Cellular Mechanisms for Insulin Resistance in Normal Pregnancy and Gestational Diabetes

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The incidence of gestational diabetes mellitus (GDM) has doubled over the last 6–8 years and is paralleling the obesity epidemic. GDM carries long-term implications for the subsequent development of type 2 diabetes in the mother and increased risk for obesity and glucose intolerance in the offspring. Insulin resistance exists before pregnancy in women with a history of GDM but worsens during gestation. Insulin secretion is inadequate to compensate for the insulin resistance, leading to hyperglycemia that is detected by routine glucose screening in pregnancy. Thus, chronic insulin resistance is a central component of the pathophysiology of GDM.

Human pregnancy is characterized by a series of metabolic changes that promote adipose tissue accretion in early gestation, followed by insulin resistance and facilitated lipolysis in late pregnancy. In early pregnancy, insulin secretion increases, while insulin sensitivity is unchanged, decreased, or may even increase (1,2). However, in late gestation, maternal adipose tissue depots decline, while postprandial free fatty acid (FFA) levels increase and insulin-mediated glucose disposal worsens by 40–60% compared with prepregnancy (2). The ability of insulin to suppress whole-body lipolysis is also reduced during late pregnancy (3), and this is further reduced in GDM subjects (4), contributing to greater postprandial increases in FFAs, increased hepatic glucose production, and severe insulin resistance (2,5–7). Although various placental hormones have been suggested to reprogram maternal physiology to meet fetal needs, the cellular mechanisms for this complex transition remain obscure (8). Further, the critical molecular mechanisms involved in increasing maternal lipid flux in obese women throughout pregnancy that may underlie skeletal muscle insulin resistance and increased fetal fuels are just beginning to be investigated.

RECENT INSIGHTS RELATING PLACENTAL HORMONES AND ADIPOKINES IN THE INSULIN RESISTANCE OF PREGNANCY

— Skeletal muscle is the principal site of whole-body glucose disposal, and along with adipose tissue, becomes severely insulin resistant during the latter half of pregnancy. Normal pregnancy is characterized by an ~50% decrease in insulin-mediated glucose disposal in humans and a 200–250% increase in insulin secretion to maintain euglycemia in the mother (2,9). Placental-derived hormones are believed to be a major factor in reprogramming maternal physiology to achieve an insulin-resistant state. However, it is important to note that, with the exception of tumor necrosis factor (TNF)-α, changes in placental hormones in human pregnancy do not directly correlate with changes in maternal insulin resistance (10). Therefore, a synergy with other obesity- or pregnancy-related factors may hold the key to understanding how insulin resistance develops during pregnancy.

Human placental lactogen (hPL) increases up to 30-fold throughout pregnancy and induces insulin release from the pancreas in pregnancy (11). Studies outside of pregnancy indicate that hPL can cause peripheral insulin resistance (12), although the results have been variable (13). Another hormone recently implicated in the insulin resistance of pregnancy is human placental growth hormone (hPGH), which differs from pituitary growth hormone by 13 amino acids. hPGH increases six- to eightfold during gestation and replaces normal pituitary growth hormone in the maternal circulation by ~20 weeks' gestation (8). Much like the well-documented effects of excess pituitary growth hormone on insulin sensitivity, overexpression of hPGH in transgenic mice comparable to levels seen in the third trimester of pregnancy causes severe peripheral insulin resistance (14). Surprisingly, little work has been done to determine the molecular mechanisms of insulin resistance in skeletal muscle in response to elevated hPL or hPGH. Recent evidence has shown that an important effect of hPGH is to specifically increase the expression of the p85α subunit of phosphatidylinositol (PI) 3-kinase in skeletal muscle. Studies in pregnant and nonpregnant humans (15,16) indicate that increases in the p85α subunit of PI 3-kinase acts as a dominant-negative competitor to forming a PI 3-kinase heterodimer with the p110 subunit, thereby inhibiting PI 3-kinase activity and preventing further

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Abbreviations: FFA, free fatty acid; GDM, gestational diabetes mellitus; hPGH, human placental growth hormone; hPL, human placental lactogen; IR, insulin receptor; IRS, insulin receptor substrate; PI, phosphatidylinositol; PPAR, peroxisome proliferator–activated receptor; TNF, tumor necrosis factor.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

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insulin signaling downstream (17), as discussed below.

