Type 2 Diabetes—a Matter of β-Cell Life and Death?

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In type 2 diabetes, the β cells of the pancreas fail to produce enough insulin to meet the body's demand, in part because of an acquired decrease in β-cell mass. In adults, pancreatic β-cell mass is controlled by several mechanisms, including β-cell replication, neogenesis, hypertrophy, and survival. Here, I discuss evidence supporting the notion that increased β-cell apoptosis is an important factor contributing to β-cell loss and the onset of type 2 diabetes. Interestingly, a key signaling molecule that promotes β-cell growth and survival, insulin receptor substrate 2 (IRS-2), is a member of a family of proteins whose inhibition contributes to the development of insulin resistance in the liver and other insulin-responsive tissues. Thus, the IRS-2 pathway appears to be a crucial participant in the tenuous balance between effective pancreatic β-cell mass and insulin resistance.

Concurrent with the obesity epidemic, the incidence of type 2 diabetes is increasing at an alarming rate (*1*). Type 2 diabetes arises when the endocrine pancreas fails to secrete sufficient insulin to cope with the metabolic demand (*2*, *3*), because of acquired β-cell secretory dysfunction and/or decreased β-cell mass. Insulin secretory dysfunction in type 2 diabetes is well documented and has been reviewed elsewhere (*4*, *5*). Whether insulin secretory dysfunction is a cause or consequence of the disease is still debated, but there is mounting evidence that it may be symptomatic of changes in β-cell mass (*3*, *6*). Although proposed nearly 50 years ago (*7*), the hypothesis that β-cell loss plays an important role in the pathogenesis of type 2 diabetes has only recently come to the fore (*3*). β-cell mass in the adult is plastic, and adjustments in β-cell growth and survival maintain a balance between insulin supply and metabolic demand. For example, obese individuals who do not develop diabetes exhibit an increase in β-cell mass that appears to compensate for the increased metabolic load and obesity-associated insulin resistance. However, this β-cell adaptation eventually fails in the subset of obese individuals who develop type 2 diabetes (*2*, *7–12*). Indeed, most individuals with type 2 diabetes, whether obese or lean, show a net decrease in β-cell mass (*8*). Thus, type 2 diabetes is a disease of relative insulin deficiency.

Given the pivotal role of β-cell mass in determining whether an individual will progress to type 2 diabetes (*2*, *10*), there is growing interest in understanding the mechanisms that control the life and death of β cells. Here, I outline current concepts of β-cell growth and survival, with an emphasis on one particular cell survival mechanism that is thought to go awry in type 2 diabetes.

Cellular Mechanisms Controlling Adult β-Cell Mass

Pancreatic β-cell mass is regulated by at least four independent mechanisms: (i) β-cell replication (i.e., the mitogenic division of existing β cells), (ii) β-cell size, (iii) β-cell neogenesis [i.e., the emergence of "new β cells" from certain common pancreatic ductal epithelial cells (*8*, *13*, *14*)], and (iv) β-cell apoptosis (*10*, *12*, *13*). The sum of the rates of β-cell replication, size, and neogenesis, minus the rate of β-cell apoptosis gives the net rate of β-cell growth (*12*, *15*). The contribution made by each of these mechanisms is variable and may change at different stages of life or when the β-cell mass adapts to changes in metabolic load. Moreover, the relative contribution of each may be species specific. For example, recent evidence indicates that maintenance of β-cell mass in young adult mice is primarily due to β-cell replication (*16*, *17*), yet in humans both β-cell neogenesis and replication appear to play a role (*8*). Adding further complexity, β-cell neogenesis has been documented in older mice (*18*), an observation that underscores the age dependence of these mechanisms.

