

Lipids and Glucose in Type 2 Diabetes

What is the cause and effect?

GUENTHER BODEN, MD¹
MARKKU LAAKSO, MD²

Historically, type 2 diabetes was considered to revolve around a glucose-insulin axis. The foundations for this thinking were probably laid down by two momentous discoveries in diabetes research. According to popular legend, Oskar Minkowski noticed that urine from his pancreatectomized dogs attracted an inordinate number of flies. He is then alleged to have tasted the urine and noted its sweetness. From this observation came the supposition that the pancreas produced a substance that controlled sugar concentration, and diabetes occurred in the absence of this substance. The second landmark was the discovery by Frederick Banting that insulin was the active element from the pancreas. As a consequence of these two discoveries, the concept that the glucose-insulin relationship was the central element of fuel metabolism gained a firm hold. Diabetes has since been considered to be a disorder primarily associated with abnormal glucose metabolism. This notion is given credence by the considerable amount of data indicating that the chronic elevation of plasma glucose causes many of the microvascular complications of diabetes.

In 1992, McGarry (1) asked what would have happened if Minkowski had lacked a sense of taste but had a good nose. If that had been the case, he may have smelled acetone, and this may have led him to the conclusion that removal of the pancreas affects fatty acid metabolism.

From the ¹Division of Endocrinology/Diabetes/Metabolism and the General Clinical Research Center, Temple University Hospital, Philadelphia, Pennsylvania; and the ²Department of Medicine, University of Kuopio, Kuopio, Finland.

Address correspondence and reprint requests to G. Boden, MD, Temple University Hospital, 3401 N. Broad St., Philadelphia, PA 19140. E-mail: bodengh@tuhs.temple.edu.

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Abbreviations: DAG, diacylglycerol; FFA, free fatty acid; IL, interleukin; IRS, insulin receptor substrate; NF, nuclear factor; NHANES III, Third National Health and Nutrition Examination Survey; PKC, protein kinase C; PPAR, peroxisome proliferator-activated receptor; TNF, tumor necrosis factor; TZD, thiazolidinedione.

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

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If this had happened, our understanding of the pathogenesis of type 2 diabetes may have developed along a different route and led us more quickly to our current awareness that obesity, or more accurately, the products of excess adipose tissue, precede the perturbations of glucose metabolism.

It is now apparent that elevation of plasma free fatty acids (FFAs) plays a pivotal role in the development of type 2 diabetes by causing insulin resistance. Type 2 diabetes develops because pancreatic β -cells eventually fail to produce enough insulin to compensate for the ongoing insulin resistance. There is a tight association between type 2 diabetes and dyslipidemia. The latter is characterized by raised small, dense LDL levels, elevated levels of triglycerides, and low levels of HDL. Individually, the latter two factors increase the risk of cardiovascular disease, and the combination of the two is a risk factor for cardiovascular heart disease that is at least as strong as a high level of LDL cholesterol (2). Some (3,4) but not all (5) studies have demonstrated that increased levels of small, dense LDL are associated with an elevated risk of cardiovascular disease.

EVOLUTIONARY RATIONALE FOR FAT ACCUMULATION

According to the thrifty gene hypothesis (6), the probability of an individual sur-

living times of famine would be increased if they could maximize energy storage (as fat) during times of surplus food availability. The stored fat could then be used during periods of starvation.

Adipocytes take up and store FFAs. The two main types of adipose tissue are subcutaneous and visceral adipose tissue. About 80% of body fat is located in the subcutaneous adipose tissue, and ~10% is located in visceral adipose tissue (7). The remainder is in various other locations, such as perirenal and peritoneal adipose tissue (7).

The body uses its fat reserves during periods of low energy intake, when FFAs are being released for other tissues to be used as fuel. However, if plasma FFA levels are elevated for more than a few hours, they will cause insulin resistance (8). In certain conditions, the FFA-induced insulin resistance has the beneficial effect of preserving carbohydrate for use by vital tissues, such as the central nervous system. This is the case during starvation and during the second half of pregnancy, when the insulin resistance of the mother preserves glucose for the growing fetus.