Recent prospective studies have implicated adiponectin from adipocytes and secreted factors, such as TNF-α, as active candidates in mediating insulin resistance of pregnancy. Collectively, these factors, known as "adipokines," include leptin, adiponectin, TNF-α, interleukin-6, resistin, and others. TNF-α is a cytokine, produced not only from monocytes and macrophages, but also from T-cells, neutrophils, fibroblasts, and adipocytes. Obese animals and humans show a positive correlation between TNF-α levels and BMI and hyperinsulinemia (18–20). Infusion of TNF-α results in increased insulin resistance in rat and human skeletal muscle cells incubated in culture (21), although TNF-α neutralization over a period of 4 weeks had no effect on insulin sensitivity in obese type 2 diabetic subjects (22). Although the concentration of TNF-α in plasma of obese patients is much lower compared with that found in burn patients and patients with cachexia, some evidence suggests that local skeletal muscle TNF-α may act in a paracrine fashion to contribute to skeletal muscle insulin resistance (21,22). TNF-α impairs insulin signaling by increasing serine phosphorylation of insulin receptor substrate (IRS)-1 (18,19) and diminishing insulin receptor (IR) tyrosine kinase activity (23) as discussed below. Studies in pregnancy have reported that changes in insulin sensitivity from early (22–24 weeks) to late (34–36 weeks) gestation correlate with plasma TNF-α ($r = 0.45$) (10) and that circulating TNF-α may be produced by the placenta and skeletal muscle to induce or exacerbate insulin resistance through mechanisms that remain to be determined.

Increasing adiposity is correlated with the secretion of proinflammatory cytokines from adipose tissue (24–26), suggesting their release may play an important role in fuel availability during pregnancy. The signals that regulate the secretion of these molecules are far from clear and include stimuli as diverse as mechanical stress induced by expansion of adipocytes (27), increased glucose flux (28), and endoplasmic reticulum stress (29). Of the adipokines released by adipose tissue, adiponectin is the most abundant in the circulation. Adiponectin is a secreted globular protein synthesized exclusively in adipocytes. In humans, low plasma adiponectin concentrations correlate highly with insulin resistance in obesity, type 2 diabetes, and GDM (30–32). Recent findings also show that adiponectin secretion and adiponectin mRNA levels in white adipose tissue decline with advancing gestation, even in lean women (33), suggesting that there are pregnancy-associated factors that reduce adiponectin levels. Circulating levels of adiponectin have been shown to correlate with whole-body insulin sensitivity, presumably working though adiponectin receptors in skeletal muscle and liver (34). Adiponectin stimulates glucose uptake in skeletal muscle and reduces hepatic glucose production through its effect on AMP-activated protein kinase. Thus, adiponectin can be viewed as an endogenous insulin-sensitizing hormone. Several studies have demonstrated that, similar to obese patients and type 2 diabetic patients, adiponectin levels are reduced in former GDM patients (35,36) and are lower in GDM women during late pregnancy compared with pregnant control subjects matched for BMI (37–40). TNF-α and other proinflammatory mediators suppress the transcription of adiponectin in adipocytes (41,42), which might explain the lower levels of serum adiponectin in individuals with GDM.

MECHANISMS UNDERLYING REDUCED GLUCOSE TRANSPORT IN SKELETAL MUSCLE FIBERS ISOLATED FROM OBESE PREGNANT WOMEN AND FURTHER REDUCTION IN GDM