Measuring dynamic changes in β-cell mass is technically difficult and subtleties can be overlooked. Markers of cell division, such as Ki-67, correspond to a small transient window in the cell cycle and may underestimate the incidence of β-cell replication. Likewise, apoptotic and necrotic cells are efficiently cleared by macrophages in vivo and so the extent of β-cell apoptosis, especially when analyzed in ex vivo pancreatic sections, may be underappreciated. Pancreatic β-cell neogenesis (often measured as the abundance of insulin-positive cells in the pancreatic ductal epithelium) is relatively rare and its detection requires analysis of multiple pancreatic sections. Moreover, without specific markers for "precursor β cells," it is difficult to judge whether "insulin-positive cells" actually mature into fully differentiated β cells or are alternative cell types that have been misdiagnosed (*19*, *20*). Such studies are particularly challenging in humans, where retrograde analyses from autopsy specimens cannot be done and pancreatic biopsies are difficult to obtain. In addition, pancreatic specimens from patients with type 2 diabetes often represent only the end stages of the disease.

These technical difficulties notwithstanding, a model of postnatal pancreatic β-cell growth in humans is emerging from studies of both humans (*7–9*) and rodents (*21*, *22*) (Figs. 1 and 2). Under normal circumstances, there is a transient burst of β-cell replication just after birth, followed by a transitory rise in β-cell neogenesis (*21*) (Fig. 2). In the later phase of this neonatal burst of β-cell replication and neogenesis, there is also a modest amount of apoptosis that parallels pancreatic islet rearrangement. Because this rate of apoptosis is low, the net effect is a marked increase in β-cell growth early in life. It should be noted that the early burst of β-cell growth has been observed primarily in rodent models. This is obviously difficult to substantiate in newborn humans, although it is generally thought that a similar spurt of human postnatal β-cell growth occurs (*21*). Thereafter, through childhood and adolescence the rates of β-cell replication, neogenesis, and apoptosis drop markedly (Fig. 2). In adults, β cells have an estimated life-span of ~60 days (*13*). Under normal conditions, ~0.5% of adult β cells undergo apoptosis, but this is balanced by β-cell replication and, to a lesser extent, β-cell neogenesis (*2*, *21*). Normally, β-cell size stays relatively constant and, as such, β-cell mass is maintained at an optimal level through most of adulthood (Fig. 2). In the senior years of life, β-cell mass may decrease as apoptosis slightly outweighs β-cell replication and neogenesis (Fig. 2). This may partially explain why the elderly have an increased susceptibility to type 2 diabetes.

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Adaptation of β-Cell Mass to Metabolic Load

During adulthood, the β-cell mass is highly adaptive to changes in metabolic homeostasis. A good example is pregnancy: Rodent studies have shown that the maternal β-cell population can almost double to compensate for the increased metabolic load of a developing fetus (23). Although the evidence is limited, a similar adaptation likely occurs in humans (23). This is achieved primarily by increased β-cell replication driven by the pregnancy hormones prolactin and placental lactogen (23). Postpartum, the rate of β-cell replication decreases and a concomitant increase in β-cell apoptosis ensures a rapid return of β-cell mass to normal levels (21).

More pertinent to this article, however, is that β-cell mass adapts to an increased metabolic load caused by obesity and the inherent insulin resistance. In humans, this increased β-cell mass is thought to occur through an increase in β-cell replication and neogenesis, as well as β-cell hypertrophy (Figs. 1 and 2) (8, 11). A small increase in β-cell apoptosis has also been observed in nondiabetic obesity (8); however, this is outweighed by increases in β-cell replication, neogenesis, and cell size, resulting in a net increase in β-cell mass (Fig. 2). As previously mentioned, the relative contributions of β-cell replication, neogenesis, and size to this compensatory increase in β-cell mass may vary between species and even between different strains of rats and mice (12). Whether humans have a genetically based variability in the adaptive mechanisms that increase β-cell mass remains to be determined.

