainstem, and sympathetic innervation comes from preganglionic motor neurons in the spinal cord. Motor neurons, in turn, receive projections of neurons placed in different hypothalamic and brainstem nuclei, the so-called "second order" neurons, which are connected with other "third order" neurons in relation to the pancreas [192]. A complete sagittal scheme representing these marked neuronal nuclei is available in Buijs et al. [192].

Parasympathetic Branch The DMV is the main effector of the parasympathetic response, stimulating insulin secretion directly by β-cells and glucagon by α-cells. The DMV is innervated by the NTS, home of GLUT2-AMPK-nNOS-sGC-K2P GABAergic GI neurons. In hypoglycemia, these neurons inhibit DMV GABAergic neurons, the latter ones being presynaptic in relation to premotor neurons in the vagus nerve [19].

Regarding the hypothalamus, the DMV is directly innervated by the LH, ARC, DMN, and PVN. The PVN is, in turn, innervated by the VMN [193–195]. The control over glucose levels exercised by these nuclei is essential to inducing the parasympathetic response (Fig. 7). Insulin secretion is dependent on hypothalamic GLUT2-GK-K_{ATP} GE neurons. A central loss of GLUT2 expression results in decreased insulin secretion. In fact, it was shown in knockout models for GLUT2 that parasympathetic activity induces proliferation of β-cells in the first months of life, resulting in poor insulin secretion at later stages and subsequently glucose intolerance and diabetes [196].

Sympathetic Branch Hypothalamic control of the SANS relies on MCH and orexin neurons in the LH and neurons in the PVN, which send axonal projections to sympathetic preganglionic motor neurons as well as to A1/C1 catecholaminergic neurons located in the RVLM. This group of neurons is also innervated by NTS neurons and the locus coeruleus (LC). In addition, the presence of presumably GLUT2-GK-K_{ATP} GE neurons within A5 neurons has been described [197, 198].

Anterograde and retrograde virus neuronal tracing verified the latter efferent projections [199, 200]. In addition, systemic glycoprivation increases the depolarization rate as well as c-Fos and dopamine-β-hydroxylase expression in C1 neurons, while ablation of this group of neurons reduces sympathetic counterregulatory response to hypoglycemia simulated by the injection of 2-DG (nonmetabolizable glucose analog) in the hypothalamus [201, 202].

Hypoglycemia induces orexin secretion by GI neurons in the LH, producing a stimulating effect on the C1 neurons of the RVLM. Both C1 and glutamatergic neurons in the RVLM make connections with adrenal sympathetic preganglionic neurons, stimulating the secretion of glucagon and epinephrine [203] (Fig. 7).

Control by Hypothalamic and Brainstem Glucose-Sensing Neurons over the ANS Metabolites and hormonal signals, including ghrelin, insulin, and leptin, contact the ARC directly from the bloodstream, given its proximity to the ME. Ghrelin is produced in fasting periods and has an orexigenic effect, while insulin and leptin are produced in response to increased glucose and adipose tissue respectively, producing an anorexigenic effect. Anorexigenic NPY/AgRP GI neurons in the ARC are stimulated by ghrelin and inhibited by glucose, insulin and leptin, while orexigenic POMC/CART GE neurons are stimulated by insulin, leptin, and glucose, and inhibited indirectly by ghrelin through activation of NPY/AgRP neurons, exercising GABAergic transmission over POMC/CART neurons [167, 204, 205].

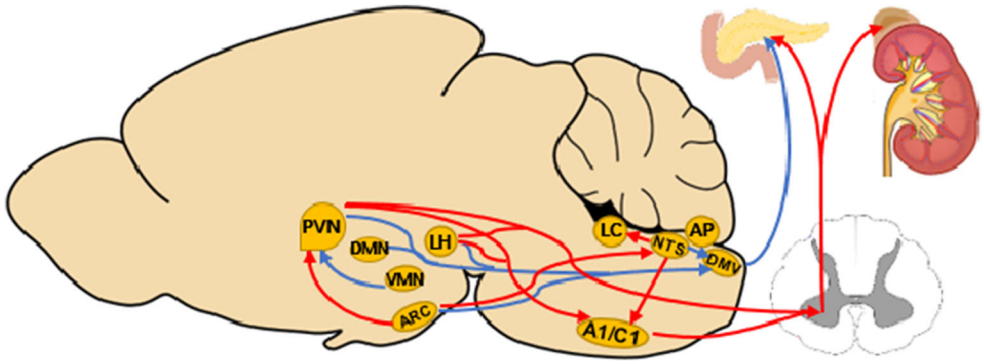

Fig. 7 Representation of the neuronal circuit controlling parasympathetic innervation in the vagus nerve to the pancreas and sympathetic innervation in the spinal cord to the pancreas and the adrenal medulla. A1/C1 catecholaminergic neurons of the rostral ventrolateral medulla, AP area postrema, ARC arcuate nucleus, DMN dorsomedial nucleus, DMV dorsal motor nucleus of the vagus, LC locus coeruleus, LH lateral hypothalamus, NTS nucleus of the solitary tract, PVN paraventricular nucleus, VMN ventromedial nucleus

Establishing a simple model for energy control, under conditions of hypoglycemia and low energy availability, activation of anorexigenic GI neurons leads to a response in the SANS, ending in the release of glucagon from the pancreas and adrenaline from the adrenal medulla, as well as the hypothalamic release of CRH followed by ACTH and glucocorticoids [206–208].

Under conditions of hyperglycemia or high levels of energy, orexigenic GE neurons respond by activating the PANS and releasing insulin, in addition to the sympathetic activation of thermogenesis [20, 209] (Fig. 8).

Appetite Control

Two neuronal nuclei are essential for the central control of appetite: (1) the ARC as the "starter," defining the synaptic connections that reach the parabrachial nucleus (PBN) in the brainstem, which is the other neuronal nucleus and (2) the "end effector" of anorexigenic signals, suppressing appetite [210] (Fig. 9).

In situations of hyperglycemia after food intake, activity of POMC/CART GE neurons increase. These neurons secrete α-MSH, activating the PVN glutamatergic neurons. Successively, PBN is activated, inducing an anorexigenic effect, thus suppressing appetite.

Moreover, during hypoglycemia, NPY/AgRP GI neurons suppress anorexigenic behavior sending GABAergic projections on PVN glutamatergic neurons and the PBN. In fact, chronic injection of bretazenil (partial agonist for $GABA_A$ receptors) into the PBN prevents starvation [211].

In addition, the NTS, which contains glutamatergic neurons that integrate metabolic signals from the gastrointestinal

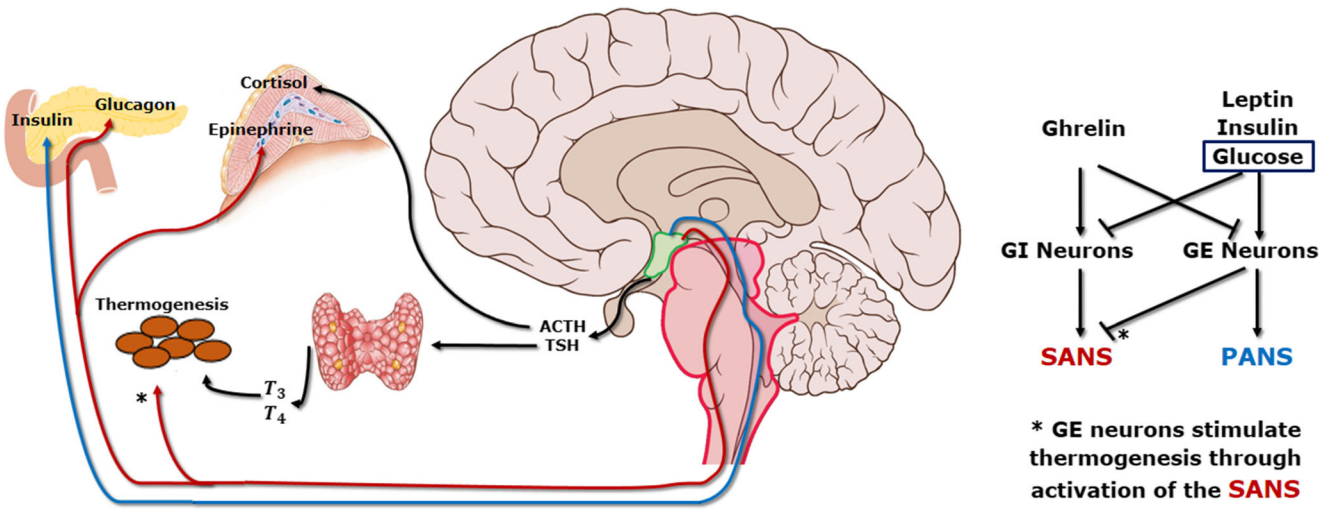

Fig. 8 GE and GI neurons in the hypothalamus (green) and the brainstem (pink) respond to metabolic signals, establishing the autonomic response. High energy signals (leptin, insulin, glucose) enhance activation of GE neurons, setting a parasympathetic response (blue). Low energy signals (ghrelin) enhance activation of GI neurons, inducing a sympathetic response (red) and the release of glucocorticoids. ACTH adenocorticotropic hormone, CRH corticotrophin releasing hormone, GE glucose-excited, GI glucose-inhibited, PANS parasympathetic autonomic nervous system, SANS sympathetic autonomic nervous system, T_3 triiodothyronine, T_4 tetraiodothyronine, TSH thyroid-stimulating hormone

tract, acts as a modulator and integration center, receiving excitatory projections from the PVN (glutamatergic) and the Raphe (serotonergic), inducing activation of the PBN [210, 212].