GDM — The action of insulin to increase glucose uptake is controlled initially by the transport of glucose across the cell membrane, which takes place by facilitated diffusion through glucose transporters. We have investigated the effects of pregnancy and GDM on the ability of insulin to stimulate glucose transport in human skeletal muscle fiber strips in vitro. Freshly isolated skeletal muscle fibers were obtained from obese GDM women during elective cesarean delivery and compared with muscle fibers from obese pregnant subjects with normal glucose tolerance matched for BMI, age, and ethnicity (90% Caucasian). We also obtained samples of *rectus abdominus* from obese nonpregnant subjects during elective abdominal procedures to examine the mechanisms for the insulin resistance of pregnancy compared with the nonpregnant state. We demonstrated directly in skeletal muscle fibers that pregnancy alone was associated with a marked reduction (40%) in insulin-stimulated glucose transport, and this impairment in insulin action was significantly worse in GDM subjects (65% reduced) compared with obese pregnant subjects (5). The reduction in glucose transport occurs in the absence of any detectable change in total GLUT4 abundance in skeletal muscle from pregnant or GDM subjects (43). These results are analogous to those of Garvey and Birnbaum (44), who measured glucose transport in isolated adipocytes and found a more severe decrease in glucose transport in obese GDM subjects, although those subjects were compared with nonobese pregnant control subjects.

Upon binding to the insulin receptor, insulin stimulates tyrosine phosphorylation of the β-subunit of the receptor on at least six tyrosine residues (45). The initial step in insulin receptor signaling is defective in GDM subjects who have significantly less maximal tyrosine phosphorylation of the IR compared with women with normal glucose tolerance during pregnancy (5,46), with no change in receptor binding or numbers (5,46). This finding indicates that GDM subjects carry an intrinsic defect in IR tyrosine phosphorylation per receptor protein that could be due to an endogenous inhibitory pathway for receptor signaling. The autophosphorylation of these tyrosine residues activates the receptor to dock intracellular substrates (IRS-1 to IRS-6). IRS-1 is the major docking protein in human skeletal muscle. After IRS-1 is phosphorylated on tyrosine domains, it triggers the recruitment of PI 3-kinase, a crucial event required for stimulating glucose transport (47). Tyrosine phosphorylation of IR and IRS proteins is balanced by dephosphorylation reactions carried out by cellular and membrane-bound protein tyrosine phosphatases. Evidence indicates that protein tyrosine phosphatase activity can serve as a negative regulator of insulin receptor phosphorylation to alter reduced sensitivity in human and animal models (48–51). We investigated the level of the major protein phosphatase, PTP1B protein, but found that this was not altered in pregnancy or GDM compared with obese nonpregnant subjects (52).

In contrast to IR phosphorylation on tyrosine, which stimulates downstream insulin signaling, phosphorylation of the IR on serine/threonine residues decreases signaling and can act as a dampening signaling mechanism. We obtained IR receptors from skeletal muscle of women in late