Failure of β-Cell Mass to Compensate for Metabolic Load

Although there may be an initial compensatory increase in β-cell mass, the onset of type 2 diabetes in both humans and rodent models is accompanied by a progressive decrease in β-cell mass. As a result, the body can no longer adapt to any increases in metabolic load, including insulin resistance associated with obesity. This β-cell loss arises from a marked increase in β-cell apoptosis, which far outweighs modest increases in β-cell replication and neogenesis (Fig. 2) (2, 8, 11, 22). As the type 2 diabetic state progresses, the situation worsens; the incidence of β-cell replication decreases and the β-cell population declines (Fig. 2). In humans, the increased β-cell apoptosis in type 2 diabetes is further exacerbated by the formation of amyloid plaque deposits in

islets (24) (Fig. 1). Eventually, in the most severe cases, a "point of no return" in β-cell mass can be reached and a permanent type 2 diabetic state arises that must be treated with insulin replacement therapy (6).
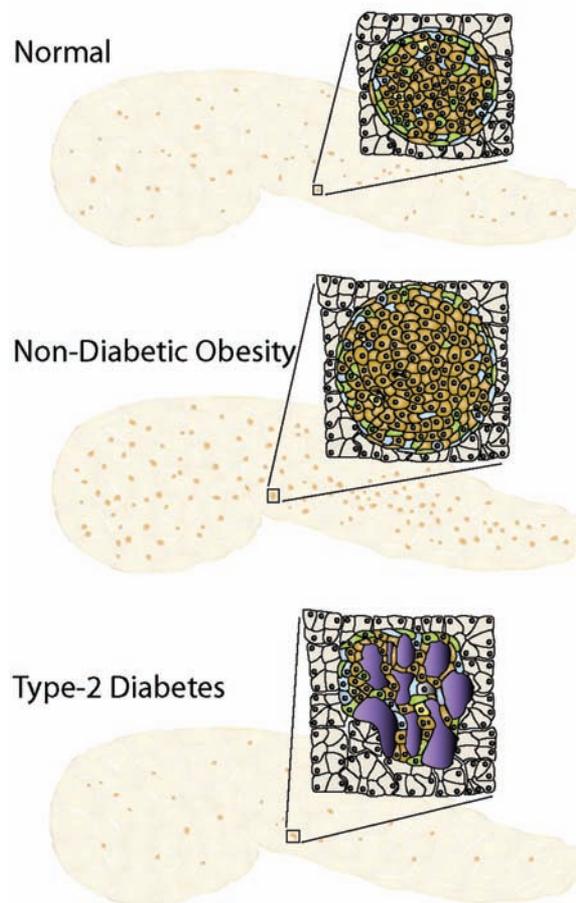
Fig. 1. Adult pancreatic morphology in normal, nondiabetic obesity and type 2 diabetes. The whole pancreas, which is composed primarily of exocrine cells (which produce enzymes that are delivered directly to the gut by way of the pancreatic duct for digestion), retains the same shape and size in all of these conditions. The endocrine islets (which secrete insulin) are more dynamic. Under normal circumstances, these islets are scattered throughout the pancreas and comprise about 1% of the total pancreatic population of cells. In normal islets (inset), the β cells (brown) tend to populate the core of the islet, representing about 70% of the islet endocrine cells. In nondiabetic obesity, the number of islets in the pancreas increases and these islets (inset) tend to be larger, primarily because of increased numbers of β cells per islet or in some instances because of increased β-cell size. As a result, >90% of the islet endocrine cells are β cells. In type 2 diabetes, the number of islets in the pancreas can decrease and these islets (inset) tend to be disorganized and misshapen. There is a marked reduction in the number of β cells per islet and amyloid plaques (purple) can dominate the islet area.