The LH, considered the most important "hunger center," is essential to induce food intake. Establishing a scheme to fully understand the orexigenic effect generated is hardly feasible due to the diverse and biased innervation from the LH to the brainstem nuclei. In turn, the ARC is one of the areas innervated by the LH. Orexin secretion produces excitatory effects on NPY/AgRP GI neurons and inhibitory effects on POMC/CART neurons. Addition of an NPY antagonist only partially reduces intake, confirming that the LH induces food intake complementarily to the ARC [213–215].

In addition to neurotransmitters, astrocytes and tanycytes have also been reported to modify the appetite of the organism, mainly by secreting gliotransmitters [216]. The best known in this role are endozepines, such as the diazepam-binding inhibitor (DBI) and its active fragment, octadecaneuropeptide (ODN). When administered directly, ODN acts as an anorexigenic factor, reducing food intake in rodents [217]. In addition, post-glucoprivation hyperphagia is suppressed after central injection of an ODN agonist. However, anorexigenic signaling by endozepines involves MC3R and MC4R and binding the $GABA_A$ receptor, but not the benzodiazepine receptors [218]. Thus, GABAergic inhibitory tone would be enhanced to reduce appetite (Fig. 9).

Conclusion and Future Perspectives

In this work, we have reviewed the various mechanisms through which glucose changes in the internal environment of the brain are detected from a molecular and anatomical-functional perspective. The existence of such diverse mechanisms, both dependent and independent of glucose metabolism, leaves open the question of what is the specific function of each and is reflective of the variety of processes used to sense this molecule. Accordingly, glucose fluctuations are detected by GE and GI neurons, which are distributed in several hypothalamic and brainstem nuclei. Many components of these glucose-sensing mechanisms contribute to diabetes development, such as gene variants of GLUT2 [219], glucokinase [220], or AMPK activity [221]. Understanding the specific role of these elements in the brain glucose-sensing elements has the potential to identify novel therapeutic targets.

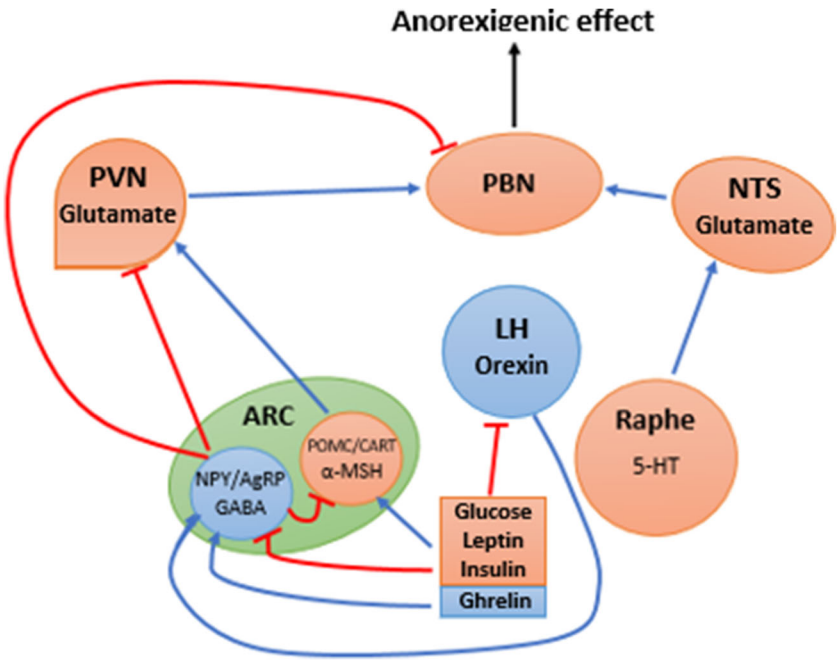

Fig. 9 Wiring diagram established in the regulation of food intake control. Red: anorexigenic neurons and nuclei. Blue: orexigenic neurons and nuclei. Green: first-order nucleus. The ARC is the main controller of the anorexigenic response by sensing energy status from the periphery. In turn, the ARC receives signals from the LH. The PVN receives inhibitory projections from NPY/AgRP GI neurons and excitatory projections from POMC/CART GE neurons in the ARC. Furthermore, the PVN innervates the PBN, the final anorexigenic effectors. The PBN also receives inhibitory projections from NPY/AgRP GI neurons in the ARC and glutamatergic excitatory projections from the NTS, which is, in turn, innervated by serotonergic Raphe projections. 5-HT serotonin (5-HT), AgRP agouti-related protein, ARC Arcuate nucleus, CART cocaine amphetamine regulated transcript peptide, LH lateral hypothalamus, NPY neuropeptide Y, NTS nucleus of the solitary tract, PBN parabrachial nucleus, POMC proopiomelanocortin, PVN paraventricular nucleus, α-MSH alpha-melanocyte stimulating hormone

Several of the mechanisms described are subject to the transport kinetics of glucose, especially those dependent on GLUT2. It has been previously questioned if the entry of glucose through the ME reaches sufficient levels for its transport through GLUT2 in the hypothalamus. Therefore, it has been suggested that glucose uptake by glial cells and its subsequent metabolism to lactate as a source of ATP to neurons, could imply a direct relationship between both cell types in glucose sensing. A subtype of glial cell of special relevance are the tanycytes surrounding the ME and ventricular walls. In addition to forming the initial barrier for glucose entry (tanycytes beta2), tanycytes alpha and beta 1 could also present their own glucose-sensing mechanism analogous to those described in neurons.

Acknowledgements This work was supported by FONDECYT Initiation into Research grant Fondecyt 11150678 (Fernando MartÍnez), Fondecyt 11140405 (Katterine Salazar), Fondecyt 1181243 (Francisco Nualart), and a CONICYT PIA ECM-12 grant (Francisco Nualart). The funders had no role in the study design, data collection, data analysis, decision to publish, or preparation of the manuscript.