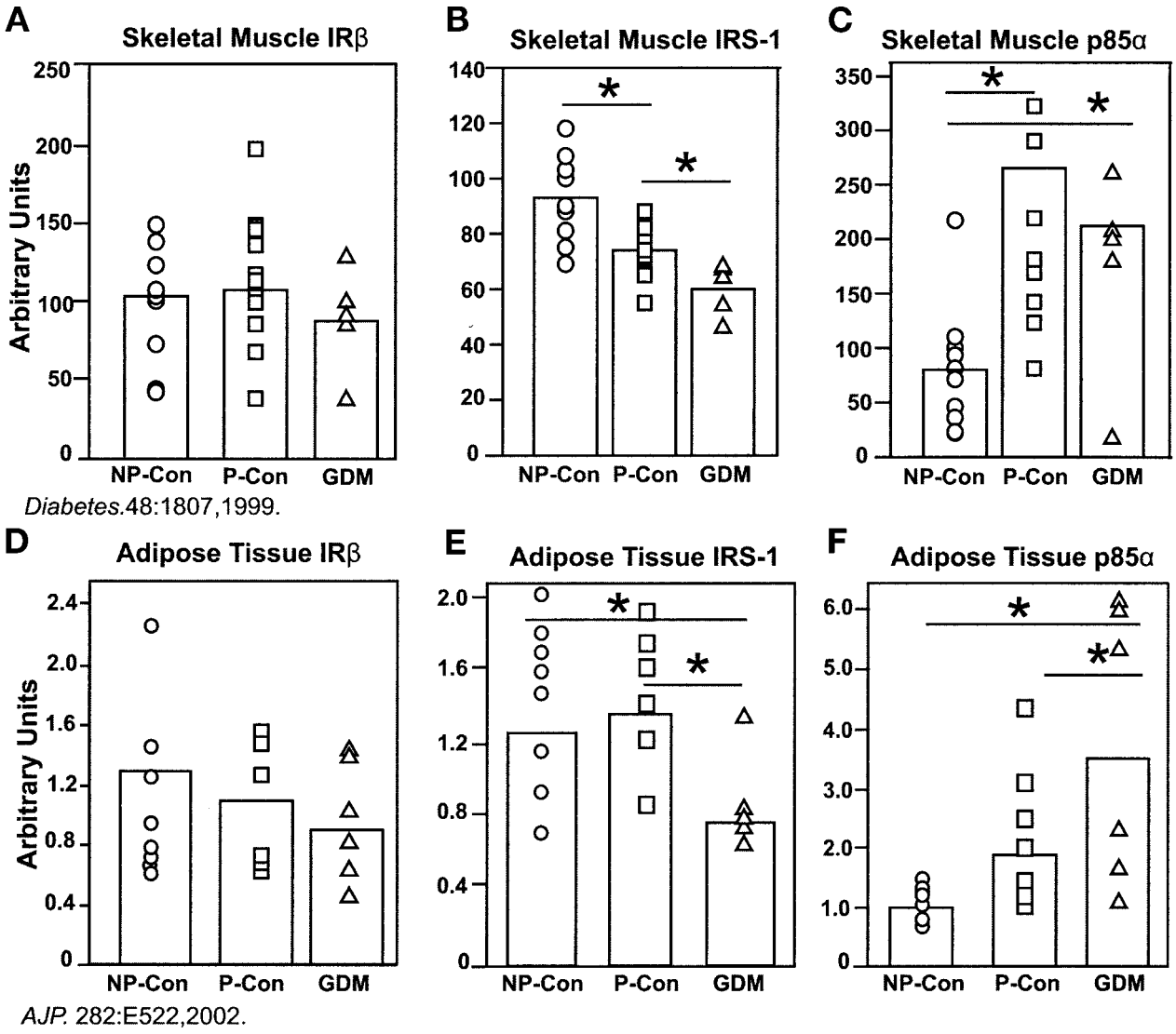

Figure 1—*Changes in insulin signaling proteins in pregnant and GDM women during the third trimester. Insulin signaling proteins, IR, IRS-1, and p85α were analyzed by Western blot in skeletal muscle* (A–C) *and adipose tissue* (D–F) *biopsies from obese nonpregnant control (NP-Con), obese pregnant women (P-Con) with normal glucose tolerance, and weight-matched GDM women (GDM). Individual data points to show variability within each group along with the group average are indicated by the bar* (n = *5–10/group;* *P < *0.05*).

gestation and measured the activity of the partially purified receptors after insulin treatment. This was significantly reduced in pregnancy and more so in GDM subjects (46). However, when we pretreated these receptors with alkaline phosphatase to remove serine and tyrosine phosphorylation, the ability of insulin to activate tyrosine phosphorylation of the IR was restored to normal in receptors from pregnant women and partially restored in GDM subjects (46). The reversibility of the IR tyrosine kinase activity by alkaline phosphatase pretreatment suggests that pregnancy could involve activation of a serine kinase(s) to induce a posttranslational modification of the IR that can significantly dampen insulin signaling.

In intact cells, IR serine/threonine phosphorylation can be stimulated by prolonged insulin treatment, phorbol esters, and cAMP analogs, presumably as a result of activation by protein kinase C (53–55). In addition, the cytokine TNF-α has been shown to act as a serine/threonine kinase to inhibit both IR and IRS-1 tyrosine phosphorylation (56,57). As mentioned above, circulating TNF-α levels increase during pregnancy and correlate with the extent of insulin resistance measured in humans during pregnancy (10).

