Insulin biosynthesis: The IREny of it all

Recent studies have shown that the unfolded protein response (UPR) is essential for the survival of insulin-producing β cells. Work in this issue of *Cell Metabolism* (Lipson et al., 2006) identifies a novel role for UPR activation in β cell function by demonstrating that the UPR effector IRE1 is a positive regulator of glucose-stimulated proinsulin biosynthesis.

Pancreatic islets of Langerhans are responsible for the homeostatic control of glucose levels, maintaining them in a narrow range of 5–5.5 mM. This is accomplished by the secretion of endocrine hormones such as insulin and glucagon by β cells and α cells, respectively. Diabetes, and its attendant dysregulated glucose levels, represents a family of polygenic disorders caused by the absence of insulin production (autoimmune or type 1 diabetes), insufficient production of insulin, and/or inhibition of insulin action at peripheral tissues such as fat, liver, and muscle (type 2 diabetes). While the release of endocrine hormones is generally regulated by neuronal, nutritional, and hormonal signals, the primary regulatory signal for the secretion and synthesis of insulin by β cells is considered to be glucose. The production and release of insulin by β cells is tightly controlled by transcriptional, translational, and posttranslational mechanisms. A paper in the current issue of *Cell Metabolism* (Lipson et al., 2006) provides evidence supporting a novel role for the endoplasmic reticulum-resident protein kinase inositol-requiring enzyme (IRE) 1α in the selective regulation of postprandial biosynthesis of insulin by β cells.

The unfolded protein response (UPR) is an evolutionarily conserved cellular response that allows cells to survive the accumulation of unfolded proteins in the ER. This UPR is tightly regulated by the activity of three signal transducers, IRE1, PERK, and ATF6 (Harding et al., 2002; Zhang and Kaufman, 2006). The resident ER chaperone BiP is thought to be the primary regulator of the UPR transducers (Zhang and Kaufman, 2006). In unstressed cells, BiP is bound to each of the signaling transducers; however, in response to ER stress or protein overload, BiP is released, allowing it to assist in the proper folding of ER proteins. This also allows IRE1 and PERK to be activated by homodimerization and autophosphorylation. The activation of ATF6 occurs in the golgi, where it is cleaved to an active transcription factor that translocates to the nucleus.

The three signal transducers have different modes of action. Activated PERK attenuates general protein synthesis by phosphorylating the transcriptional elongation factor eIF2α and participates in the transcriptional activation of UPR-associated genes (Harding et al., 2002; Zhang and Kaufman, 2006). IRE1 contains not only a kinase domain but also an endoribonuclease domain that when activated mediates the site-specific splicing of a 26 base intron of X-box protein-1 (XBP-1). This splice produces a frameshift in XBP-1 mRNA leading to the synthesis of a potent transcription factor that regulates the expression of various genes involved in the degradation of unfolded proteins, or proper protein folding (Harding et al., 2002; Zhang and Kaufman, 2006).

β cells are highly sensitive to alterations in the activity of the UPR. Mutations in PERK have been identified in Wolcott-Rallison syndrome, an autosomal recessive disease characterized by a number of disorders, including diabetes that is due to selective dystrophy of β cells (Delepine et al., 2000). PERK-deficient mice develop diabetes by 2–4 weeks of age due to progressive β cell loss (Harding et al., 2001; Zhang et al., 2002), and mice expressing a Ser51Ala mutation at the PERK phosphorylation site in eIF2α show a loss of β cell mass in utero (Scheuner et al., 2001). While these studies provide evidence that UPR, or more specifically PERK, signaling is essential for β cell survival, few studies have examined the role of IRE1 in regulating pancreatic β cell function or survival.

Lipson et al. now show that glucose stimulates IRE1α phosphorylation in isolated mouse islets and insulinoma cells in a concentration-dependent fashion that correlates with the stimulatory effects of glucose on insulin secretion. While IRE1α phosphorylation correlates with glucose-stimulated insulin secretion, IRE1α depletion using siRNA or expression of a kinase-dead IRE1 mutant fails to modify the stimulatory actions of glucose on insulin secretion or insulin transcription by INS-1 cells. Instead, IRE1α appears to selectively regulate the translation of insulin, as proinsulin biosynthesis is significantly reduced while total protein synthesis is unaffected in INS-1 cells depleted of IRE1α by siRNA knockdown. These novel findings suggest that IRE1α may function as a positive and selective regulator of insulin biosynthesis in response to glucose challenge. These findings are in contrast to the more global inhibitory actions of PERK-mediated eIF2α phosphorylation as a negative regulator of protein translation (Zhang and Kaufman, 2006).