References

1. Shen J, Petersen KF, Behar KL, Brown P, Nixon TW, Mason GF, Petroff OAC, Shulman GI et al (1999) Determination of the rate of the glutamate/glutamine cycle in the human brain by in vivo 13C NMR. Proc Natl Acad Sci U S A 96(14):8235–8240
2. Haberg A, Qu H, Saether O, Unsgard G, Haraldseth O, Sonnewald U (2001) Differences in neurotransmitter synthesis and intermediary metabolism between glutamatergic and GABAergic neurons during 4 hours of middle cerebral artery occlusion in the rat: The role of astrocytes in neuronal survival. J Cereb Blood Flow Metabol Off J Int Soc Cereb Blood Flow Metabol 21(12):1451–1463
3. Howarth C, Gleeson P, Attwell D (2012) Updated energy budgets for neural computation in the neocortex and cerebellum. J Cereb Blood Flow Metabol Off J Int Soc Cereb Blood Flow Metabol 32(7):1222–1232
4. Group DR (1991) Epidemiology of severe hypoglycemia in the diabetes control and complications trial. The DCCT research group. Am J Med 90(4):450–459
5. Investigators TN-SS (2012) Hypoglycemia and risk of death in critically ill patients. N Engl J Med 367(12):1108–1118
6. Klein R (1995) Hyperglycemia and microvascular and macrovascular disease in diabetes. Diabetes Care 18(2):258–268
7. Aronoff SL, Berkowitz K, Shreiner B, Want L (2004) Glucose metabolism and regulation: Beyond insulin and glucagon. Diabetes Spectrum 17(3):183–190
8. Forhead AJ, Cutts S, Matthews PA, Fowden AL (2009) Role of thyroid hormones in the developmental control of tissue glycogen in fetal sheep near term. Exp Physiol 94(10):1079–1087
9. Ogunnowo-Bada EO, Heeley N, Brochard L, Evans ML (2014) Brain glucose sensing, glucokinase and neural control of metabolism and islet function. Diabetes Obes Metab 16(Suppl 1):26–32
10. Anand BK, Chhina GS, Sharma KN, Dua S, Singh B (1964) Activity of single neurons in the hypothalamic feeding centers: Effect of glucose. Am J Phys 207:1146–1154
11. Oomura Y, Yoshimatsu H (1984) Neural network of glucose monitoring system. J Auton Nerv Syst 10(3–4):359–372
12. Burdakov D, Luckman SM, Verkhratsky A (2005) Glucose-sensing neurons of the hypothalamus. Philos Trans R Soc Lond Ser B Biol Sci 360(1464):2227–2235
13. Melnick IV, Price CJ, Colmers WF (2011) Glucosensing in parvocellular neurons of the rat hypothalamic paraventricular nucleus. Eur J Neurosci 34(2):272–282
14. Dallaporta M, Himmi T, Perrin J, Orsini JC (1999) Solitary tract nucleus sensitivity to moderate changes in glucose level. Neuroreport 10(12):2657–2660
15. Ritter S, Li AJ, Wang Q, Dinh TT (2011) Minireview: The value of looking backward: The essential role of the hindbrain in counterregulatory responses to glucose deficit. Endocrinology 152(11):4019–4032
16. Sprague JE, Arbelaez AM (2011) Glucose counterregulatory responses to hypoglycemia. Pediatr Endocrinol Rev 9(1):463–473 quiz 74-5
17. Borg MA, Sherwin RS, Borg WP, Tamborlane WV, Shulman GI (1997) Local ventromedial hypothalamus glucose perfusion blocks counterregulation during systemic hypoglycemia in awake rats. J Clin Invest 99(2):361–365
18. Kang L, Routh VH, Kuzhikandathil EV, Gaspers LD, Levin BE (2004) Physiological and molecular characteristics of rat hypothalamic ventromedial nucleus glucosensing neurons. Diabetes 53(3):549–559
19. Boychuk CR, Gyarmati P, Xu H, Smith BN (2015) Glucose sensing by GABAergic neurons in the mouse nucleus tractus solitarii. J Neurophysiol 114(2):999–1007
20. Parton LE, Ye CP, Coppari R, Enriori PJ, Choi B, Zhang CY, Xu C, Vianna CR et al (2007) Glucose sensing by POMC neurons regulates glucose homeostasis and is impaired in obesity. Nature 449(7159):228–232
21. Ashford ML, Boden PR, Treherne JM (1990) Glucose-induced excitation of hypothalamic neurones is mediated by ATP-sensitive K+ channels. Pflugers Arch 415(4):479–483
22. Mounien L, Marty N, Tarussio D, Metref S, Genoux D, Preitner F, Foretz M, Thorens B (2010) Glut2-dependent glucose-sensing controls thermoregulation by enhancing the leptin sensitivity of NPY and POMC neurons. FASEB J Off Publ Fed Am Soc Exp Biol 24(6):1747–1758
23. Díez-Sampedro A, Hirayama BA, Osswald C, Gorboulev V, Baumgarten K, Volk C et al (2003) A glucose sensor hiding in a family of transporters. Proc Natl Acad Sci 100(20):11753–11758
24. Benford H, Bolborea M, Pollatzek E, Lossow K, Hermans-Borgmeyer I, Liu B et al (2017) A sweet taste receptor-dependent mechanism of glucosensing in hypothalamic tanycytes. Glia 65(5):773–789
25. Leloup C, Arluison M, Lepetit N, Cartier N, Marfaing-Jallat P, Ferre P et al (1994) Glucose transporter 2 (GLUT 2): Expression in specific brain nuclei. Brain Res 638(1–2):221–226
26. Lamy CM, Sanno H, Labouebe G, Picard A, Magnan C, Chatton JY et al (2014) Hypoglycemia-activated GLUT2 neurons of the nucleus tractus solitarius stimulate vagal activity and glucagon secretion. Cell Metab 19(3):527–538
27. Garcia M, Millan C, Balmaceda-Aguilera C, Castro T, Pastor P, Montecinos H et al (2003) Hypothalamic ependymal-glial cells express the glucose transporter GLUT2, a protein involved in glucose sensing. J Neurochem 86(3):709–724
28. Kang L, Dunn-Meynell AA, Routh VH, Gaspers LD, Nagata Y, Nishimura T, Eiki J, Zhang BB et al (2006) Glucokinase is a critical regulator of ventromedial hypothalamic neuronal glucosensing. Diabetes 55(2):412–420
29. Dunn-Meynell AA, Routh VH, Kang L, Gaspers L, Levin BE (2002) Glucokinase is the likely mediator of glucosensing in both glucose-excited and glucose-inhibited central neurons. Diabetes 51(7):2056–2065