About one-third of obese individuals develop type 2 diabetes. This may be due to a genetic predisposition at the level of the β cell. Consistent with this idea, certain inherited forms of type 2 diabetes, called maturity-onset diabetes of the young (MODY), are

caused by mutations in proteins required for normal β-cell function, such as glucokinase and the transcription factor PDX-1 (25). However, environmental contributions should also be considered. In humans, low birth weight has been associated with an increased risk of developing type 2 diabetes in adulthood (26). In this regard, the surges in β-cell replication and neogenesis early in postnatal life become critical for laying down the baseline population of β cells in adult life (Fig. 2). In a smaller neonate, a proportionally smaller baseline β-cell mass may be acquired in the adult (12). A limited adult β-cell mass could compromise the body's capacity to compensate for an increased metabolic load, as in pregnancy or obesity, which in turn may contribute to an increased risk of gestational diabetes or earlier onset of obesity-linked type 2 diabetes (12).

The Role of IRS-2 Signaling in β-Cell Survival

Many mechanisms could trigger the increase in β-cell apoptosis that occurs during the pathogenesis of type 2 diabetes (2, 3, 11, 27). Among them are the development of endoplasmic reticulum (ER) stress, chronic hyperglycemia, chronic hyperlipidemia, oxidative stress, and certain cytokines (2, 3, 11, 27). Here, I consider how certain circulating factors that are elevated in obesity-linked diabetes might disrupt signal transduction pathways that promote normal β-cell turnover and survival. I focus on factors that affect expression levels of IRS-2 because IRS-2 is especially potent in promoting β-cell survival (28) and because dampening of IRS-2 signaling leads to insulin resistance as well as β-cell apoptosis.

Members of the IRS protein family are intracellular tyrosine kinase substrates that act as signaling interfaces immediately downstream of cell surface receptors, such as the insulin and insulin-like growth factor-1 (IGF-1) receptors (29). Deletion of IRS-1 and IRS-2 leads to marked insulin resistance, indicating that these genes play a key role in insulin action (30–33). Once an IRS molecule is tyrosine phosphorylated, certain signaling proteins selectively dock by means of their SH2-domains to specific IRS-phosphotyrosine sites, resulting in activation of downstream signaling pathways. Examples include the phosphatidylinositol-3′-kinase (PI3′K)/protein kinase-B (PKB, also known as Akt) pathway and the Ras pathway that leads to activation of

the mitogen-activated protein (MAP) kinases Erk-1 and Erk-2 (29). Both IRS-1 and IRS-2 are expressed in pancreatic β-cells. However, IRS-1 is not involved in the control of β-cell mass (30), but instead appears to function in cellular Ca^{2+} homeostasis (34). In contrast, IRS-2 plays a critical role in regulation of β-cell growth (30, 33, 35). Increased IRS-2 expression can promote β-cell replication, neogenesis, and survival (12, 18, 28), whereas decreased IRS-2 expression causes spontaneous β-cell apoptosis (30, 35, 36). Thus, IRS-2 is critically important for maintaining β-cell mass, especially by promoting β-cell survival (12, 28). It follows then that mechanisms leading to suppression of IRS-2 expression in β-cells may be linked to the increased incidence of β-cell apoptosis and consequently the onset of type 2 diabetes (29, 37).

What mechanisms might contribute to the loss of IRS-2 expression in β cells? Although experimental evidence is limited, some interesting parallels can be drawn to mechanisms by which insulin resistance develops in insulin-responsive tissues, such as skeletal muscle and the liver. Tyrosine phosphorylation of IRS-2 leads to increased β-cell growth and survival (2). However, IRS-2 contains multiple sites for serine/threonine phosphorylation, and these, for the most part, have a negative effect on IRS signal transduction by promoting IRS degradation (37, 38). Several mechanisms relevant to the pathogenesis of type-2 diabetes could potentially increase IRS-2 serine/threonine phosphorylation, subsequently resulting in IRS-2 ubiquitination, proteosomal degradation, and ultimately β-cell apoptosis (Fig. 3).