IRS-1 IS DOWNREGULATED IN PREGNANCY AND MORE SO IN GDM

— In addition to the activity of the insulin receptor, the level of IRS-1 protein is critical for regulation of glucose uptake in insulin-sensitive tissues. Our studies show that unlike the IR, IRS-1 protein is decreased in skeletal muscle by 30–50% in normal obese pregnant and GDM subjects compared with obese nonpregnant control subjects (5), suggesting a strong effect of pregnancy and possibly a stronger effect of GDM in downregulating IRS-1 levels (Fig. 1). In adipose tissue biopsies from the abdominal wall of the same patients in late pregnancy, we also confirmed that IRS-1 is downregulated in GDM women only (4). In a follow-up study, vastus lateralis muscle biopsies were obtained antepartum and postpartum. The IRS-1 protein levels were measured and were reduced by 52%

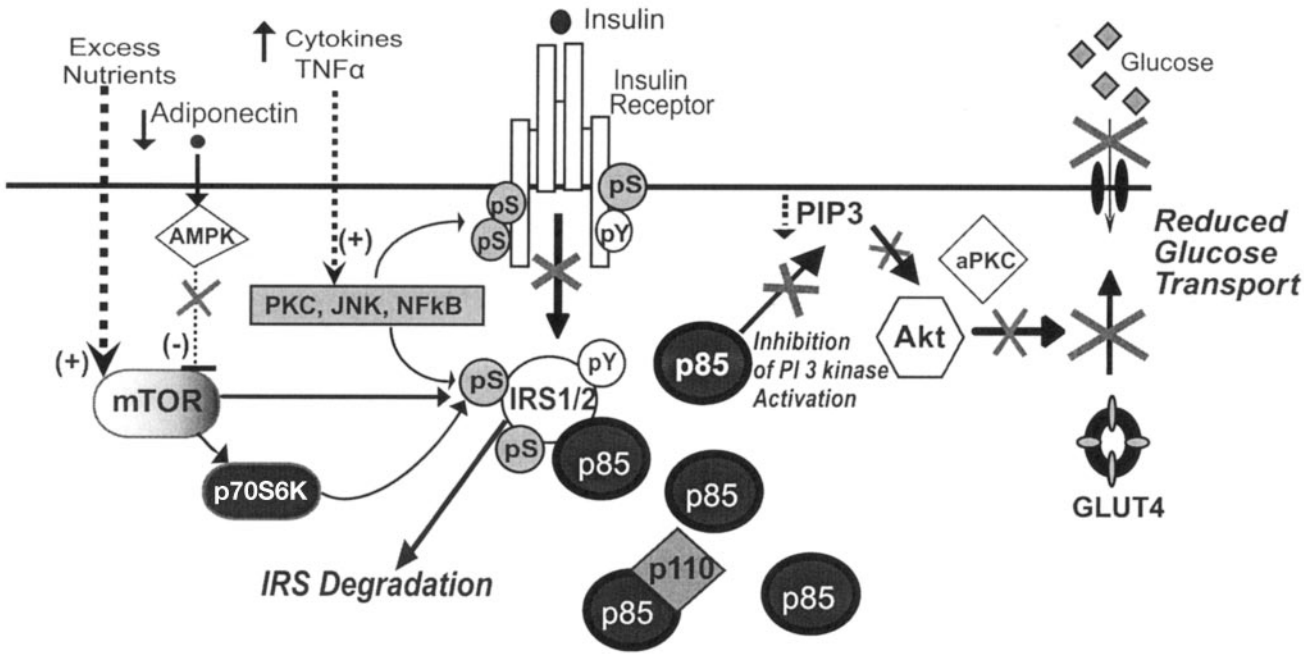

Figure 2—*Summary of potential mechanisms for insulin resistance in skeletal muscle during late pregnancy in human gestational diabetes. The pathway for insulin stimulation of glucose transport in muscle involves activation of the insulin receptor protein, which docks IRS-1 and IRS-2 and phosphorylates these proteins on tyrosine residues (pY). IRS-1 recruits the p85α regulatory subunit of PI 3-kinase (p85-p110), resulting in phosphorylation of membrane-bound phospholipids at the 3′ position (phosphoinositol-3,4,5-phosphate [PIP_3]). Production of PIP_3 is required for activation of Akt and signaling for GLUT4 translocation. Defective tyrosine phosphorylation of IR and IRS-1 was observed in GDM subjects and is associated with increased inhibitory serine phosphorylation (pS) and a striking loss of IRS-1 protein levels compared with normal obese nondiabetic subjects. Increased serine phosphorylation of IR and IRS-1 has been linked to increased activation of JNK and protein kinase C (PKC), enzymes activated by inflammatory cytokines (such as TNF-α). Alternatively, the increase in IRS-1 serine phosphorylation may be attributed to enhanced activation of the mTOR-p70S6 pathway. mTOR, p70S6K1, and AMPK act as nutrient and energy sensors within the cell. Higher basal phosphorylation of p70S6K1 was seen in GDM versus control subjects and may account for the increase in basal serine IRS-1 phosphorylation and lead to IRS-1 degradation. AMPK, a target for adiponectin signaling, is a negative regulator of mTOR. In GDM subjects, adiponectin levels are lower, which could contribute to the increased activation of the mTOR pathway. In both normal pregnant and GDM subjects, p85α levels are elevated antepartum but return to normal postpartum. Excess p85α acts as a dominant-negative signaling molecule by blocking the association of PI 3-kinase (p85-p110) with IRS-1 and thereby attenuating PI 3-kinase activation during pregnancy. Loss of PI 3-kinase activation from increased p85 and increased serine phosphorylation of IRS-1 both lead to reduced translocation of GLUT4 to the plasma membrane and result in decreased insulin-stimulated glucose uptake to skeletal muscle.*

in GDM women compared with obese control subjects antepartum (58). More importantly, IRS-1 content of skeletal muscle returned to normal postpartum by 6 weeks (58) and in non-GDM women who returned to normal weight 1 year postpartum (15). Thus, while the reduction in skeletal muscle IRS-1 protein is more severe in GDM women, it appears to be reversible after pregnancy.