PATHOLOGICAL CONSEQUENCES OF FAT ACCUMULATION

In contrast to its beneficial effects during periods of starvation and gestation, during prolonged periods of energy excess, the FFA-induced insulin resistance becomes counterproductive because there is no need for preservation of carbohydrate for use by vital tissues. Under these conditions, glucose levels remain normal only as long as the basal and postprandial secretion of insulin by the pancreas is sufficient to compensate for the insulin resistance.

INDUCTION OF INSULIN RESISTANCE BY FFAS

The mechanisms by which elevated FFA levels result in insulin resistance have been determined in skeletal muscle, where most insulin-stimulated glucose uptake occurs. Formerly, it was believed that FFA production from overloaded fat

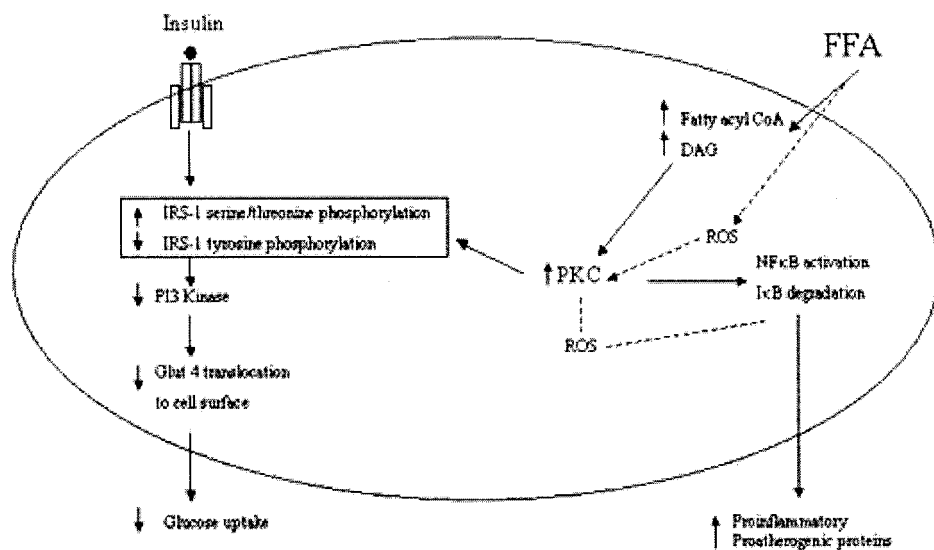


Figure 1—Potential mechanism of FFA on insulin resistance and atherogenesis in human muscle. The key initiating event is an increase in plasma FFA followed by increased uptake of FFA into muscle. This leads to intramyocellular accumulation of fatty acyl-CoA and DAG and activation of PKC (the β II and δ isoforms). It is assumed that activation of PKC interrupts insulin signaling by serine phosphorylation of IRS-1, resulting in a decrease in tyrosine phosphorylation of IRS-1. At the same time, activation of PKC also leads to production of inflammatory and proatherogenic proteins through activation of the $I\kappa B$ - α /NF- κB pathway. The broken lines indicate that activation of PKC by reactive oxygen species (ROS) and activation of the $I\kappa B$ - α /NF- κB pathway by ROS has not been demonstrated in human muscle but only in bovine aortic smooth muscle and endothelial cells (ref. 84). PI, phosphatidylinositol.

cells disrupted glucose homeostasis via the Randle glucose–fatty acid cycle. First described by Randle et al. (9) in 1963, the hypothesis was that glucose uptake is reduced when tissue energy needs are being met by FFA oxidation. The oxidation of FFAs was thought to result in decreased glucose oxidation and an increase in intracellular citrate levels, which would decrease glycolysis and glucose uptake. In vivo and in vitro studies have only partially confirmed Randle's hypothesis (8, 10–12). For instance, insulin-stimulated glucose uptake has been found to proceed normally for several hours after maximal inhibition of carbohydrate oxidation by fatty acids (8, 11–13).

