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REGULATION OF GLUCOSE METABOLISM BY CENTRAL INSULIN ACTION

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Insulin has been known to act on the hypothalamus, in particular the arcuate nucleus, in the central nervous system. Such central insulin action is not only involved in the regulation of energy metabolism via the regulation of food intake and heat production, but also plays an important role in glucose metabolism by regulating hepatic glucose production and glucose uptake by skeletal muscles. Studies on the intracerebroventricular administration of PI-3K inhibitors or sulfonylureas have demonstrated that hyperpolarization of agouti-related protein neurons induced by the activation of PI-3K signaling/K_{ATP} channels in the hypothalamic arcuate nucleus plays an important role in the suppression of hepatic glucose production mediated by central insulin action. Cutting of the vagus nerve overrides the suppression of hepatic glucose production by intracerebroventricular insulin administration, which suggests the involvement of autonomic nerves in central insulin action in the liver. The central insulin action-mediated suppression of hepatic glucose production is associated with decreased gene expression of enzymes involved in hepatic gluconeogenesis, and both increased interleukin-6 expression in hepatic non-parenchymal cells induced by central insulin action and associated activation of hepatic STAT3 play an important role in the suppression of gene expression of hepatic gluconeogenesis-related enzymes. In animal models of obesity and insulin resistance, the central insulin action-mediated hepatic glucose production control mechanism is impaired in both the hypothalamus and liver. Increased hepatic gluconeogenesis in obesity and type-2 diabetes has been attributed to impaired hepatic insulin signaling and increased expression of enzymes involved in hepatic gluconeogenesis due to hyperglycemia, but may also be partially attributed to the impairment of the central insulin action-mediated suppression of hepatic gluconeogenesis. ***Biomed Rev*** **2011; 22: 31-39.**

Key words: central nervous system, gluconeogenesis, glucose, insulin, STAT3 signaling

INTRODUCTION

Homeostasis of energy metabolism is maintained by the close interaction between the central nervous system (CNS) and peripheral tissues. In response to changes such as food intake, a stimulus is sent from peripheral tissues to the CNS to regulate energy intake, and this is followed by the transmission of a command from the CNS to the peripheral tissues to regulate energy metabolism, such as increasing heat production (1, 2). In such CNS-peripheral tissue interactions, a variety of humoral

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factors play an important role especially in the transmission of information from peripheral tissues to the CNS. Previous studies have demonstrated that factors such as leptin, adiponectin, both secreted by adipose tissue, and glucagon like-peptide 1 secreted by intestinal tissue, play an important role in energy metabolism through their action on the CNS (3-5). Insulin, a potent regulator of glucose metabolism acting on the liver, skeletal muscle, and adipose tissue, has also been shown to regulate energy metabolism through its action on the CNS. Recent studies have also demonstrated that insulin action on the CNS not only regulates energy metabolism via the regulation of food intake and heat production, but is also involved in the regulation of glucose metabolism (6,7). These observations indicate that insulin regulates glucose metabolism via both the direct action on peripheral organs and the indirect regulation through its action on the CNS. The purpose of this article is to discuss the role of central insulin action in the regulation of glucose metabolism, with a focus on its recently-discovered action on the regulation of hepatic glucose production.

CENTRAL INSULIN ACTION AND ENERGY METABOLISM

Studies using ^{125}I-labeled insulin have demonstrated that circulating insulin acts on limited areas of the brain such as the hypothalamus, in particular the arcuate nucleus, and the choroid plexus (8-10). In particular, insulin action in the hypothalamic arcuate nucleus, which has both insulin and leptin receptors (11), plays an important role in the regulation of energy metabolism (Fig. 1). Brain endothelial cells are connected to each other by tight junctions to form the blood-brain barrier (BBB), which inhibits the entry of molecules larger than 400 Da into the CNS (12). Several mechanisms of how insulin, a molecule of some 5800 Da, is transported into the brain have been reported and include diffusion from periventricular regions lacking the BBB, such as the median eminence near the arcuate nucleus (13), and the involvement of insulin receptor-mediated transport mechanisms in the BBB (12). Studies using hyperinsulinemic euglycemic clamps have shown that an increase in insulin level in cerebrospinal fluid is limited, even in the presence of a significantly high level of