30. Lee K, Dixon AK, Richardson PJ, Pinnock RD (1999) Glucose-receptive neurones in the rat ventromedial hypothalamus express K(ATP) channels composed of Kir6.1 and SUR1 subunits. J Physiol 515(Pt 2):439–452
31. Karschin A, Brockhaus J, Ballanyi K (1998) KATP channel formation by the sulphonylurea receptors SUR1 with Kir6.2 subunits in rat dorsal vagal neurons in situ. J Physiol 509(Pt 2):339–346
32. Aguilar-Bryan L, Clement JP, Gonzalez G, Kunjilwar K, Babenko A, Bryan J (1998) Toward understanding the assembly and structure of KATP channels. Physiol Rev 78(1):227–245
33. Zhu W, Czyzyk D, Paranjape SA, Zhou L, Horblitt A, Szabo G et al (2010) Glucose prevents the fall in ventromedial hypothalamic GABA that is required for full activation of glucose counterregulatory responses during hypoglycemia. Am J Physiol Endocrinol Metab 298(5):E971–E977
34. Chan O, Lawson M, Zhu W, Beverly JL, Sherwin RS (2007) ATP-sensitive K(+) channels regulate the release of GABA in the ventromedial hypothalamus during hypoglycemia. Diabetes 56(4): 1120–1126
35. Kong D, Vong L, Parton LE, Ye C, Tong Q, Hu X et al (2010) Glucose stimulation of hypothalamic MCH neurons involves K(ATP) channels, is modulated by UCP2, and regulates peripheral glucose homeostasis. Cell Metab 2(15):545–552
36. Ricquier D, Bouillaud F (2000) The uncoupling protein homologues: UCP1, UCP2, UCP3, StUCP and AtUCP. Biochem J 345(Pt 2):161–179
37. Bouillaud F (2009) UCP2, not a physiologically relevant uncoupler but a glucose sparing switch impacting ROS production and glucose sensing. Biochim Biophys Acta 1787(5):377–383
38. Pecqueur C, Alves-Guerra C, Ricquier D, Bouillaud F (2009) UCP2, a metabolic sensor coupling glucose oxidation to mitochondrial metabolism? IUBMB Life 61(7):762–767
39. Archer SL, Wu XC, Thebaud B, Moudgil R, Hashimoto K, Michelakis ED (2004) O2 sensing in the human ductus arteriosus: Redox-sensitive K+ channels are regulated by mitochondria-derived hydrogen peroxide. Biol Chem 385(3–4):205–216
40. Avshalumov MV, Rice ME (2003) Activation of ATP-sensitive K+ (K(ATP)) channels by H2O2 underlies glutamate-dependent inhibition of striatal dopamine release. Proc Natl Acad Sci U S A 100(20):11729–11734
41. Leloup C, Magnan C, Benani A, Bonnet E, Alquier T, Offer G, Carriere A, Periquet A et al (2006) Mitochondrial reactive oxygen species are required for hypothalamic glucose sensing. Diabetes 55(7):2084–2090
42. O'Malley D, Reimann F, Simpson AK, Gribble FM (2006) Sodium-coupled glucose cotransporters contribute to hypothalamic glucose sensing. Diabetes 55(12):3381–3386
43. Yu AS, Hirayama BA, Timbol G, Liu J, Diez-Sampedro A, Kepe V, Satyamurthy N, Huang SC et al (2013) Regional distribution of SGLT activity in rat brain in vivo. Am J Physiol Cell Physiol 304(3):C240–C2C7
44. Bianchi L, Diez-Sampedro A (2010) A single amino acid change converts the sugar sensor SGLT3 into a sugar transporter. PLoS One 5(4):e10241
45. Lemon CH, Margolskee RF (2009) Contribution of the T1r3 taste receptor to the response properties of central gustatory neurons. J Neurophysiol 101(5):2459–2471
46. Ren X, Zhou L, Terwilliger R, Newton SS, de Araujo IE (2009) Sweet taste signaling functions as a hypothalamic glucose sensor. Front Integr Neurosci 3:12
47. Welcome MO, Mastorakis NE, Pereverzev VA (2015) Sweet taste receptor signaling network: Possible implication for cognitive functioning. Neurol Res Int 2015:606479
48. Kinnamon SC (2012) Taste receptor signalling - from tongues to lungs. Acta Physiol (Oxf) 204(2):158–168
49. Zhang Y, Hoon MA, Chandrashekar J, Mueller KL, Cook B, Wu D, Zuker CS, Ryba NJP (2003) Coding of sweet, bitter, and umami tastes: Different receptor cells sharing similar signaling pathways. Cell 112(3):293–301
50. Clapp TR, Medler KF, Damak S, Margolskee RF, Kinnamon SC (2006) Mouse taste cells with G protein-coupled taste receptors lack voltage-gated calcium channels and SNAP-25. BMC Biol 4:7
51. Finger TE, Danilova V, Barrows J, Bartel DL, Vigers AJ, Stone L, Hellekant G, Kinnamon SC (2005) ATP signaling is crucial for communication from taste buds to gustatory nerves. Science (New York, NY) 310(5753):1495–1499
52. Margolskee RF, Dyer J, Kokrashvili Z, Salmon KS, Ilegems E, Daly K et al (2007) T1R3 and gustducin in gut sense sugars to regulate expression of Na+−glucose cotransporter 1. Proc Natl Acad Sci U S A 104(38):15075–15080
53. Shin YJ, Park JH, Choi JS, Chun MH, Moon YW, Lee MY (2010) Enhanced expression of the sweet taste receptors and alpha-gustducin in reactive astrocytes of the rat hippocampus following ischemic injury. Neurochem Res 35(10):1628–1634
54. Shiraishi T (1991) Noradrenergic neurons modulate lateral hypothalamic chemical and electrical stimulation-induced feeding by sated rats. Brain Res Bull 27(3–4):347–351
55. Tong Q, Ye C, McCrimmon RJ, Dhillon H, Choi B, Kramer MD et al (2007) Synaptic glutamate release by ventromedial hypothalamic neurons is part of the neurocircuitry that prevents hypoglycemia. Cell Metab 5(5):383–393
56. Diggs-Andrews KA, Zhang X, Song Z, Daphna-Iken D, Routh VH, Fisher SJ (2010) Brain insulin action regulates hypothalamic glucose sensing and the counterregulatory response to hypoglycemia. Diabetes 59(9):2271–2280
57. Grabauskas G, Song I, Zhou S, Owyang C (2010) Electrophysiological identification of glucose-sensing neurons in rat nodose ganglia. J Physiol 588(Pt 4):617–632
58. Routh VH (2002) Glucose-sensing neurons: Are they physiologically relevant? Physiol Behav 76(3):403–413
59. Sohn JW (2013) Ion channels in the central regulation of energy and glucose homeostasis. Front Neurosci 7:85
60. Silver IA, Erecinska M (1998) Glucose-induced intracellular ion changes in sugar-sensitive hypothalamic neurons. J Neurophysiol 79(4):1733–1745
61. Kurita H, Xu KY, Maejima Y, Nakata M, Dezaki K, Santoso P, Yang Y, Arai T et al (2015) Arcuate Na+,K+-ATPase senses systemic energy states and regulates feeding behavior through glucose-inhibited neurons. Am J Physiol Endocrinol Metab 309(4):E320–E333
62. Fioramonti X, Song Z, Vazirani RP, Beuve A, Routh VH (2011) Hypothalamic nitric oxide in hypoglycemia detection and counterregulation: A two-edged sword. Antioxid Redox Signal 14(3):505–517
63. Ramamurthy S, Ronnett G (2012) AMP-activated protein kinase (AMPK) and energy-sensing in the brain. Exp Neurobiol 21(2): 52–60
64. Ronnett GV, Ramamurthy S, Kleman AM, Landree LE, Aja S (2009) AMPK in the brain: Its roles in energy balance and neuroprotection. J Neurochem 109(Suppl 1):17–23
65. Sanders Matthew J, Grondin Pascal O, Hegarty Bronwyn D, Snowden Michael A, Carling D (2007) Investigating the mechanism for AMP activation of the AMP-activated protein kinase cascade. Biochem J 403(1):139–148
66. Almeida A, Moncada S, Bolanos JP (2004) Nitric oxide switches on glycolysis through the AMP protein kinase and 6-phosphofructo-2-kinase pathway. Nat Cell Biol 6(1):45–51
67. Murphy BA, Fakira KA, Song Z, Beuve A, Routh VH (2009) AMP-activated protein kinase and nitric oxide regulate the glucose sensitivity of ventromedial hypothalamic glucose-inhibited neurons. Am J Physiol Cell Physiol 297(3):C750–C758