It is now thought that FFAs induce insulin resistance in human muscle at the level of insulin-stimulated glucose transport or phosphorylation by impairing the insulin-signaling pathway (Fig. 1) (13,14). In a study of nondiabetic men and women, insulin resistance developed 2–4 h after an acute elevation in plasma FFA concentration and took a similar amount of time to disappear after plasma FFA levels had returned to normal (8,15). This delay indicates that FFAs have an indirect effect, a contention that is supported by the observation that acute increases in plasma FFAs increased triglyceride content in muscle cells of human volunteers (16). This rise in intramyocellular triglyceride concentration occurred several hours after the elevation of FFAs and coincided with the development of insulin resistance (16).

However, it is probably not the accumulation of fat in muscle cells that causes insulin resistance but rather the accumulation of other metabolites, including diacylglycerol (DAG), that occur at the same time (17).

DAG, which is an intermediate of triglyceride metabolism, is a potent activator of protein kinase C (PKC) (18). In healthy volunteers, along with the rise in intramyocellular levels of DAG, there was a concomitant increase in PKC activity (17). PKC is an enzyme that can phosphorylate serine and threonine residues on both the insulin receptor (19,20) and insulin receptor substrate (IRS)-1 (20, 21). The latter two molecules are important for insulin signaling. Serine phosphorylation of IRS-1 can lead to its destruction and to insulin resistance (22).

A change in intracellular DAG levels is also accompanied by activation of the nuclear factor (NF)- κB pathway (Fig. 1) (17). NF- κB has been linked to fatty acid-induced impairment of insulin action in rodents (23,24). NF- κB is also increasingly recognized as playing a crucial role in the pathogenesis of coronary artery disease (25). Thus, the activation of NF- κB may also help to explain the increased prevalence of vascular disease in obese patients with type 2 diabetes. Consequently, lowering FFA concentrations may prevent activation of the NF- κB pathway and have benefits beyond increasing insulin sensitivity and improving regulation of glucose levels.

Another mechanism by which FFAs

can cause insulin resistance is by increasing oxidative stress (26). Reactive oxygen species can activate PKC and the NF- κB pathway (Fig. 1) and thereby contribute to insulin resistance (17,27).

FFAs also affect the functioning of insulin in the liver and thus contribute to hepatic overproduction of glucose and to elevated circulating blood glucose levels (28). The main role of insulin in the liver is control of glucose production. The mechanism by which insulin acutely (within 1–2 h) suppresses hepatic glucose production is by inhibiting glycogenolysis (29). FFAs produce insulin resistance in the liver by inhibiting the acute insulin suppression of glycogenolysis (30). Insulin also promotes hepatic uptake of FFAs and production of intracellular triglycerides. Thus, insulin resistance in the liver may contribute to elevated plasma FFA levels.

An increase in visceral fat could also cause insulin resistance by mechanisms that do not directly involve FFAs (Fig. 1). Adipose tissue is a source of inflammatory mediators, such as tumor necrosis factor (TNF)- α (31) and interleukin (IL)-6 (32) and peptides that include resistin (33), leptin (34), and adiponectin (35). So far, however, the physiological relevance of these adipokines for the development of insulin resistance in humans has not been established.

ADIPONECTIN AND INSULIN RESISTANCE

Of all the adipokines mentioned, adiponectin is most likely to affect insulin

sensitivity. Adiponectin is produced exclusively in adipocytes. It stimulates fatty acid oxidation, decreases plasma triglycerides, and improves glucose metabolism by increasing insulin sensitivity (36). Adiponectin levels are negatively correlated with the development of insulin resistance in rhesus monkeys (37). The plasma levels of adiponectin in Caucasians and Pima Indians are negatively correlated with percent body fat and plasma insulin levels and are positively correlated with insulin-mediated glucose uptake (35,38). In addition, plasma levels of adiponectin are lower in individuals with type 2 diabetes than in age- and body mass-matched individuals without diabetes (35). Thus, the impaired release of adiponectin that occurs in obesity may contribute to insulin resistance and development of type 2 diabetes.