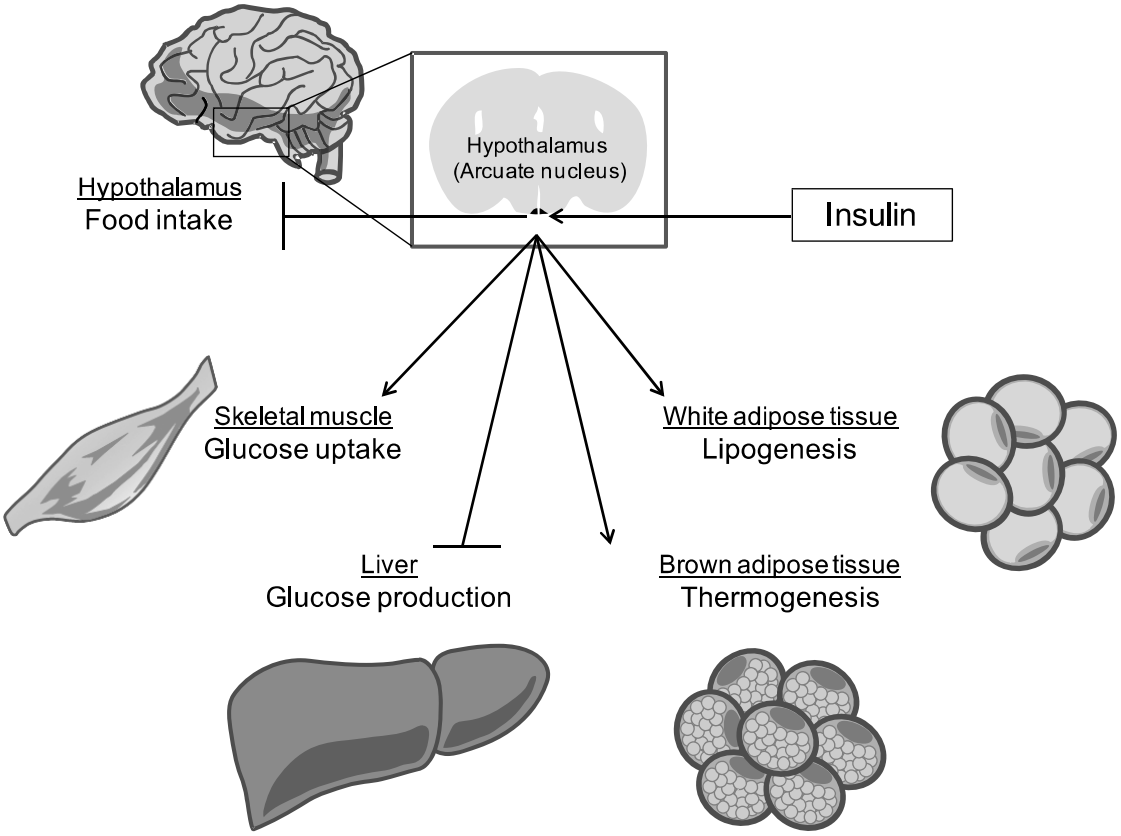

Figure 1. Central insulin action and energy metabolism. *Insulin acts on the hypothalamus, especially the arcuate nucleus. Hypothalamic insulin action suppresses food intake, induces glucose uptake in the muscle, decreases hepatic glucose production, and increases thermogenesis in brown adipose tissue and lipogenesis in white adipose tissue.*

circulating insulin, indicating the saturation of insulin transport into the CNS. These observations suggest the involvement of not only simple diffusion, but also carrier-mediated transport mechanisms in the saturable transport of insulin into the CNS (14-16).