68. Zhang J, Xie Z, Dong Y, Wang S, Liu C, Zou MH (2008) Identification of nitric oxide as an endogenous activator of the AMP-activated protein kinase in vascular endothelial cells. J Biol Chem 283(41):27452–27461
69. Canabal DD, Song Z, Potian JG, Beuve A, McArdle JJ, Routh VH (2007) Glucose, insulin, and leptin signaling pathways modulate nitric oxide synthesis in glucose-inhibited neurons in the ventromedial hypothalamus. Am J Physiol Regul Integr Comp Physiol 292(4):R1418–R1428
70. Sayed N, Baskaran P, Ma X, van den Akker F, Beuve A (2007) Desensitization of soluble guanylyl cyclase, the NO receptor, by S-nitrosylation. Proc Natl Acad Sci U S A 104(30):12312–12317
71. Bayliss DA, Sirois JE, Talley EM (2003) The TASK family: Two-pore domain background K+ channels. Mol Interv 3(4):205–219
72. Czirjak G, Enyedi P (2002) Formation of functional heterodimers between the TASK-1 and TASK-3 two-pore domain potassium channel subunits. J Biol Chem 277(7):5426–5432
73. Wyatt CN, Mustard KJ, Pearson SA, Dallas ML, Atkinson L, Kumar P, Peers C, Hardie DG et al (2007) AMP-activated protein kinase mediates carotid body excitation by hypoxia. J Biol Chem 282(11):8092–8098
74. Hayes MR, Skibicka KP, Bence KK, Grill HJ (2009) Dorsal hindbrain 5′-adenosine monophosphate-activated protein kinase as an intracellular mediator of energy balance. Endocrinology 150(5): 2175–2182
75. Burdakov D, Jensen LT, Alexopoulos H, Williams RH, Fearon IM, O'Kelly I, Gerasimenko O, Fugger L et al (2006) Tandem-pore K+ channels mediate inhibition of orexin neurons by glucose. Neuron 50(5):711–722
76. Kim Y, Bang H, Kim D (2000) TASK-3, a new member of the tandem pore K+ channel family. J Biol Chem 275(13):9340–9347
77. Kréneisz O, Benoit JP, Bayliss DA, Mulkey DK (2009) AMP-activated protein kinase inhibits TREK channels. J Physiol 587(Pt 24):5819–5830
78. Arluison M, Quignon M, Nguyen P, Thorens B, Leloup C, Penicaud L (2004) Distribution and anatomical localization of the glucose transporter 2 (GLUT2) in the adult rat brain–an immunohistochemical study. J Chem Neuroanat 28(3):117–136
79. Young JK, McKenzie JC (2004) GLUT2 immunoreactivity in Gomori-positive astrocytes of the hypothalamus. J Histochem Cytochem 52(11):1519–1524
80. Burcelin R, Thorens B (2001) Evidence that extrapancreatic GLUT2-dependent glucose sensors control glucagon secretion. Diabetes 50(6):1282–1289
81. Marty N, Dallaporta M, Foretz M, Emery M, Tarussio D, Bady I, Binnert C, Beermann F et al (2005) Regulation of glucagon secretion by glucose transporter type 2 (glut2) and astrocyte-dependent glucose sensors. J Clin Invest 115(12):3545–3553
82. Leloup C, Orosco M, Serradas P, Nicolaidis S, Penicaud L (1998) Specific inhibition of GLUT2 in arcuate nucleus by antisense oligonucleotides suppresses nervous control of insulin secretion. Brain Res Mol Brain Res 57(2):275–280
83. Dringen R, Gebhardt R, Hamprecht B (1993) Glycogen in astrocytes: Possible function as lactate supply for neighboring cells. Brain Res 623(2):208–214
84. Suh SW, Bergher JP, Anderson CM, Treadway JL, Fosgerau K, Swanson RA (2007) Astrocyte glycogen sustains neuronal activity during hypoglycemia: Studies with the glycogen phosphorylase inhibitor CP-316,819 ([RR*, S*]-5-chloro-N-[2-hydroxy-3-(methoxymethylamino)-3-oxo-1-(phenylmethyl) propyl]-1H-indole-2-carboxamide). J Pharmacol Exp Ther 321(1):45–50
85. Wender R, Brown AM, Fern R, Swanson RA, Farrell K, Ransom BR (2000) Astrocytic glycogen influences axon function and survival during glucose deprivation in central white matter. J Neurosci Off J Soc Neurosci 20(18):6804–6810
86. Pellerin L, Magistretti PJ (1994) Glutamate uptake into astrocytes stimulates aerobic glycolysis: A mechanism coupling neuronal activity to glucose utilization. Proc Natl Acad Sci U S A 91(22): 10625–10629
87. Genc S, Kurnaz IA, Ozilgen M (2011) Astrocyte-neuron lactate shuttle may boost more ATP supply to the neuron under hypoxic conditions–in silico study supported by in vitro expression data. BMC Syst Biol 5:162
88. Bittar PG, Charnay Y, Pellerin L, Bouras C, Magistretti PJ (1996) Selective distribution of lactate dehydrogenase isoenzymes in neurons and astrocytes of human brain. J Cereb Blood Flow Metabol Off J Int Soc Cereb Blood Flow Metabol 16(6):1079–1089
89. Laughton JD, Charnay Y, Belloir B, Pellerin L, Magistretti PJ, Bouras C (2000) Differential messenger RNA distribution of lactate dehydrogenase LDH-1 and LDH-5 isoforms in the rat brain. Neuroscience 96(3):619–625
90. Gerhart DZ, Enerson BE, Zhdankina OY, Leino RL, Drewes LR (1997) Expression of monocarboxylate transporter MCT1 by brain endothelium and glia in adult and suckling rats. Am J Phys 273(1 Pt 1):E207–E213
91. Broer S, Rahman B, Pellegri G, Pellerin L, Martin JL, Verleysdonk S et al (1997) Comparison of lactate transport in astroglial cells and monocarboxylate transporter 1 (MCT 1) expressing Xenopus laevis oocytes. Expression of two different monocarboxylate transporters in astroglial cells and neurons. J Biol Chem 272(48):30096–30102
92. Pellerin L, Bergersen LH, Halestrap AP, Pierre K (2005) Cellular and subcellular distribution of monocarboxylate transporters in cultured brain cells and in the adult brain. J Neurosci Res 79(1–2):55–64
93. Cortes-Campos C, Elizondo R, Llanos P, Uranga RM, Nualart F, Garcia MA (2011) MCT expression and lactate influx/efflux in tanycytes involved in glia-neuron metabolic interaction. PLoS One 6(1):e16411
94. Rinholm JE, Hamilton NB, Kessaris N, Richardson WD, Bergersen LH, Attwell D (2011) Regulation of oligodendrocyte development and myelination by glucose and lactate. J Neurosci Off J Soc Neurosci 31(2):538–548
95. Ainscow EK, Mirshamsi S, Tang T, Ashford MLJ, Rutter GA (2002) Dynamic imaging of free cytosolic ATP concentration during fuel sensing by rat hypothalamic neurones: Evidence for ATP-independent control of ATP-sensitive K(+) channels. J Physiol 544(Pt 2):429–445
96. Walz W, Mukerji S (1988) Lactate release from cultured astrocytes and neurons: A comparison. Glia 1(6):366–370
97. Song Z, Routh VH (2005) Differential effects of glucose and lactate on glucosensing neurons in the ventromedial hypothalamic nucleus. Diabetes 54(1):15–22
98. Borg MA, Tamborlane WV, Shulman GI, Sherwin RS (2003) Local lactate perfusion of the ventromedial hypothalamus suppresses hypoglycemic counterregulation. Diabetes 52(3):663–666
99. Guillod-Maximin E, Lorsignol A, Alquier T, Penicaud L (2004) Acute intracarotid glucose injection towards the brain induces specific c-fos activation in hypothalamic nuclei: Involvement of astrocytes in cerebral glucose-sensing in rats. J Neuroendocrinol 16(5):464–471
100. Bolanos JP, Almeida A (2006) Modulation of astroglial energy metabolism by nitric oxide. Antioxid Redox Signal 8(5–6):955–965
101. Fioramonti X, Marsollier N, Song Z, Fakira KA, Patel RM, Brown S, Duparc T, Pica-Mendez A et al (2010) Ventromedial hypothalamic nitric oxide production is necessary for hypoglycemia detection and counterregulation. Diabetes 59(2):519–528
102. Cidad P, Almeida A, Bolanos JP (2004) Inhibition of mitochondrial respiration by nitric oxide rapidly stimulates cytoprotective