The mechanism leading to decreased adiponectin levels in obesity (39) is not clear. Adiponectin is inhibited by insulin and TNF- α . Therefore, hyperinsulinemia caused by obesity-induced insulin resistance, together with enhanced TNF- α expression, may contribute to reduced adiponectin secretion. It has also been suggested that visceral adipose tissue may produce an as yet unidentified substance that destabilizes adiponectin mRNA (40).

FFAs AND INSULIN SECRETION

Insulin resistance in the liver results in overproduction of glucose, whereas insulin resistance in skeletal muscle produces underutilization of glucose. Because FFAs can induce insulin resistance in both liver and muscle, all overweight or obese people, who are likely to have elevated plasma FFA levels, might be expected to have elevated glucose levels. This, however, is not the case because only approximately half of overweight individuals have abnormal glucose levels; in the Third National Health and Nutrition Examination Survey (NHANES III), 23% of overweight or obese (BMI ≥ 25 kg/m²) individuals had impaired fasting glucose (fasting glucose concentration of 110–125 mg/dl) or impaired glucose tolerance (2-h glucose concentration of 140–199 mg/dl), and 23% had diabetes (41). NHANES III was conducted using a lower limit of fasting glucose of 110 mg/dl to define impaired fasting glucose, but the American Diabetes Association has since defined the cutoff as 100 mg/dl. On the

basis of this new definition, the actual proportion of overweight people with impaired glucose metabolism will be higher than the estimates from published surveys (including NHANES III) that used the older definition.

Recently, it has become clearer why many obese, insulin-resistant people will never develop diabetes. In obese people with normal pancreatic β -cells, FFAs are potent insulin secretagogues and can compensate for the insulin resistance that they produce. Acute elevations of plasma FFAs have long been known to stimulate insulin secretion (42). More importantly, prolonged elevations of plasma FFAs (2–4 days) have been shown to potentiate glucose-stimulated insulin secretion in healthy volunteers (43–45). Moreover, when chronic elevated plasma FFA levels were lowered in obese diabetic and non-diabetic subjects, insulin secretion rates decreased by 30–50% (46), indicating that the elevated plasma FFAs had supported 30–50% of basal insulin secretion.

In contrast, in first-degree relatives of patients with type 2 diabetes (i.e., in individuals who are genetically predisposed to develop diabetes), FFAs are unable to fully compensate with adequately increased insulin secretion for the insulin resistance that they produce (45,47). This defect in FFA-stimulated insulin secretion can also be demonstrated in patients with impaired glucose tolerance (47,48) and in patients with overt type 2 diabetes (46). These observations suggest that the obese individuals who develop type 2 diabetes have a genetic predisposition to pancreatic β -cell failure. This hypothesis is supported by the fact that these individuals have a defect in both FFA and glucose-stimulated insulin secretion (49) and by *in vitro* studies showing that basal insulin secretion from normal Wistar rat islets increased when cultured for 7 days in 2 mmol/l FFAs but decreased in islets from Zucker fatty rats who are predisposed to develop diabetes (50).

PEROXISOME PROLIFERATOR-ACTIVATED RECEPTORS: A TARGET FOR THERAPEUTIC INTERVENTION

Prolonged exposure to elevated FFA levels is central to the development of insulin resistance and type 2 diabetes, making modulation of these levels an attractive therapeutic strategy. Nicotinic acid and

nicotinic acid analogs are drugs that lower plasma FFA levels. Their usefulness is limited, however, because the initial lowering of plasma FFA levels by nicotinic acid is invariably followed by a sharp FFA rebound (51) that increases insulin resistance, at least temporarily. There has therefore been considerable interest in drugs that activate the peroxisome proliferator-activated receptors (PPARs), which are nuclear transcription factors that regulate the expression of numerous genes involved in lipid and carbohydrate metabolism, inflammation, and vascular tone.