Central insulin action plays an important role in food intake regulation. In a study using monkeys, intracerebroventricular administration of insulin led to decreases in food intake and body weight (17). Rat studies have also demonstrated similar effects following the intracerebroventricular administration of insulin or low-molecular weight compounds that can activate the insulin receptor (1,18). The neuron-specific insulin receptor knockout (NIRKO) mouse is an animal model of impaired central insulin action (19). The female NIRKO mouse exhibits a significant increase in food intake, and both males and females increase adipose tissue and body weight and develop insulin resistance (19). Such central insulin action-mediated regulation of food intake has been attributed to an action on neuropeptide tyrosine (NPY)/agouti-related protein (AgRP) neurons, which mediate orexigenic effects, and pro-opiomelanocortin (POMC)/cocaine-amphetamine-regulated transcript (CART) neurons, which mediate the anorexic effects in the hypothalamus. Intracerebroventricular administration of insulin leads to decreased expression of NPY and increased expression of POMC, resulting in suppressed food intake (1,6,20). When insulin binds to its receptor, it transmits information to cells by activating the phosphoinositide-3-kinase (PI3-K) and mitogen-activated protein kinase/extracellular-regulated kinase (MAPK) signaling pathways (21). The PI3-K signaling pathway has been shown to play an important role in the suppression of food intake mediated by insulin signaling in the CNS. Actually, studies have demonstrated that the anorexic effect of intracerebroventricular insulin administration can be inhibited by intracerebroventricular pretreatment with a PI3-K inhibitor (22). Leptin, which acts on the hypothalamus, suppresses food intake, and regulates energy metabolism, is reported to activate the hypothalamic PI3-K signaling pathway (23). Leptin regulates the expression of AgRP and POMC through activation of the PI3-K signaling pathway and it also regulates the expression of signal transducer and activator of transcription-3 (STAT3) signaling (24). STAT3-dependent regulation of AgRP and POMC expression is suppressed by competitive binding of each transcriptional promoter by Forkhead box O1 (FoxO1) (25), which is inhibited by the activation of PI3-K signaling (26, 27). Given that insulin does not activate hypothalamic STAT3, insulin regulates AgRP and POMC expression via inactivation of hypothalamic FoxO1 by PI-3K signaling (25,28). Central insulin action also plays an important role in increasing heat production following food intake via the regulation of sympathetic activity. The dietary control of sympathetic activity is critically mediated by insulin action in the ventromedial hypothalamus (VMH) (29). This finding is supported by the observation that the destruction of the VMH by the administration of gold thioglucose leads to suppression of heat production following food intake and impaired regulation of sympathetic activity (29, 30). Studies have suggested the involvement of hypothalamic PI-3K and MAPK in the regulation of sympathetic activity mediated by central insulin action. In particular, regulation of sympathetic stimulation of brown adipocytes, a major source of heat production in mice, has been suggested to be critically mediated by the activation of hypothalamic MAPK (31).

The involvement of central insulin action has also been suggested in the regulation of lipogenesis in white adipose tissue (32). A 7-day continuous intracerebroventricular insulin administration with an osmotic pump resulted in increased adipose tissue weight and increased expression of lipoprotein lipase in adipose tissue (32). However, the detailed role of central insulin action in white adipose tissue has not been elucidated. These observations indicate that insulin plays an important role in the homeostasis of energy metabolism by decreasing food intake and increasing heat production through its action on the hypothalamus.

CENTRAL INSULIN ACTION REGULATES GLUCOSE METABOLISM

There is an increasing amount of evidence to suggest that central insulin action plays an important role in the regulation of glucose metabolism as well as energy metabolism. Obici and colleagues developed rats deficient in hypothalamic insulin receptors by intracerebroventricular administration of antisense oligonucleotides and found that these rats exhibited insulin resistance (33). A study using hyperinsulinemic euglycemic clamps has shown that insulin resistance produced by knocking down hypothalamic insulin receptors is not associated with impaired glucose uptake in muscles and fat tissues but is attributed to impaired suppression of glucose production; in other words, increased glucose production in the liver (33). The NIRKO mouse, which also exhibits insulin resistance, has been shown to exhibit increased hepatic glucose production in a hyperinsulinemic euglycemic clamp study (34). Another study has demonstrated that the intracerebroventricu-

lar administration of insulin in rat or mouse models leads to suppressed glucose production in the liver (35,36). Central insulin action, acting mainly on the hypothalamic arcuate nucleus, has been shown to regulate hepatic glucose production *via* the vagus nerve (33). Cutting of the vagal hepatic branch has been demonstrated to result in reduced hepatic glucose production following intracerebroventricular administration of insulin (36). These studies suggest that the central insulin action regulates glucose metabolism through the suppression of hepatic glucose production via the vagal nerve. It has been reported that central insulin action activates hepatic glycogen synthesis (37). Considering that hepatic glycogen synthase is known to be activated by hepatic vagal nerve activation (38), central insulin action also regulates hepatic glycogen synthesis via the vagal nerve.