GLUT3-mediated glucose uptake through 5'-AMP-activated protein kinase. Biochem J 384(Pt 3):629–636
103. Bolanos JP, Peuchen S, Heales SJ, Land JM, Clark JB (1994) Nitric oxide-mediated inhibition of the mitochondrial respiratory chain in cultured astrocytes. J Neurochem 63(3):910–916
104. Chan O, Paranjape SA, Horblitt A, Zhu W, Sherwin RS (2013) Lactate-induced release of GABA in the ventromedial hypothalamus contributes to Counterregulatory failure in recurrent hypoglycemia and diabetes. Diabetes 62(12):4239–4246
105. Chan O, Paranjape S, Czyzyk D, Horblitt A, Zhu W, Ding Y, Fan X, Seashore M et al (2011) Increased GABAergic output in the ventromedial hypothalamus contributes to impaired hypoglycemic Counterregulation in diabetic rats. Diabetes 60(5):1582–1589
106. Bolaños JP, Heales SJR (2010) Persistent mitochondrial damage by nitric oxide and its derivatives: Neuropathological implications. Front Neuroenerg 2:1
107. Swanson RA, Choi DW (1993) Glial glycogen stores affect neuronal survival during glucose deprivation in vitro. J Cereb Blood Flow Metabol Off J Int Soc Cereb Blood Flow Metabol 13(1): 162–169
108. Herzog RI, Jiang L, Herman P, Zhao C, Sanganahalli BG, Mason GF, Hyder F, Rothman DL et al (2013) Lactate preserves neuronal metabolism and function following antecedent recurrent hypoglycemia. J Clin Invest 123(5):1988–1998
109. Millán C, Martínez F, Cortés-Campos C, Lizama I, Yañez MJ, Llanos P, Reinicke K, Rodríguez F et al (2010) Glial glucokinase expression in adult and post-natal development of the hypothalamic region. ASN NEURO 2(3):e00035
110. Salgado M, Tarifeno-Saldivia E, Ordenes P, Millan C, Yanez MJ, Llanos P et al (2014) Dynamic localization of glucokinase and its regulatory protein in hypothalamic tanycytes. PLoS One 9(4): e94035
111. Steffens AB, Scheurink AJ, Porte D Jr, Woods SC (1988) Penetration of peripheral glucose and insulin into cerebrospinal fluid in rats. Am J Phys 255(2 Pt 2):R200–R204
112. Shram NF, Netchiporouk LI, Martelet C, Jaffrezic-Renault N, Cespuglio R (1997) Brain glucose: Voltammetric determination in normal and hyperglycaemic rats using a glucose microsensor. Neuroreport 8(5):1109–1112
113. Nigrovic LE, Kimia AA, Shah SS, Neuman MI (2012) Relationship between cerebrospinal fluid glucose and serum glucose. N Engl J Med 366(6):576–578
114. Akmayev IG, Popov AP (1977) Morphological aspects of the hypothalamic-hypophyseal system. VII. The tanycytes: Their relation to the hypophyseal adrenocorticotrophic function. An ultrastructural study. Cell Tissue Res 180(2):263–282
115. Rodriguez EM, Blazquez JL, Pastor FE, Pelaez B, Pena P, Peruzzo B et al (2005) Hypothalamic tanycytes: A key component of brain-endocrine interaction. Int Rev Cytol 247:89–164
116. Cortes-Campos C, Elizondo R, Carril C, Martínez F, Boric K, Nualart F, Garcia-Robles MA (2013) MCT2 expression and lactate influx in anorexigenic and orexigenic neurons of the arcuate nucleus. PLoS One 8(4):e62532
117. Lam TK, Gutierrez-Juarez R, Pocai A, Rossetti L (2005) Regulation of blood glucose by hypothalamic pyruvate metabolism. Science (New York, NY) 309(5736):943–947
118. Robins SC, Stewart I, DE MN, Taylor V, Giachino C, Goetz M et al (2013) Alpha-Tanycytes of the adult hypothalamic third ventricle include distinct populations of FGF-responsive neural progenitors. Nat Commun 4:2049
119. Guadaño-Ferraz A, Obregón MJ, Germain DLS, Bernal J (1997) The type 2 iodothyronine deiodinase is expressed primarily in glial cells in the neonatal rat brain. Proc Natl Acad Sci U S A 94(19): 10391–10396
120. Gonzalez JA, Reimann F, Burdakov D (2009) Dissociation between sensing and metabolism of glucose in sugar sensing neurones. J Physiol 587(1):41–48
121. Tarussio D, Metref S, Seyer P, Mounien L, Vallois D, Magnan C, Foretz M, Thorens B (2014) Nervous glucose sensing regulates postnatal β cell proliferation and glucose homeostasis. J Clin Invest 124(1):413–424
122. Kang L, Sanders NM, Dunn-Meynell AA, Gaspers LD, Routh VH, Thomas AP, Levin BE (2008) Prior hypoglycemia enhances glucose responsiveness in some ventromedial hypothalamic glucosensing neurons. Am J Physiol Regul Integr Comp Physiol 294(3):R784–R792
123. Williams RH, Alexopoulos H, Jensen LT, Fugger L, Burdakov D (2008) Adaptive sugar sensors in hypothalamic feeding circuits. Proc Natl Acad Sci U S A 105(33):11975–11980
124. Colville CA, Seatter MJ, Jess TJ, Gould GW, Thomas HM (1993) Kinetic analysis of the liver-type (GLUT2) and brain-type (GLUT3) glucose transporters in Xenopus oocytes: Substrate specificities and effects of transport inhibitors. Biochem J 290(Pt 3):701–706
125. Burant CF, Bell GI (1992) Mammalian facilitative glucose transporters: Evidence for similar substrate recognition sites in functionally monomeric proteins. Biochemistry 31(42):10414–10420
126. de Vries MG, Arseneau LM, Lawson ME, Beverly JL (2003) Extracellular glucose in rat ventromedial hypothalamus during acute and recurrent hypoglycemia. Diabetes 52(11):2767–2773
127. Silver IA, Erecinska M (1994) Extracellular glucose concentration in mammalian brain: Continuous monitoring of changes during increased neuronal activity and upon limitation in oxygen supply in normo-, hypo-, and hyperglycemic animals. J Neurosci Off J Soc Neurosci 14(8):5068–5076
128. Farrell CL, Pardridge WM (1991) Blood-brain barrier glucose transporter is asymmetrically distributed on brain capillary endothelial lumenal and ablumenal membranes: An electron microscopic immunogold study. Proc Natl Acad Sci U S A 88(13): 5779–5783
129. Leino RL, Gerhart DZ, van Bueren AM, McCall AL, Drewes LR (1997) Ultrastructural localization of GLUT 1 and GLUT 3 glucose transporters in rat brain. J Neurosci Res 49(5):617–626
130. Maher F (1995) Immunolocalization of GLUT1 and GLUT3 glucose transporters in primary cultured neurons and glia. J Neurosci Res 42(4):459–469
131. Nagamatsu S, Sawa H, Kamada K, Nakamichi Y, Yoshimoto K, Hoshino T (1993) Neuron-specific glucose transporter (NSGT): CNS distribution of GLUT3 rat glucose transporter (RGT3) in rat central neurons. FEBS Lett 334(3):289–295
132. Orellana JA, Saez PJ, Cortes-Campos C, Elizondo RJ, Shoji KF, Contreras-Duarte S et al (2012) Glucose increases intracellular free Ca(2+) in tanycytes via ATP released through connexin 43 hemichannels. Glia 60(1):53–68
133. Frayling C, Britton R, Dale N (2011) ATP-mediated glucosensing by hypothalamic tanycytes. J Physiol 589(Pt 9):2275–2286
134. Guillemain G, Loizeau M, Pincon-Raymond M, Girard J, Leturque A (2000) The large intracytoplasmic loop of the glucose transporter GLUT2 is involved in glucose signaling in hepatic cells. J Cell Sci 113(Pt 5):841–847
135. Stolarczyk E, Guissard C, Michau A, Even PC, Grosfeld A, Serradas P, Lorsignol A, Pénicaud L et al (2010) Detection of extracellular glucose by GLUT2 contributes to hypothalamic control of food intake. Am J Physiol Endocrinol Metab 298(5): E1078–E1087
136. Stolarczyk E, Le Gall M, Even P, Houllier A, Serradas P, Brot-Laroche E et al (2007) Loss of sugar detection by GLUT2 affects glucose homeostasis in mice. PLoS One 2(12):e1288