Chronic hyperglycemia can lead to chronic activation of the nutrient-sensing serine/threonine protein kinase, mammalian Target of Rapamycin (mTOR) in β cells. Activated mTOR triggers serine/threonine phosphorylation of IRS-2 and its subsequent proteosomal degradation, leading to increased β-cell apoptosis (39). It should be noted, however, that chronic hyperglycemia can also trigger β-cell apoptosis by additional mechanisms, collectively referred to as "glucotoxicity." These mechanisms include the generation of potentially damaging reactive-oxygen species (ROS) as a consequence of chronically increased glucose metabolism in β cells (40); chronic elevation of intra-

cellular $[Ca^{2+}]$ to cytotoxic levels (3); a marked up-regulation in the synthesis of β-cell secretory granule proteins, including pro-insulin and pro-Islet Amyloid Associated Peptide (proIAPP), which in turn could promote ER stress (3, 41); and a glucose-induced increase in local interleukin-1β (IL-1β) production (42). Some of these "metabolic stresses," such as dangerously high levels of ROS and $[Ca^{2+}]$ in the β cell, may activate the Jnk/p38

Fig. 2. A hypothetical model for postnatal pancreatic β-cell growth in humans. Three conditions are depicted: normal (black), nondiabetic obesity (green), and type 2 diabetes (red). The upper panel shows changes in the total β-cell mass. This reflects the sum of changes in β-cell replication, β-cell neogenesis, and β-cell size, minus the incidence of β-cell apoptosis, depicted individually in the other four panels. The changes in β-cell mass, replication, neogenesis, size, and apoptosis are in arbitrary units that reflect the magnitude of change from birth onward.

stress protein kinases in β cells (43), in turn leading to serine/threonine phosphorylation of IRS-2 and its degradation (37, 44).

Hyperlipidemia, another feature of obesity-linked type 2 diabetes, can lead to abnormal accumulation of lipids in β cells with detrimental effects, including fatty acid–induced β-cell apoptosis (11, 45). Fatty acids, through the production of intracellular long chain acyl-CoA, can activate a novel class of protein kinase C isoforms (nPKC) indepen-

dent of $[Ca^{2+}]$ (46). Chronic activation of certain nPKCs can lead to serine/threonine phosphorylation of IRS molecules (38, 47) that in β cells would promote IRS-2 degradation and ultimately lead to β-cell apoptosis (37) (Fig. 3). As with chronic hyperglycemia, this is only one possible mechanism by which chronic accumulation of lipids in the β cell can contribute to fatty acid–induced β-cell apoptosis, commonly known as "lipotoxicity" (11).

Other lipotoxic mechanisms include the generation of ceramide from palmitate, which induces a mitochondrial apoptotic pathway (11); inhibition of PKB (45); and fatty acid/ceramide–induced activation of the c-Jun N-terminal kinase (Jnk)/p38 stress kinase pathway (48). Interestingly, Jnk/p38-mediated phosphorylation of IRS-2 may also be induced by hyperglycemia, underscoring a likely interplay between glucotoxicity and lipotoxicity mechanisms in β-cell apoptosis (49).

A variety of cytokines play a role in the pathogenesis of type 2 diabetes. The discovery that there is local induction of IL-1β production within islets in response to chronic glucose implies that IL-1β plays a role in inducing β-cell apoptosis in type 2 diabetes (42), as well as in type 1 diabetes (27, 50). In obesity-linked diabetes, certain adipocyte-derived cytokines are elevated in the circulation, including leptin, tumor necrosis factor α (TNFα), and IL-6. Intriguingly, leptin has recently been shown to modulate IL-1β–induced apoptosis in human β cells (51). Some of these cytokines can induce β-cell apoptosis through induction of signaling pathways that activate the transcription factor NFκB (27). However, they may also activate signaling pathways that trigger increased degradation of IRS-2. Certain cytokines, such as leptin, IL-6, and IFN-γ activate the Janus Kinase-2/Signal Transducer and Activator of Transcription (JAK/STAT) post-receptor signaling pathway. This leads to increased expression of Suppressor of Cytokine Signaling-1 (SOCS-1) and SOCS-3 proteins, which normally bind to the leptin, IL-6, and IFN-γ receptors and inhibit JAK-2/STAT signaling. SOCS-1 and SOCS-3 have also been shown to bind to the C terminus of IRS molecules, leading to their ubiquitination and subsequent degradation (52, 53). Thus, it is conceivable that leptin and/or IL-6 may cause β-cell apoptosis by decreasing β-cell IRS-2 levels through a similar mechanism. IL-1β and TNFα promote