Hyperinsulinemic euglycemic clamp studies with insulin administration at high concentrations are used to examine in detail the effect of insulin on glucose uptake by peripheral organs, including skeletal muscles (39). A recent study using the euglycemic clamp with high-dose insulin administration has shown that the inhibition of central insulin action does not alter glucose uptake by adipose tissue, but reduces glucose uptake by skeletal muscles (40). Increased synthesis of glycogen in skeletal muscles has also been observed following intracerebroventricular administration of insulin in mice (41). Thus, the central insulin action may also play a certain role in glucose metabolism in skeletal muscles.

The PI-3K signaling pathway in the hypothalamic arcuate nucleus plays an important role in the regulation of glucose metabolism mediated by central insulin action. In rats, the intracerebroventricular administration of a PI-3K inhibitor to inhibit central insulin action leads to a decrease in insulin-dependent hypoglycemic response and an increase in glucose production in the liver (35,42). The use of an adenovirus vector to induce overexpression of a constitutively active mutant of Akt, which is activated by PI-3K, in the arcuate nucleus results in an enhanced hypoglycemic response to insulin with no change in food intake (42). These findings suggest that activation of the PI-3K signaling pathway in the CNS, especially in the hypothalamic arcuate nucleus, leads to suppressed glucose production in the liver and thus plays a certain role in the hypoglycemic response to insulin.

The K_{ATP} channels in the hypothalamus have been shown to play an important role in the suppression of hepatic glucose production mediated by the central insulin action. The intracerebroventricular administration of K_{ATP} channel blocker sulfonylurea can reverse the suppression of hepatic glucose production by central insulin action (35). Insulin has been shown to induce hyperpolarization of hypothalamic neurons by opening K_{ATP} channels via PI-3K (43). This is further supported by the fact that insulin opens K_{ATP} channels in both AgRP and POMC neurons and induces hyperpolarization of these neurons (44,45). It has also been demonstrated that MAPK inhibitors do not affect insulin-induced, hypothalamic K_{ATP} channel-dependent hyperpolarization (43). AgRP neuron-specific insulin receptor-deficient mice exhibited increased hepatic glucose production, whereas in POMC neuron-specific insulin receptor-deficient mice, insulin successfully suppressed glucose production in the liver (45). These results suggest that hyperpolarization and inactivation of AgRP neurons, resulting from the activation of PI-3K/K_{ATP} channels, play an important role in the suppression of hepatic glucose production mediated by the central insulin action.

The involvement of central insulin action has also been suggested in the regulation of glucose uptake by skeletal muscles, but its mechanism has been unclear (40). Leptin, which also acts on K_{ATP} channels in the hypothalamus (43), has been shown to increase glucose uptake by skeletal muscles by activating 5'-AMP-activated protein kinase (AMPK) via the sympathetic nervous system (46), and the hypothalamic administration of leptin has been shown to increase glucose uptake by adipose tissue (47). Intracerebroventricular administration of sulfonylurea in a euglycemic clamp study with a high-concentration insulin infusion resulted in increased hepatic glucose production and reduced glucose uptake by skeletal muscles due to blockage of central insulin action by K_{ATP} channel blockade (40). At the same time, no change in glucose uptake was observed in adipose tissue or the heart, and no significant change in AMPK activity was observed in skeletal muscles (40). These results suggest the regulation of glucose metabolism in skeletal muscles mediated by central insulin action has different mechanisms than the regulation of glucose metabolism in skeletal muscles mediated by central leptin action.