137. Kang J, Kang N, Lovatt D, Torres A, Zhao Z, Lin J, Nedergaard M (2008) Connexin 43 hemichannels are permeable to ATP. J Neurosci Off J Soc Neurosci 28(18):4702–4711
138. Routh VH, Hao L, Santiago AM, Sheng Z, Zhou C (2014) Hypothalamic glucose sensing: Making ends meet. Front Syst Neurosci 8:236
139. Moriguchi T, Sakurai T, Nambu T, Yanagisawa M, Goto K (1999) Neurons containing orexin in the lateral hypothalamic area of the adult rat brain are activated by insulin-induced acute hypoglycemia. Neurosci Lett 264(1–3):101–104
140. Cai XJ, Evans ML, Lister CA, Leslie RA, Arch JR, Wilson S et al (2001) Hypoglycemia activates orexin neurons and selectively increases hypothalamic orexin-B levels: Responses inhibited by feeding and possibly mediated by the nucleus of the solitary tract. Diabetes 50(1):105–112
141. Paxinos G, Franklin KB (2001) The mouse brain in stereotaxic coordinates. Academic Press, San Diego
142. Coll AP, Yeo GS (2013) The hypothalamus and metabolism: Integrating signals to control energy and glucose homeostasis. Curr Opin Pharmacol 13(6):970–976
143. Mountjoy PD, Bailey SJ, Rutter GA (2007) Inhibition by glucose or leptin of hypothalamic neurons expressing neuropeptide Y requires changes in AMP-activated protein kinase activity. Diabetologia 50(1):168–177
144. Fioramonti X, Contie S, Song Z, Routh VH, Lorsignol A, Penicaud L (2007) Characterization of glucosensing neuron subpopulations in the arcuate nucleus: Integration in neuropeptide Y and pro-opio melanocortin networks? Diabetes 56(5):1219–1227
145. Stanley BG, Leibowitz SF (1984) Neuropeptide Y: Stimulation of feeding and drinking by injection into the paraventricular nucleus. Life Sci 35(26):2635–2642
146. Ollmann MM, Wilson BD, Yang YK, Kerns JA, Chen Y, Gantz I, Barsh GS (1997) Antagonism of central melanocortin receptors in vitro and in vivo by agouti-related protein. Science (New York, NY) 278(5335):135–138
147. Tong Q, Ye C-P, Jones JE, Elmquist JK, Lowell BB (2008) Synaptic release of GABA by AgRP neurons is required for normal regulation of energy balance. Nat Neurosci 11(9):998–1000
148. Spanswick D, Smith MA, Mirshamsi S, Routh VH, Ashford ML (2000) Insulin activates ATP-sensitive K+ channels in hypothalamic neurons of lean, but not obese rats. Nat Neurosci 3(8):757–758
149. Hu J, Jiang L, Low MJ, Rui L (2014) Glucose rapidly induces different forms of excitatory synaptic plasticity in hypothalamic POMC neurons. PLoS One 9(8):e105080
150. Park YB, Choi YJ, Park SY, Kim JY, Kim SH, Song DK, Won KC, Kim YW (2011) ATP-sensitive potassium channel-deficient mice show hyperphagia but are resistant to obesity. Diabetes Metabol J 35(3):219–225
151. Vaisse C, Clement K, Guy-Grand B, Froguel P (1998) A frameshift mutation in human MC4R is associated with a dominant form of obesity. Nat Genet 20(2):113–114
152. Huszar D, Lynch CA, Fairchild-Huntress V, Dunmore JH, Fang Q, Berkemeier LR, Gu W, Kesterson RA et al (1997) Targeted disruption of the melanocortin-4 receptor results in obesity in mice. Cell 88(1):131–141
153. Yamanaka A, Beuckmann CT, Willie JT, Hara J, Tsujino N, Mieda M, Tominaga M, Yagami KI et al (2003) Hypothalamic orexin neurons regulate arousal according to energy balance in mice. Neuron 38(5):701–713
154. Burdakov D, Gerasimenko O, Verkhratsky A (2005) Physiological changes in glucose differentially modulate the excitability of hypothalamic melanin-concentrating hormone and orexin neurons in situ. J Neurosci Off J Soc Neurosci 25(9):2429–2433
155. Shiuchi T, Haque MS, Okamoto S, Inoue T, Kageyama H, Lee S, Toda C, Suzuki A et al (2009) Hypothalamic orexin stimulates feeding-associated glucose utilization in skeletal muscle via sympathetic nervous system. Cell Metab 10(6):466–480
156. Hara J, Yanagisawa M, Sakurai T (2005) Difference in obesity phenotype between orexin-knockout mice and orexin neuron-deficient mice with same genetic background and environmental conditions. Neurosci Lett 380(3):239–242
157. Shimada M, Tritos NA, Lowell BB, Flier JS, Maratos-Flier E (1998) Mice lacking melanin-concentrating hormone are hypophagic and lean. Nature 396(6712):670–674
158. Segal-Lieberman G, Bradley RL, Kokkotou E, Carlson M, Trombly DJ, Wang X, Bates S, Myers MG et al (2003) Melanin-concentrating hormone is a critical mediator of the leptin-deficient phenotype. Proc Natl Acad Sci U S A 100(17):10085–10090
159. Karlsson C, Zook M, Ciccocioppo R, Gehlert DR, Thorsell A, Heilig M, Cippitelli A (2012) Melanin-concentrating hormone receptor 1 (MCH1-R) antagonism: Reduced appetite for calories and suppression of addictive-like behaviors. Pharmacol Biochem Behav 102(3):400–406
160. de Araujo IE, Oliveira-Maia AJ, Sotnikova TD, Gainetdinov RR, Caron MG, Nicolelis MA et al (2008) Food reward in the absence of taste receptor signaling. Neuron 57(6):930–941
161. Domingos AI, Sordillo A, Dietrich MO, Liu ZW, Tellez LA, Vaynshteyn J et al (2013) Hypothalamic melanin concentrating hormone neurons communicate the nutrient value of sugar. elife 2:e01462
162. Schwartz MW, Woods SC, Porte D Jr, Seeley RJ, Baskin DG (2000) Central nervous system control of food intake. Nature 404(6778):661–671
163. Cone RD, Cowley MA, Butler AA, Fan W, Marks DL, Low MJ (2001) The arcuate nucleus as a conduit for diverse signals relevant to energy homeostasis. Int J Obes Relat Metabol Disord J Int Assoc Stud Obes 25(Suppl 5):S63–S67
164. Hervey GR (1959) The effects of lesions in the hypothalamus in parabiotic rats. J Physiol 145(2):336–52.3
165. Teitelbaum P, Epstein AN (1962) The lateral hypothalamic syndrome: Recovery of feeding and drinking after lateral hypothalamic lesions. Psychol Rev 69:74–90
166. Lytle LD, Campbell BA (1975) Effects of lateral hypothalamic lesions on consummatory behavior in developing rats. Physiol Behav 15(3):323–331
167. Ahima RS, Antwi DA (2008) Brain regulation of appetite and satiety. Endocrinol Metab Clin N Am 37(4):811–823
168. Cui H, Sohn J-W, Gautron L, Funahashi H, Williams KW, Elmquist JK, Lutter M (2012) Neuroanatomy of melanocortin-4 receptor pathway in the lateral hypothalamic area. J Comp Neurol 520(18):4168–4183
169. Thornton SN, Nicolaidis S, Larue-Achagiotis C, Campfield A (1991) Body weight gain after VMH lesions in adult female rats guanethidine-sympathectomized at birth. Appetite 17(1):47–53
170. Meek TH, Nelson JT, Matsen ME, Dorfman MD, Guyenet SJ, Damian V, Allison MB, Scarlett JM et al (2016) Functional identification of a neurocircuit regulating blood glucose. Proc Natl Acad Sci U S A 113(14):E2073–E2082
171. Kamitakahara A, Xu B, Simerly R (2016) Ventromedial hypothalamic expression of Bdnf is required to establish normal patterns of afferent GABAergic connectivity and responses to hypoglycemia. Mol Metabol 5(2):91–101
172. Chan O, Zhu W, Ding Y, McCrimmon RJ, Sherwin RS (2006) Blockade of GABA(A) receptors in the ventromedial hypothalamus further stimulates glucagon and sympathoadrenal but not the hypothalamo-pituitary-adrenal response to hypoglycemia. Diabetes 55(4):1080–1087