INCREASED IRS-1 SERINE PHOSPHORYLATION IS AN ADDITIVE FACTOR REDUCING INSULIN SIGNALING IN GDM

— Although the underlying defects are still being investigated, growing evidence suggests that local production/action of inflammatory molecules may be particularly important in generating skeletal muscle insulin resistance in obesity and type 2 diabetes. Evidence suggests that there are at least two potential cellular mechanisms for inflammatory mediators to provoke insulin resistance involving the IR-IRS-PI 3-kinase signaling pathway. One mechanism with strong experimental evidence is IRS-1 serine phosphorylation. As mentioned, elevated serine phosphorylation reduces insulin-stimulated IR and IRS-1 tyrosine phosphorylation and PI 3-kinase activity and is accompanied by impaired glucose uptake. In many in vitro systems, this pathway for insulin resistance correlates with serine phosphorylation of IRS-1 on 307/312 (mouse/human) and other phosphorylation sites (59,60). Most recently, this has been observed in skeletal muscle of obese and type 2 diabetic patients compared with lean patients (16). In a recent longitudinal study, we performed vastus lateralis muscle biopsies at 30–34 weeks' gestation and found increased basal levels of 312-IRS-1 serine phosphorylation relative to total IRS-1 abundance in skeletal muscle from GDM women compared with obese pregnant women with normal glucose tolerance (58). Basal 312-serine phosphorylation of IRS-1 to total IRS-1 was increased by 62% in GDM women compared with pregnant control subjects. Furthermore, after insulin stimulation with a glucose load, tyrosine phosphorylation of IRS-1 was correspondingly reduced in GDM women, consistent with the increase in inhibitory serine kinase expression. Increased serine phosphorylation of IRS-1 decreases IRS-1 association with the IR and can inhibit PI 3-kinase activity, thereby inhibiting insulin signaling from activating GLUT4 translocation (61), as shown in Fig. 2.

The serine kinase(s) responsible for the increased IR or IRS-1 serine phos-

phorylation is unknown. However, several signaling cascades have been implicated in the serine phosphorylation of IRS-1 in states of insulin resistance including JNK1 (62), NF-κB (63), protein kinase C-θ (64), mTOR (65), and p70 S6K1 (66), which can phosphorylate IRS-1 on serine residues and inhibit its function. Of these kinases, JNK and NF-κB are activated by inflammatory mediators, such as TNF-α, whereas other inhibitory kinases (mTOR, p70 S6K1, and protein kinase C-θ) are increased in insulin-resistant states by conditions of nutrient excess. In a preliminary human study, we found that GDM subjects had a significant increase in basal p70 S6K1 phosphorylation levels during late pregnancy compared with women with normal glucose tolerance, but it was restored to normal postpartum (58). These data suggest that increased p70 S6K activation, which increases IRS-1 serine phosphorylation and its degradation (67), could help to explain the increased IRS-1 depletion in GDM subjects. Because p70 S6K1 is activated by excess amino acids and glucose, nutrient excess in GDM might underlie the activation of this important serine kinase.

