Mechanisms of glucose sensing and multiplicity of glucose sensors

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INTRODUCTION

Increases in blood glucose concentrations are detected by pancreatic β cells which then secrete appropriate amounts of insulin to normalize blood glucose concentrations. A decreased capability of these endocrine cells to sense glucose is a characteristic of type 2 diabetes. It is therefore an important goal of basic research to elucidate the glucose signalling mechanisms which control the secretion of insulin. This signalling pathway depends on glucose metabolism and requires the presence of specific molecules such as the glucose transporter GLUT2, glucokinase and the KATP channel subunits Kir6.2 and SUR1. Even though the pancreatic β cells may be a critical glucose sensor in the control glucose homeostasis, the secretory activity of these cells is also controlled by other glucose sensitive cells, which modulate the β cell activity through endocrine or nervous signals. These other glucose sensors include endocrine cells from the gut and which secrete the insulinotropic hormones GIP and GLP-1 following glucose absorption, but also cells controlling the activity of the autonomic nervous system which control in a positive (parasympathetic) or negative (sympathetic) way insulin secretion.

Here, we will discuss recent physiological studies performed in my laboratory and which explored, by physiological evaluation of genetically modified mice, the role of GLUT2 and of the gluco-incretin hormone receptors, in the mechanisms of gluco-detection not only by pancreatic β cells but also by the hepatoportal sensor which controls peripheral glucose utilization and the first peak of insulin secretion.

PANCREATIC β CELL GLUCOSE DETECTION

Glucose-stimulated insulin secretion (GSIS) is initiated by glucose uptake by GLUT2 and phosphorylation by glucokinase. Glucose-6-phosphate is then metabolized through the glycolytic pathway and activates mitochondrial metabolism to generate an increase in the cytoplasmic ATP/ADP concentration ratio [18]. This leads to closure of KATP-channels, depolarization of the plasma membrane and opening of voltage-sensitive Ca++ channels. The entry of Ca++ then triggers exocytosis of the insulin granules. A KATP-channel-independent pathway has also been described in which glucose stimulates insulin secretion even when the KATP channel is maintained open by diazoxide and the cells depolarized by extracellular K+ [13]. Both the KATP channel-dependent and -independent pathways require energy production from glucose and the first steps in glucose utilisation is its uptake by GLUT2 and phosphorylation by glucokinase [12].

INACTIVATION OF THE GLUT2 GENE SUPPRESSES GLUCOSE-STIMULATED INSULIN SECRETION

Inactivation of the Glut2 gene in the mouse by homologous recombination generated mice with a diabetic-like phenotype and which died at 2-3 weeks of age [14]. Islet perfusion experiments revealed that the absence of GLUT2 expression caused a markedly suppressed glucose-stimulated insulin secretion. The absence of GLUT2 also suppressed glucose stimulation of gene transcription and insulin mRNA translation.

Thus, as expected, suppressing glucose entry in the β cells suppresses glucose sensing. To evaluate whether GLUT2 was also controlling glucose detection by other glucose sensors, we rescued the Glut2-/- mice by transgenic reexpression specifically in the ⍺-cell of a glucose transporter (GLUT1) under the control of the rat insulin promoter. These mice (RipGlut1Glut2-/-) grew normally, could breed and their islets displayed normal GSIS [31].

THE HEPATOPORTAL GLUCOSE SENSOR

A glucose sensor is located in the portal vein region [16] and is connected through afferent branches of
the vagus nerve to glucose sensitive neurons in the lateral hypothalamus [26, 28] and in the nucleus of the solitary tract [1]. This sensor is probably activated in the absorptive phase; experimentally it can be activated by direct portal glucose infusion. The hepatoportal glucose sensor controls many physiological functions, including an induction of anorexia [25], a stimulation of hepatic glucose uptake [11] and an inhibition of counterregulation induced by peripheral hypoglycemia [7, 15].

We showed that activation of the hepatoportal sensor by portal glucose infusion induced a slow development of hypoglycemia, which reached a value of approximately 2.5 mM [5]. This effect was caused by a stimulation of glucose uptake by peripheral tissues. Using different experimental protocols and genetically modified mice, in particular the RipGlut1Glut2-/− mice, we presented evidence that glucose detection by the hepatoportal sensor was dependent on the presence of GLUT2 and was inhibited by somatostatin, two characteristics shared with the β cell glucose sensor [4]. We further demonstrated that this sensor required the presence of the GLP-1 receptor to be glucose competent, as this sensor could not be activated in Gip-1R-/− mice or in control mice infused in the portal vein with the GLP-1R antagonist exendin-(9-39) [3].