As it becomes increasingly clear how the central insulin action regulates hepatic glucose metabolism, studies have suggested that the contribution of this mechanism varies depending on the type of animal model. A dog model has been developed where insulin is infused from the carotid or vertebral artery to selectively increase the level of insulin circulating in the head compared with the peripheral insulin level (48). This model, where the insulin level in the head was selectively increased to a level 4 times higher than that achieved by the

standard hyperinsulinemic euglycemic clamp, demonstrated the same level of suppression of hepatic glucose production as the standard hyperinsulinemic euglycemic clamp. This finding indicates that the central insulin action may exert a milder suppressive effect on hepatic glucose production in dogs than in mice and rats.

MECHANISM OF CENTRAL INSULIN ACTION-MEDIATED REGULATION OF HEPATIC GLUCOSE PRODUCTION

Hepatic glucose production is composed of glycogenolysis and gluconeogenesis. Central insulin action-mediated suppression of hepatic glucose production has been attributed primarily to a decrease in gluconeogenesis (36). Hepatic gluconeogenesis is strongly regulated by the expression of the genes of related metabolic enzymes, and the central insulin action suppresses gene expression of enzymes involved in hepatic gluconeogenesis, such as phosphoenolpyruvate carboxykinase and glucose-6-phosphatase. Gene expression of hepatic gluconeogenic enzymes is regulated by a number of transcription factors, including cyclic-AMP response element binding protein (CREB) and FoxO1 (49). Insulin decreases the gene expression of hepatic gluconeogenic enzymes by the inhibition of transcriptional activity of CREB and FoxO1 (50-53). Central insulin action also suppresses hepatic gluconeogenic gene expression as described above, and hepatic STAT3 plays an important role as a transcription factor involved in this suppression by central insulin action (34). Hepatic STAT3 suppresses gene expression of enzymes involved in hepatic gluconeogenesis when activated by interleukin-6 (IL-6) (54,55). This is further supported by liver-specific STAT3-deficient mice that develop insulin resistance as the gene expression of hepatic gluconeogenic enzymes increases, leading to glucose intolerance under the condition of obesity induced by high-fat diet feeding (54). Activation of hepatic STAT3 induced by the central insulin action has been demonstrated to be due to increased secretion of IL-6 from hepatic non-parenchymal cells (34). However, the role of the vagus nerve in hepatic STAT3 activation by central insulin action has not been fully elucidated. These observations indicate that insulin inhibits the expression of hepatic gluconeogenic genes by the direct action on the hepatocyte inhibiting CREB and FoxO1 activity and by central insulin action activating hepatic STAT3 (Fig. 2).