INCREASED P85 MONOMER OF PI 3-KINASE AND ITS ROLE IN THE INSULIN RESISTANCE OF NORMAL PREGNANCY

— IRS-1 docks the important insulin-signaling protein p85α, which is a critical step for generating PI 3-kinase activity in response to insulin. PI 3-kinase is composed of an 85-kDa regulatory subunit (p85α) and a catalytic 110-kDa subunit (p110). The protein levels of the p85α subunit were unexpectedly higher in skeletal muscle and adipose tissue obtained from pregnant and GDM subjects compared with obese nonpregnant women (4,5), a finding that prompted us to investigate the role of p85 further. For PI 3-kinase activation to occur, both the regulatory p85α and catalytic p110 subunit must bind as a heterodimer to phosphorylated IRS-1. Association of PI 3-kinase with IRS-1 brings it in close proximity to its phospholipid substrates in the plasma membrane, resulting in the formation of phosphoinositol-3,4,5-phosphate, which is necessary to propagate downstream signaling to Akt and atypical protein kinase C, necessary for glucose transport (68). Studies in several laboratories have shown that a disrupted balance between the levels of the PI 3-kinase subunits directly regulates insulin sensitivity in mice and in cells (rev. in 69). In our human studies, the p85α levels in rectus abdominus and vastus lateralis muscle and adipose tissue were elevated 1.5- to 2.0-fold in obese pregnant women compared with obese nonpregnant control subjects (4). The levels returned to normal in both pregnant and GDM subjects 1 month postpartum (17) and in women who return to normal body weight 1 year after pregnancy (15).

We recently demonstrated that expression of the p85 monomers is increased in transgenic mice overexpressing placental growth hormone (14) and that mice with a heterozygous deletion for p85α were protected from growth hormone–induced insulin resistance (17). After insulin stimulation, excess p85α competes with p85-p110 heterodimers for specific PI 3-kinase binding sites on IRS-1. Binding of p85α monomers to IRS-1 effectively prevents access of p85-p110 heterodimers binding to IRS-1 (dominant-negative effect), resulting in a marked decrease in IRS-1–associated PI 3-kinase activation (17). Therefore, our human and animal data support an important role for hPGH in driving increased p85α abundance, resulting in a decrease in the IRS-1–associated PI 3-kinase activity. This additional mechanism may compound the insulin resistance found in skeletal muscle due to IR and IRS-1 serine phosphorylation as shown in Fig. 2.

INSULIN RESISTANCE IN ADIPOSE TISSUE: IMPLICATIONS FOR EXCESS FUELS AND THE ORIGINS OF INSULIN RESISTANCE

— Unlike in skeletal muscle, GLUT4 protein is downregulated in adipose tissue of pregnant women, and the decrease is more profound in women with GDM (70). In addition, insulin-induced translocation of GLUT4 to plasma membranes is abnormal in GDM patients (71). Our studies in adipose tissue biopsies from obese GDM subjects also show that IRS-1 protein is downregulated and this downregulation is related to impaired insulin-induced suppression of FFAs in these subjects (4). These findings suggest that insulin resistance in adipose tissue could lead to important metabolic changes in cytokine expression and FFA release that may figure prominently in the mechanisms underlying insulin resistance, increased nutrient availability, and subsequent transfer to the fetus.