173. Fan X, Chan O, Ding Y, Zhu W, Mastaitis J, Sherwin R (2015) Reduction in SGLT1 mRNA expression in the ventromedial hypothalamus improves the counterregulatory responses to hypoglycemia in recurrently hypoglycemic and diabetic rats. Diabetes 64(10):3564–3572
174. Bellinger LL, Bernardis LL (2002) The dorsomedial hypothalamic nucleus and its role in ingestive behavior and body weight regulation: Lessons learned from lesioning studies. Physiol Behav 76(3):431–442
175. Wamsteeker Cusulin JI, Fuzesi T, Watts AG, Bains JS (2013) Characterization of corticotropin-releasing hormone neurons in the paraventricular nucleus of the hypothalamus of Crh-IRES-Cre mutant mice. PLoS One 8(5):e64943
176. Kadar A, Sanchez E, Wittmann G, Singru PS, Fuzesi T, Marsili A et al (2010) Distribution of hypophysiotropic thyrotropin-releasing hormone (TRH)-synthesizing neurons in the hypothalamic paraventricular nucleus of the mouse. J Comp Neurol 518(19):3948–3961
177. Arase K, York DA, Shimizu H, Shargill N, Bray GA (1988) Effects of corticotropin-releasing factor on food intake and brown adipose tissue thermogenesis in rats. Am J Phys 255(3 Pt 1): E255–E259
178. Choi YH, Hartzell D, Azain MJ, Baile CA (2002) TRH decreases food intake and increases water intake and body temperature in rats. Physiol Behav 77(1):1–4
179. Leibowitz SF, Hammer NJ, Chang K (1981) Hypothalamic paraventricular nucleus lesions produce overeating and obesity in the rat. Physiol Behav 27(6):1031–1040
180. Kalsbeek A, La Fleur S, Van Heijningen C, Buijs RM (2004) Suprachiasmatic GABAergic inputs to the paraventricular nucleus control plasma glucose concentrations in the rat via sympathetic innervation of the liver. J Neurosci Off J Soc Neurosci 24(35): 7604–7613
181. Geerling JC, Shin J-W, Chimenti PC, Loewy AD (2010) Paraventricular hypothalamic nucleus: Axonal projections to the brainstem. J Comp Neurol 518(9):1460–1499
182. Paxinos G, Watson C (1998) The rat brain in stereotaxic coordinates, 4th edn. Academic Press, San Diego
183. Yang L, Scott KA, Hyun J, Tamashiro KL, Tray N, Moran TH, Bi S (2009) Role of dorsomedial hypothalamic neuropeptide Y in modulating food intake and energy balance. J Neurosci Off J Soc Neurosci 29(1):179–190
184. Adachi A, Kobashi M, Funahashi M (1995) Glucose-responsive neurons in the brainstem. Obes Res 3(Suppl 5):735s–740s
185. Buller K, Xu Y, Dayas C, Day T (2001) Dorsal and ventral medullary catecholamine cell groups contribute differentially to systemic interleukin-1beta-induced hypothalamic pituitary adrenal axis responses. Neuroendocrinology 73(2):129–138
186. Day TA, Sibbald JR (1989) A1 cell group mediates solitary nucleus excitation of supraoptic vasopressin cells. Am J Phys 257(5 Pt 2):R1020–R1026
187. Mussa BM, Verberne AJ (2013) The dorsal motor nucleus of the vagus and regulation of pancreatic secretory function. Exp Physiol 98(1):25–37
188. Miller AD, Leslie RA (1994) The area postrema and vomiting. Front Neuroendocrinol 15(4):301–320
189. Verberne AJ, Sartor DM (2010) Rostroventrolateral medullary neurons modulate glucose homeostasis in the rat. Am J Physiol Endocrinol Metab 299(5):E802–E807
190. Verberne AJ, Sabetghadam A, Korim WS (2014) Neural pathways that control the glucose counterregulatory response. Front Neurosci 8:38
191. Taborsky GJ Jr, Mundinger TO (2012) Minireview: The role of the autonomic nervous system in mediating the glucagon response to hypoglycemia. Endocrinology 153(3):1055–1062
192. Buijs RM, Chun SJ, Niijima A, Romijn HJ, Nagai K (2001) Parasympathetic and sympathetic control of the pancreas: A role for the suprachiasmatic nucleus and other hypothalamic centers that are involved in the regulation of food intake. J Comp Neurol 431(4):405–423
193. ter Horst GJ, Luiten PG (1986) The projections of the dorsomedial hypothalamic nucleus in the rat. Brain Res Bull 16(2):231–248
194. Sim LJ, Joseph SA (1991) Arcuate nucleus projections to brainstem regions which modulate nociception. J Chem Neuroanat 4(2):97–109
195. Canteras NS, Simerly RB, Swanson LW (1994) Organization of projections from the ventromedial nucleus of the hypothalamus: A Phaseolus vulgaris-leucoagglutinin study in the rat. J Comp Neurol 348(1):41–79
196. Tarussio D, Metref S, Seyer P, Mounien L, Vallois D, Magnan C, Foretz M, Thorens B (2014) Nervous glucose sensing regulates postnatal beta cell proliferation and glucose homeostasis. J Clin Invest 124(1):413–424
197. Ross CA, Ruggiero DA, Reis DJ (1985) Projections from the nucleus tractus solitarii to the rostral ventrolateral medulla. J Comp Neurol 242(4):511–534
198. Dunn-Meynell AA, Rawson NE, Levin BE (1998) Distribution and phenotype of neurons containing the ATP-sensitive K+ channel in rat brain. Brain Res 814(1–2):41–54
199. Allen GV, Cechetto DF (1992) Functional and anatomical organization of cardiovascular pressor and depressor sites in the lateral hypothalamic area: I. Descending projections. J Comp Neurol 315(3):313–332
200. Shafton AD, Ryan A, Badoer E (1998) Neurons in the hypothalamic paraventricular nucleus send collaterals to the spinal cord and to the rostral ventrolateral medulla in the rat. Brain Res 801(1–2):239–243
201. Ritter S, Dinh TT, Li AJ (2006) Hindbrain catecholamine neurons control multiple glucoregulatory responses. Physiol Behav 89(4): 490–500
202. Madden CJ, Stocker SD, Sved AF (2006) Attenuation of homeostatic responses to hypotension and glucoprivation after destruction of catecholaminergic rostral ventrolateral medulla neurons. Am J physiol Regul Integr Comp Physiol 291(3):R751–R759
203. Puskas N, Papp RS, Gallatz K, Palkovits M (2010) Interactions between orexin-immunoreactive fibers and adrenaline or noradrenaline-expressing neurons of the lower brainstem in rats and mice. Peptides 31(8):1589–1597
204. Unger RH, Zhou YT, Orci L (1999) Regulation of fatty acid homeostasis in cells: Novel role of leptin. Proc Natl Acad Sci U S A 96(5):2327–2332
205. Nogueiras R, Tschöp MH, Zigman JM (2008) CNS regulation of energy metabolism: Ghrelin versus leptin. Ann N Y Acad Sci 1126:14–19
206. Routh VH, Song Z, Liu X (2004) The role of glucosensing neurons in the detection of hypoglycemia. Diabetes Technol Ther 6(3):413–421
207. Biggers DW, Myers SR, Neal D, Stinson R, Cooper NB, Jaspan JB, Williams PE, Cherrington AD et al (1989) Role of brain in counterregulation of insulin-induced hypoglycemia in dogs. Diabetes 38(1):7–16
208. Frizzell RT, Jones EM, Davis SN, Biggers DW, Myers SR, Connolly CC, Neal DW, Jaspan JB et al (1993) Counterregulation during hypoglycemia is directed by widespread brain regions. Diabetes 42(9):1253–1261
209. Fenselau H, Campbell JN, Verstegen AM, Madara JC, Xu J, Shah BP et al (2017) A rapidly acting glutamatergic ARC–>PVH satiety circuit postsynaptically regulated by alpha-MSH. Nat Neurosci 20(1):42–51
210. Wu Q, Clark MS, Palmiter RD (2012) Deciphering a neuronal circuit that mediates appetite. Nature 483(7391):594–597

211. Wu Q, Boyle MP, Palmiter RD (2009) Loss of GABAergic signaling by AgRP neurons to the parabrachial nucleus leads to starvation. Cell 137(7):1225–1234
212. Herbert H, Moga MM, Saper CB (1990) Connections of the parabrachial nucleus with the nucleus of the solitary tract and the medullary reticular formation in the rat. J Comp Neurol 293(4):540–580
213. Yamanaka A, Kunii K, Nambu T, Tsujino N, Sakai A, Matsuzaki I, Miwa Y, Katsutoshi Goto et al (2000) Orexin-induced food intake involves neuropeptide Y pathway. Brain Res 859(2):404–409
214. Ma X, Zubcevic L, Bruning JC, Ashcroft FM, Burdakov D (2007) Electrical inhibition of identified anorexigenic POMC neurons by orexin/hypocretin. J Neurosci Off J Soc Neurosci 27(7):1529–1533
215. Muroya S, Funahashi H, Yamanaka A, Kohno D, Uramura K, Nambu T, Shibahara M, Kuramochi M et al (2004) Orexins (hypocretins) directly interact with neuropeptide Y, POMC and glucose-responsive neurons to regulate Ca 2+ signaling in a reciprocal manner to leptin: Orexigenic neuronal pathways in the mediobasal hypothalamus. Eur J Neurosci 19(6):1524–1534
216. Lanfray D, Arthaud S, Ouellet J, Compère V, Do Rego J-L, Leprince J et al (2013) Gliotransmission and brain glucose sensing: Critical role of endozepines. Diabetes 62(3):801–810
217. de Mateos-Verchere JG, Leprince J, Tonon MC, Vaudry H, Costentin J (2001) The octadecaneuropeptide [diazepam-binding inhibitor (33-50)] exerts potent anorexigenic effects in rodents. Eur J Pharmacol 414(2–3):225–231
218. do Rego JC, Orta MH, Leprince J, Tonon MC, Vaudry H, Costentin J (2007) Pharmacological characterization of the receptor mediating the anorexigenic action of the octadecaneuropeptide: Evidence for an endozepinergic tone regulating food intake. Neuropsychopharmacol Off Publ Am Coll Neuropsychopharmacol 32(7):1641–1648
219. Laukkanen O, Lindstrom J, Eriksson J, Valle TT, Hamalainen H, Ilanne-Parikka P et al (2005) Polymorphisms in the SLC2A2 (GLUT2) gene are associated with the conversion from impaired glucose tolerance to type 2 diabetes: The Finnish Diabetes Prevention Study. Diabetes 54(7):2256–2260
220. Muller YL, Piaggi P, Hoffman D, Huang K, Gene B, Kobes S, Thearle MS, Knowler WC et al (2014) Common genetic variation in the glucokinase gene (GCK) is associated with type 2 diabetes and rates of carbohydrate oxidation and energy expenditure. Diabetologia 57(7):1382–1390
221. Namkoong C, Kim MS, Jang PG, Han SM, Park HS, Koh EH, Lee WJ, Kim JY et al (2005) Enhanced hypothalamic AMP-activated protein kinase activity contributes to hyperphagia in diabetic rats. Diabetes 54(1):63–68