activation of the protein kinase IκK (*27*). IκK phosphorylates the cytosolic inhibitory protein IκB, resulting in release and activation of NFκB (*27*). However, IκK has also been shown to phosphorylate IRS molecules on serine/threonine sites, promoting their degradation (*54*). If this occurs with IRS-2 in β cells, it should then trigger apoptosis. In addition, IL-1β and TNFα are known to activate Jnk/p38 "stress protein kinases" as well as the nPKC isoform, PKCδ (*27*). As mentioned previously, Jnk/p38 and PKCδ may also increase the serine/threonine phosphorylation state of IRS-2, leading to IRS-2 degradation and ultimately β-cell apoptosis.

IRS-2 also plays a pivotal role in insulin signal transduction pathways in insulin target tissues, especially the liver (*29*, *37*). A decrease in IRS-2 expression causes insulin resistance in insulin-responsive tissues (*30*, *33*, *37*). Indeed, several of the mechanisms proposed above to explain the loss of IRS-2 expression in β cells have also been proposed to reduce IRS-1 and IRS-2 levels in liver, muscle, and/or fat, thereby contributing to insulin resistance (*29*, *38*, *44*, *52–55*). Thus, there are parallels between the molecular mechanisms that control insulin sensitivity and those that promote β-cell survival. If these mechanisms go awry, then the balance between insulin resistance and compensatory β-cell mass will become ever more disrupted as time goes on, accelerating the onset of type 2 diabetes.

Summary

Although there is a tenuous balance between insulin resistance and an effective β-cell mass, for the most part, the β-cell mass adapts adequately to compensate for changes in the metabolic load. However, β cells can be pushed too far in susceptible individuals. Eventually the β-cell mass fails to compensate for insulin resistance, and type 2 diabetes ensues. I have argued here that this failure is caused by a marked increase in β-cell apoptosis, most likely induced by a combination of chronic hyperglycemia, hyperlipidemia, and/or certain cytokines that interfere with the signaling pathways that maintain normal β-cell growth and survival. The net effect is a reduction in functional β-cell mass in the type 2 diabetic state. There are many signal transduction pathways that affect β-cell growth and survival (*2*), and here I have focused only on one component of those pathways, IRS-2, because it is vital to normal maintenance of the adult β-cell population. One can envisage that pharmacological manipulations that increase IRS-2 expression in β cells may be a valuable strategy for promoting β-cell survival and delaying the onset of diabetes (*12*, *28*). Because decreased expression of IRS-2 in insulin-responsive tissues contributes to the insulin-resistant state, such a therapeutic strategy—at least in principle—would not only protect β cells but also help alleviate insulin resistance.

Analyses of the other signaling pathways governing β-cell growth and survival (*2*) will likely identify additional drug targets for preventing β-cell apoptosis. The future holds promise that strategies directed at prolonging the survival of the β cell will be successful in delaying or even preventing the onset of type 2 diabetes.