The molecular changes in adipose tissue during pregnancy include a reduction in the transcription factor peroxisome proliferator–activated receptor (PPAR)-γ1 (4). PPAR-γ1 binds to several adipose-specific genes and is a central regulator of the adipogenic transcriptional cascade (72,73). PPAR-γ1 is normally highly expressed in adipose tissue and plays an essential role in fat cell differentiation, insulin sensitivity, and lipid storage (74–76). PPAR-γ is also strongly implicated in the regulation of systemic insulin sensitivity (76). For example, PPAR-γ agonists (thiazolidinediones) are used for treatment of human diabetes to increase insulin sensitivity (77). Additionally, patients with dominant-negative mutations in the PPAR-γ gene are nonobese yet have severe insulin resistance and type 2 diabetes (78). The target genes induced by PPAR-γ include (among others) adiponectin (79), lipoprotein lipase (80), the intracellular fatty acid binding protein aP2, and the mitochondrial uncoupling protein UCP2 (rev. in 81). We found a 40–50% decrease in PPAR-γ mRNA and protein in abdominal white adipose tissue from both obese pregnant control subjects and obese GDM subjects at term compared with obese nonpregnant subjects (4). One factor that might suppress PPAR-γ during pregnancy is the inflammatory cytokine TNF-α (10). TNF-α downregulates PPAR-γ expression in 3T3-L1 cells and can inhibit adipocyte differentiation (82). Interestingly, growth hormone has also been shown to suppress PPAR-γ mRNA in vitro and in vivo (83,84). Our observations have led us to hypothesize that placental growth hormone may play an important role in accelerating the transition from lipid storage to lipolysis and insulin resistance during pregnancy. This transition may be accelerated in obese or GDM women either owing to an increase in maternal sensitivity to the hormone or possibly a synergy with other obesity- or pregnancy-related factors, such as hPL or TNF-α.

SUMMARY

—The insulin resistance of normal pregnancy is multifactorial, involving reduced ability of insulin to phosphorylate the IR, decreased expression of IRS-1, and increased levels of the p85α subunit of PI 3-kinase. IRS-1 is further decreased in most GDM subjects compared with obese pregnant women at term. However, in GDM, there are reciprocal and inverse changes in the degree of serine and tyrosine phosphorylation of IR and IRS-1 that further inhibit signaling,

leading to substantially reduced GLUT4 translocation and decreased glucose uptake beyond that of normal pregnancy. Women with a history of GDM have evidence of subclinical inflammation (35,36), and there is evidence for increased TNF-α as well as increased p70 S6K1 in skeletal muscle from GDM women. Adiponectin, a key insulin-sensitizing hormone produced by adipose tissue, is significantly lower in women with a history of GDM and declines with advancing gestation, suggesting it could be involved in the transition to insulin resistance. In adipose tissue, the lipogenic transcription factor PPAR-γ declines in obese women during pregnancy and may shift genes in metabolic pathways to favor increased lipolysis, thus accelerating adipose tissue insulin resistance and the switch from lipid storage to lipolysis. This transition to insulin resistance contributes to greater postprandial increases in FFAs and increased hepatic glucose production and results in greater fuel availability to the fetus of women with GDM. Thus, like a perfect storm, subclinical inflammation, placental hormones, reduced adiponectin secretion, and excess lipolysis conspire to cause severe insulin resistance in liver, muscle, and adipose tissue in women with GDM.

POSTPARTUM AND BEYOND

— Although most patients diagnosed with GDM show normal glucose tolerance soon after delivery by conventional testing, a subset are found to have impaired glucose tolerance or type 2 diabetes. The majority develop type 2 diabetes in the first decade after delivery, especially those who demonstrate impaired glucose tolerance postpartum. The factors that underlie chronic insulin resistance in former GDM subjects remain to be identified. It will be particularly important in future studies to determine the candidate serine kinase(s) involved in GDM, since it may underlie both the IR and IRS-1 signaling defects that increase these patients' susceptibility to glucose intolerance and diabetes. The role of excess lipid turnover and cytokine production from adipose tissue, especially in obese patients, could potentially be very important in the overall insulin resistance and excess substrate supply that drives maternal-fetal energy transfer and increased neonatal adiposity. Very little is known about the role of reduced adiponectin or the decrease in PPAR-γ expression in adipose tissue of obese pregnant women (4) in the pathogenesis of increased fetal fat accretion. A crucial question remains as to what regulatory molecules contribute to the change from lipid storage to accelerated lipolysis in human pregnancy. Furthermore, are the metabolic changes in adipocytes from obese women, with or without GDM, the same as in lean women? The answers to these questions will have a profound impact on our understanding of glucose and lipid metabolism in pregnancy and may provide clues to the origin of excess fuels and insulin resistance in GDM.

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