PPAR- γ

Thiazolidinediones (TZDs), in contrast to nicotinic acid analogs, lower plasma FFA levels without a rebound phenomenon (52–54). The binding affinity between TZDs and PPAR- γ correlates well with their insulin-sensitizing activity. Therefore, it is generally accepted that TZDs exert their action through PPAR- γ (55,56). PPAR- γ is expressed at highest concentrations in adipose tissue and at much lower concentrations in liver and muscle (57,58), suggesting that the primary action of TZDs is on adipose tissue. In support, it has been shown that TZDs play an important role in adipocyte development (59) and that they lower plasma levels of FFAs (Fig. 2) (52–54). In addition, TZDs have been postulated to improve insulin sensitivity by redistributing fat from visceral to subcutaneous adipose tissue (60,61) and by increasing blood levels of adiponectin (62,63).

The available TZDs (pioglitazone and rosiglitazone) have a generally good safety and tolerability profile. They have been associated with increased development of edema, which can exacerbate or lead to congestive heart failure (64,65) in high-risk patients with preexisting vascular disease (66,67). The highest incidences of edema and congestive heart failure were seen when TZDs were used in combination with insulin (65,68,69). Alongside improvements in glycemia and in some lipid parameters, e.g., HDL, clinical trials have also reported dose-dependent weight gain compared with placebo and small decreases in hemoglobin and hematocrit after treatment with TZDs (70,71).

PPAR- α

FFAs are natural ligands for PPAR- α , which is preferentially expressed in tis-

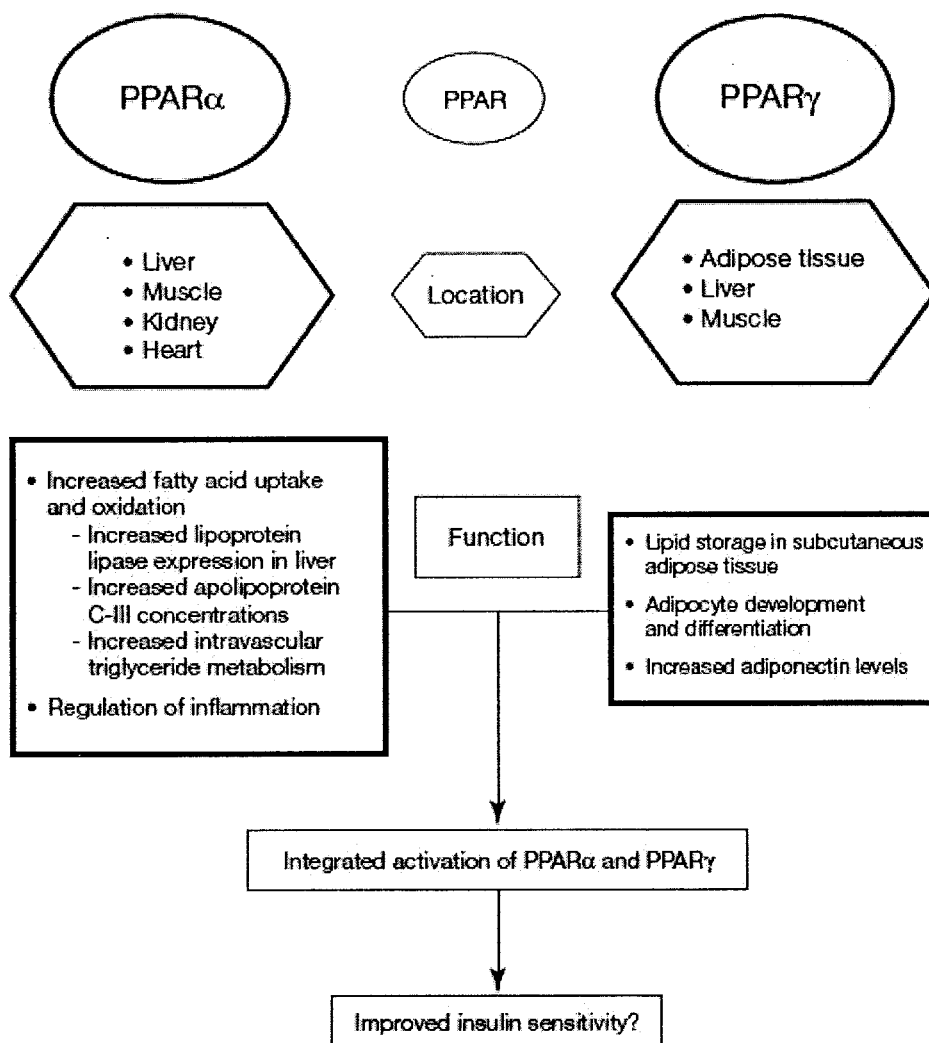


Figure 2—Potential contributions of PPAR- α and PPAR- γ to improvements in insulin sensitivity.