IRE1 possesses both a kinase and ribonuclease activity that are activated following its dissociation from BiP. While glucose stimulates IRE1α phosphorylation, it fails to stimulate XBP-1 splicing (ribonuclease activity), the dissociation of IRE1α from BiP, or the activation of JNK, a downstream target whose activation is associated with cell death under conditions of prolonged UPR activation (Urano et al., 2000). This novel form of IRE1α regulation, which the authors term “Stimulus-Coupling Adaptation to ER Folding” or SCAEF, may serve as a physiological regulator of specialized secretory cells types such as β cells, which produce specific proteins such as insulin. This form of IRE1α regulation also differs from the more classical activation under conditions of severe ER stress wherein IRE1 dissociates from BiP resulting in the activation of both ribonuclease and kinase activity. While others have shown that it is possible to activate BiP-associated IRE1 under conditions of mild ER stress (Credle et al., 2005), the current findings describe a physiological regulatory mechanism by which the selective regulation of IRE1α kinase activity participates in a specific cellular function, in this case insulin biosynthesis. In contrast to the transient and selective activation of IRE1α kinase activity in response to elevated
concentrations of glucose that occurs in response to an acute challenge, chronic exposure of β cells to elevated levels of glucose (25 mM) for 3 and 7 days results in a severe stress that is associated with the activation of both IRE1α kinase and ribonuclease activity. These chronic glucose exposure conditions are known to stimulate β cell death by a process termed glucotoxicity (Leahy, 2005; Robertson et al., 2004). These findings suggest that the normal physiological response of IRE1α in β cells to an acute glucose challenge is the selective activation of its kinase activity and the positive regulation of proinsulin biosynthesis (Figure 1). However, under pathophysiological conditions or severe stress as observed during chronic exposure of β cells to elevated levels of glucose, IRE1α kinase and endoribonuclease activity is stimulated, and this may contribute to β cell failure (or glucotoxicity).

These provocative findings of Lipson et al. suggest that IRE1α may function as a positive regulator of insulin biosynthesis, yet the mechanisms responsible for glucose-stimulated IRE1α kinase activity remain unclear. Glucose-induced IRE1α phosphorylation is attenuated by 2-deoxyglucose, suggesting that glucose metabolism is required for IRE1α activation. However, IRE1α is also phosphorylated in response to secretagogues that function by directly depolarizing β cells (arginine and KCl) and by agents that augment insulin secretion via cAMP (GLP-1). Intriguingly, this suggests the existence of multiple mechanisms of IRE1α activation, which may include roles for elevations in intracellular calcium as well as the activation of cAMP-dependent pathways. Arginine, KCl, and glucose (via metabolism and closure of ATP channels) stimulate insulin secretion by depolarizing β cells, allowing for calcium entry and calcium-mediated exocytosis. A speculated mechanism might include a hypothetical (novel, or existing) kinase capable of coupling intracellular information provided by secretagogue stimulation of β cells (i.e., calcium or cAMP accumulation) with the phosphorylation of IRE1α and the induction of proinsulin biosynthesis. Future studies designed to identify the mechanisms by which this diverse and mechanistically distinct series of secretagogues selectively activate IRE1α kinase activity independent of its ribonuclease activity are likely to provide new insights into additional cellular and physiological functions of IRE1 and alternative mechanisms by which IRE1 may be regulated under conditions of ER stress.

Figure 1. Differential regulation of IRE1α in pancreatic β cells in response to an acute or chronic glucose exposure

In response to an acute exposure (1 hr) to stimulatory concentrations of glucose, IRE1α remains bound to BiP; however, it is phosphorylated and functions to stimulate proinsulin translation. In contrast, in response to a chronic exposure (3–7 days) to elevated levels of glucose, IRE1α is fully activated as it is likely released from BiP, is phosphorylated, and stimulates XBP-1 splicing. Chronic exposure to elevated levels of glucose results in β cell death or glucotoxicity.

John A. Corbett
Saint Louis University School of Medicine
1402 South Grand Boulevard
St. Louis, Missouri 63104

Selected reading


DOI 10.1016/j.cmet.2006.08.007