References and Notes

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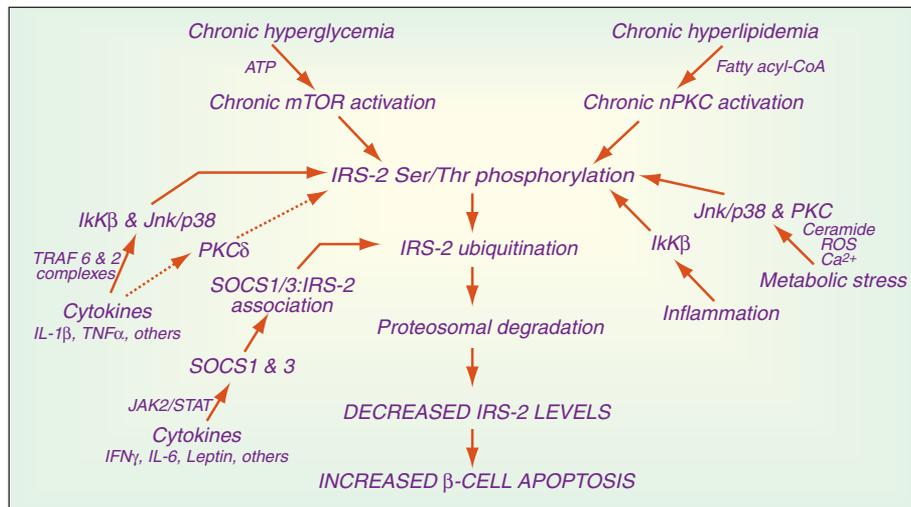
Fig. 3. Potential mechanisms that trigger IRS-2 degradation and apoptosis of pancreatic β cells. IRS-2 expression in β cells is vital for normal β-cell growth, survival, and turnover. Chronic hyperglycemia by means of mTOR activation and hyperlipidemia by means of fatty acyl-CoA–mediated activation of the novel class of PKC isoforms can lead to increased serine/threonine phosphorylation of IRS-2 that then leads to its ubiquitination and subsequent proteosomal degradation. In addition, certain cytokines, including IL-1β and TNF-α, activate IκKβ and Jnk/p38 kinases [by means of TNF-receptor associated factor (TRAF) signaling complexes] and/or PKCδ, which in turn also leads to IRS-2 serine/threonine phosphorylation. Other local inflammatory responses can activate IκKβ, and other metabolic stresses (e.g., increased ROS and ceramide generation, or chronically elevated [Ca^{2+}]) can activate PKC isoforms and/or Jnk/p38 kinases, which can reduce IRS-2 levels by a similar mechanism. Other cytokines (including IL-6 and IFN-γ) can induce expression of SOCS-1 and SOCS-3, which can then bind to IRS-2, leading to its ubiquitination and subsequent proteosomal degradation. This multipronged assault could significantly lower IRS-2 levels in the β cell. The resultant increase in β-cell apoptosis is thought to be a key factor contributing to the loss of β-cell mass in type 2 diabetes.

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VIEWPOINT

Mitochondrial Dysfunction and Type 2 Diabetes

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Maintenance of normal blood glucose levels depends on a complex interplay between the insulin responsiveness of skeletal muscle and liver and glucose-stimulated insulin secretion by pancreatic β cells. Defects in the former are responsible for insulin resistance, and defects in the latter are responsible for progression to hyperglycemia. Emerging evidence supports the potentially unifying hypothesis that both of these prominent features of type 2 diabetes are caused by mitochondrial dysfunction.

Type 2 diabetes is the most common metabolic disease in the world. In the United States, it is the leading cause of blindness, end-stage renal disease, and nontraumatic loss of limb, with associated health care costs estimated to exceed $130 billion per year (*1*). Of even greater concern, type 2 diabetes is rapidly becoming a global pandemic and is projected to afflict more than 300 million individuals worldwide by the year 2025, with most of the increase occurring in India and Asia (*2*). Although the primary cause of this disease is unknown, it is clear that insulin resistance plays an early role in its pathogenesis and that defects in insulin secretion by pancreatic β cells are instrumental in the progression to hyperglycemia. Here, we explore the potentially unifying hypothesis that these two prominent features of type 2 diabetes are both attributable to defects in mitochondria, the organelles that provide energy to the cell.

Role of Intracellular Fatty Acid Metabolites in Insulin Resistance

Several lines of evidence indicate that insulin resistance is an early feature of type 2 diabetes. First, virtually all patients with type 2 diabetes are insulin-resistant, and prospective studies have shown that this insulin-resistant state develops 1 to 2 decades before the onset of the disease (*3–5*). Second, insulin resistance in the offspring of parents with type 2 diabetes is the best predictor for later development of the disease (*6*). Lastly, perturbations that reduce insulin resistance prevent the development of diabetes (*7*).