We also showed that the effect of this sensor on stimulation of glucose uptake and induction of hypoglycemia required expression in muscle of the GLUT4 transporter, but not of the insulin receptor and that a dominant negative form of the AMP-kinase could block this effect [2].

ROLE OF THE GLUCOINCRETINS IN THE CONTROL OF THE GLUCOSE STIMULATED INSULIN SECRETION

The glucokinetic hormones GIP and GLP-1 are secreted by gut endocrine cells following nutrient absorption, in particular glucose [6, 9], and strongly potentiate glucose-stimulated insulin secretion by pancreatic β cells [21, 35], an action which requires binding of the hormones to specific β cell G-coupled receptors that activate the adenyl cyclase pathway [30, 33]. Studies in rodents have further described that GLP-1 can increase β cell mass, probably through a combination of transcriptional activation of key genes involved in β cell differentiation and function, such as Pdx-1, Glut2, glucokinase and insulin [8, 29, 32, 34, 36] and by a protection against apoptosis [17].

The physiological importance of the gluco-incretin receptors has been evaluated in gene knockout mice. GipR-/− mice showed impaired tolerance to oral glucose load but not to intraperitoneal glucose injection [20]. High fat feeding for three weeks induced a marked increase in the glycemic excursions following meal absorption. However, inactivation of this receptor gene protected mice from high fat diet-induced obesity and transfer of this mutation in ob/ob mice reduced their body weight [19]. Glp-1R-/− mice showed a marked intolerance to oral glucose absorption but only a mild intolerance to intraperitoneal glucose injection [27]. However, absence of GLP-1 receptor was compensated by upregulation of both GIP plasma levels and GIP action on pancreatic islets. Also, lack of GIP receptor led to higher sensitivity to GLP-1 in pancreatic islets [10, 22, 23]. Thus, inactivation of each receptor caused only mild impairment in whole body glucose. We therefore generated mice with inactivation of both receptor genes and evaluated glucose homeostasis and insulin secretion.

Our study [24] demonstrated that both gluco-incretin hormones have an additive effect in the stimulation of insulin secretion following oral glucose absorption. This is due to their known insulinotropic activity, which depends on the presence of their cognate receptors on β cells. Interestingly, we also showed that absence of both receptors led to glucose intolerance in female mice injected intraperitoneally with glucose, i.e., when gluco-incretin secretion is not stimulated. As no impairment in insulin sensitivity could be detected, this indicated a defect in insulin secretion. Measurement of insulin during the ipGTT revealed two defects, an absence of first phase insulin secretion in the Glp-1R-/− and double-KO mice and a reduced plasma insulin level at 30 minutes in the single receptor knockout mice. The absence of first phase insulin secretion could not be explained by a β cell defect since perfusion of islets from Glp-1R-/− mice showed perfectly normal kinetics of insulin secretion, indicating that the defect is present only when studied in vivo. We could then demonstrate that this was due to a defect in the function of the hepatoportal glucose sensor. Thus, in vivo control of first phase insulin secretion may not depend on glucose reaching the pancreatic β cells but may be controlled by other sensors, in particular that of the portal vein region.

We further showed that the absence of both, but not of single, gluco-incretin receptors, induced a β cell-autonomous secretion defect. This consisted in a 50 percent reduction in the stimulated insulin secretion, due to a defect in the glucose signaling pathway distal to plasma membrane depolarization. Our data suggest that this defect is due to changes in the expression of genes regulating the cAMP-dependent stimulation of insulin secretion.
CONCLUSION

Glucose sensing is a complex mechanism which involves the interplay of several proteins controlling glucose metabolism, mitochondrial function, ion channels and regulated secretion of insulin granules. These intricate mechanisms are further modulated by hormones and nervous signals directly affecting β cell functions. There are however other glucose sensors which are located at distinct anatomical sites and which indirectly regulate insulin secretion. Our studies have revealed, using genetically modified, mice that similar proteins participate to both the β cell and extra-pancreatic glucose sensors. This is the case for the glucose transporter GLUT2 and for the GLP-1 receptor. These studies have also led to the uncovering of the role of the hepatoportal sensor in for the GLP-1 receptor. These studies have also led to the uncovering of the role of the hepatoportal sensor in controlling the control of first phase insulin secretion, as assessed in vivo. Finally, we provided evidence that the preservation of the β cell glucose sensitivity also requires the presence of both the GIP and GLP-1 receptor which probably acts by regulating the expression of genes involved in maintaining the fully differentiated state of the β cells.

RÉFÉRENCES


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