sues where fatty acids are oxidized such as the liver, muscle, kidney, and heart. Activation of PPAR- α stimulates the expression of genes involved in fatty acid and lipoprotein oxidation in the liver and muscle (72,73). PPAR- α activators, such as fibrates, decrease plasma FFA and triglyceride concentrations by stimulating several metabolic pathways (Fig. 2). Fibrates increase fatty acid uptake and oxidation by increasing the expression of lipoprotein lipase in the liver and by decreasing apolipoprotein C-III concentrations. Apolipoprotein C-III is a protein that inhibits triglyceride hydrolysis by lipoprotein lipase, and its downregulation by fibrates results in reduced triglyceride and VLDL production by the liver (74). Simultaneously, these agents enhance intravascular triglyceride metabolism (75). The lowering of FFAs through increased oxidation may well improve insulin sen-

sitivity. Specific PPAR- α agonism can lower lipid levels in rats and improve insulin sensitivity (76,77), whereas studies of fibrates in humans have either reported improved (78,79) or unimproved (80,81) insulin sensitivity.

Dual PPAR- α / γ activation

PPAR- γ exerts its beneficial effect by lowering plasma FFA levels, increasing plasma adiponectin levels, and redistributing fat from visceral to subcutaneous depots, and PPAR- α activation lowers plasma FFA levels through increased fat oxidation. Therefore, dual PPAR- α / γ agonism may have advantages over selective PPAR subtype activation (82,83). Specifically, dual PPAR- α / γ agonism may lower plasma FFAs more than either PPAR- α or PPAR- γ alone (Fig. 2). Given the central role of FFAs in the development of insulin resistance and type 2 diabetes, this ap-

proach may offer an attractive option for therapeutic intervention. Ongoing studies are examining the efficacy and safety of these new agents.

SUMMARY

From an initial perception that a disorder of glucose metabolism was the primary event in the pathogenesis of type 2 diabetes, there is now a growing appreciation that chronic elevation of FFA levels is an early event that contributes to the development of this disease. FFAs induce insulin resistance, which increases with FFA levels, and this can be a beneficial adaptive response during starvation and pregnancy. However, insulin resistance can become counterproductive when there is an excess of energy intake associated with physical inactivity. The extra fuel is stored in visceral and subcutaneous fat depots. As fat accumulates, there is an ongoing

increase in the levels of plasma FFAs, which causes insulin resistance. In addition, the deficit of another product of adipose tissue (e.g., adiponectin) may contribute to increased insulin resistance. To counter insulin resistance and prevent hyperglycemia, insulin levels increase. In individuals with a genetic predisposition for diabetes, however, the pancreas cannot compensate for the increased secretory demands placed on it, resulting in type 2 diabetes.

The pivotal role of FFAs in the development of insulin resistance and type 2 diabetes suggests that the optimal therapeutic intervention should decrease plasma FFA levels. The PPAR family is intimately involved in lipid metabolism. Two subtypes of these receptors are the site of action of synthetic PPAR agonists: PPAR- α and PPAR- γ . The former increases fatty acid oxidation, whereas the latter results in the redistribution of fat from visceral to subcutaneous body fat and an increase in adiponectin. The outcome of activation of PPAR- γ is a lowering of plasma FFA concentrations and improved insulin sensitivity. The effects of PPAR- α on lipid metabolism may also bring about improvements in insulin sensitivity. The currently available PPAR agonists selectively activate either PPAR- α (i.e., fibrates) or PPAR- γ (i.e., TZDs) and have been shown to improve lipid metabolism. It may be that dual agonism with PPAR- α/γ agonists will provide additional benefits above and beyond those achieved with sole activation of either PPAR- α or PPAR- γ .

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