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Skeletal muscle and liver are the two key insulin-responsive organs responsible for maintaining normal glucose homeostasis, and their transition to an insulin-resistant state accounts for most of the alterations in glucose metabolism seen in patients with type 2 diabetes. Before considering whether mitochondrial dysfunction contributes to the development of insulin resistance in these organs, it is first important to understand the cellular mechanisms responsible for insulin resistance. As discussed by Lazar (*8*), there is growing evidence that circulating cytokines secreted by fat tissue can modulate the insulin responsiveness of liver and muscle. However, fatty acids (*9*) and/or intracellular fatty acid metabolites such as fatty acyl coenzyme As (fatty acyl CoAs) (*10, 11*), diacylglycerol (*10, 11*), or ceramides (*12*) are also thought to play a critical role.

Over 40 years ago, Randle *et al.* demonstrated that fatty acids caused insulin resistance in an in vitro rat muscle preparation, and they hypothesized that this occurred by a substrate competition mechanism (*13*). According to his model, increased oxidation of muscle fatty acids would produce increased levels of intracellular acetyl CoA and citrate, which in turn would inhibit, respectively, two enzymes involved in glucose utilization, pyruvate dehydrogenase and phosphofructokinase. Inhibition of the glycolytic pathway at these steps would increase intracellular glucose and glucose-6-phosphate concentrations, ultimately resulting in reduced insulin-stimulated glucose uptake.

More recent studies using ¹³C and ³¹P magnetic resonance spectroscopy (MRS) have shown that this mechanism for fatty acid–induced insulin resistance is untenable in human skeletal muscle (*14*); rather, fatty acids appear to cause insulin resistance by directly inhibiting insulin-stimulated glucose transport activity (*15*). This inhibition is likely because of the accumulation of intracellular

fatty acyl CoAs and diacylglycerol, which then activate critical signal transduction pathways that ultimately lead to suppression of insulin signaling (Fig. 1). One might therefore predict that any metabolic perturbation that promotes the accumulation of fatty acids in liver and/or muscle and/or any defect in the ability of these organs to metabolize fatty acids might result in insulin resistance (*10*). Indeed, defects in adipocyte metabolism, which occur in conditions such as severe lipodystrophy (*16*), can result in the former, and it has become increasingly evident that defects in mitochondrial fatty acid oxidation can result in the latter and may be responsible for the more common forms of insulin resistance.

Mitochondrial Dysfunction, Intracellular Fatty Acids, and Insulin Resistance

It is well established that mitochondrial function is required for normal glucose-stimulated insulin secretion from pancreatic β cells. In addition, maternally inherited defects in mitochondrial DNA that disrupt mitochondrial function are known to cause an insulin-deficient form of diabetes resembling type 1 diabetes (*17*). However, recent MRS studies of humans suggest that more subtle defects in mitochondrial function might also play a role in the pathogenesis of insulin resistance and type 2 diabetes. Petersen *et al.* found that in comparison with matched young controls, healthy lean elderly subjects had severe insulin resistance in muscle, as well as significantly higher levels of triglycerides in both muscle and liver (*18*). These changes were accompanied by decreases in both mitochondrial oxidative activity and mitochondrial adenosine triphosphate (ATP) synthesis. These data support the hypothesis that insulin resistance in humans arises from defects in mitochondrial fatty acid oxidation, which in turn lead to increases in intracellular fatty acid metabolites (fatty acyl CoA and diacylglyerol) that disrupt insulin signaling (Fig. 1).

Alterations in mitochondrial DNA (MtDNA) have been correlated with human aging in several previous studies, and a recent study of genetically manipulated mice provided evidence that such alterations may play a causal role in aging (*19*). Whether the mitochondrial