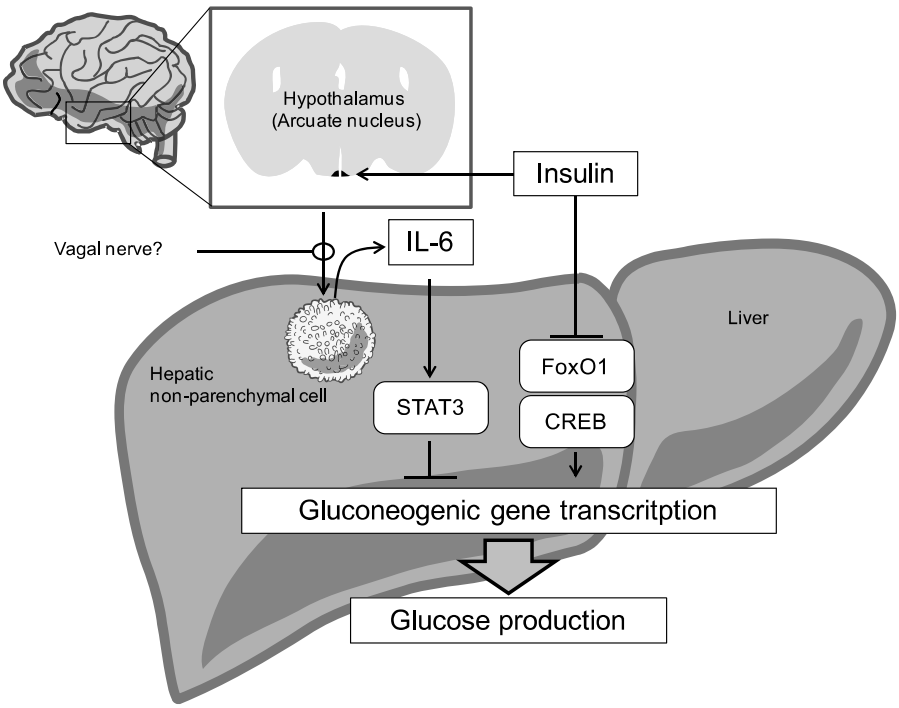

Figure 2. Direct and indirect regulation of hepatic gluconeogenic gene expression by insulin. *Insulin inhibits the transcriptional activity of FoxO1 and CREB and suppresses hepatic gluconeogenesis in a PI3-K signaling-dependent manner in hepatocytes. In addition to the direct effect on hepatocytes, central insulin action increases IL-6 expression in non-parenchymal hepatic cells, which activates hepatic STAT3 in a paracrine manner and decreases expression of hepatic gluconeogenic enzyme genes.*

Species difference has been suggested regarding the central insulin action-mediated suppression of hepatic glucose production, as evidenced by the lack of such suppressive mechanisms in dogs (37, 48). However, the central insulin action-mediated activation of hepatic STAT3 has been shown to be preserved in dogs as well as in mice (37, 56). This suggests that the contribution of hepatic STAT3 to the regulation of gene/protein expression of hepatic gluconeogenic enzymes varies among species.

CENTRAL INSULIN ACTION-MEDIATED REGULATION OF HEPATIC GLUCOSE PRODUCTION AND TYPE 2 DIABETES

Increased hepatic gluconeogenesis has been observed in obese individuals with type-2 diabetes (57). The question then arises as to the role of the central insulin action-mediated suppression of hepatic gluconeogenesis in the presence of obesity and insulin resistance. In a rat study, maintaining a high level of peripheral blood insulin did not lead to a substantial increase in central insulin action, probably due to saturation of the transport mechanisms (1). This suggests that an increased blood insulin level associated with insulin resistance does not lead to a substantial increase in insulin action in the CNS. With regard to insulin action in the hypothalamus, a study using obese Zucker rats with leptin receptor abnormalities has demonstrated that insulin fails to induce hyperpolarization of hypothalamic neurons in these rats (43). It has also been demonstrated that the intake of obesity-inducing high-fat diets, even short-term intake, induces activation of S6 kinase in the hypothalamus and thereby inhibits insulin signaling (58). A study using db/db leptin receptor-deficient obese mice has shown that the activation of hepatic STAT3, an effector of the central insulin action, is inhibited by endoplasmic reticulum stress associated with insulin resistance in obesity (59). This is further supported by the reversal of both the central insulin action-mediated suppression of glucose production in the liver and the promotion of glucose uptake by skeletal muscles in obesity model mice bred with a high-fat diet (40). Increased hepatic gluconeogenesis in type-2 diabetes has been attributed to impaired suppression of hepatic glucose production due to impaired hepatic insulin signaling and increased expression of enzymes involved in hepatic gluconeogenesis due to increased activity of CREB associated with hyperglycemia (60), but may also be partially attributed to the impairment of the central insulin action-mediated suppression of hepatic gluconeogenesis.

CONCLUSION

Studies have demonstrated that the central insulin action regulates not only food intake and energy metabolism, but also glucose metabolism via the regulation of hepatic glucose production. Hepatic glucose production plays a key role in glucose metabolism, as evidenced by the observation that increased hepatic glucose production leads to impaired glucose tolerance (61). Thus, hepatic glucose production is controlled by insulin's direct action on the liver and indirectly via its action on the CNS. The fact that various animal models of impaired hepatic insulin signaling exhibit profound glucose intolerance indicates that the direct action of insulin on the liver plays a central role in the homeostasis of glucose metabolism. At the same time, insulin resistance exhibited by CNS insulin receptor-deficient mice (19) and liver-specific STAT3-deficient mice (50) suggests a role of the central insulin action-mediated regulation of hepatic glucose production in glucose metabolism.

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