β-Cell failure in type 2 diabetes

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ABSTRACT

Type 2 diabetic patients are insulin resistant as a result of obesity and a sedentary lifestyle. Nevertheless, it has been known for the past five decades that insulin response to nutrients is markedly diminished in type 2 diabetes. There is now a consensus that impaired glucose regulation cannot develop without insulin deficiency. First-phase insulin response to glucose is lost very early in the development of type 2 diabetes. Several prospective studies have shown that impaired insulin response to glucose is a predictor of future impaired glucose tolerance (IGT) and type 2 diabetes. Recently discovered type 2 diabetes-risk gene variants influence β-cell function, and might represent the molecular basis for the low insulin secretion that predicts future type 2 diabetes. We believe type 2 diabetes develops on the basis of normal but 'weak' β-cells unable to cope with excessive functional demands imposed by overnutrition and insulin resistance. Several laboratories have shown a reduction in β-cell mass in type 2 diabetes and IGT, whereas others have found modest reductions and most importantly, a large overlap between β-cell masses of diabetic and normoglycemic subjects. Therefore, at least initially, the β-cell dysfunction of type 2 diabetes seems more functional than structural. However, type 2 diabetes is a progressive disorder, and animal models of diabetes show β-cell apoptosis with prolonged hyperglycemia/hyperlipemia (glucolipotoxicity). β-Cells exposed *in vitro* to glucolipotoxic conditions show endoplasmic reticulum (ER) and oxidative stress. ER stress mechanisms might participate in the adaptation of β-cells to hyperglycemia, unless excessive. β-Cells are not deficient in antioxidant defense, thioredoxin playing a major role. Its inhibitor, thioredoxin-interacting protein (TXNIP), might be important in leading to β-cell apoptosis and type 2 diabetes. These topics are intensively investigated and might lead to novel therapeutic approaches.
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KEY WORDS: β-Cell failure, Insulin secretion, Insulin resistance

INTRODUCTION

To present day readers of this journal, it might seem strange that until the 1970s[1] it was not known that type 1 diabetes is an autoimmune disease entirely distinct from type 2 diabetes. Diabetes specialists who have reached a certain age today, including the senior author of this review, were trained to view the differences between various types of diabetes as a matter of degree, not of kind; some patients required insulin treatment to survive, whereas others managed their metabolic state with dietary means or pharmacological agents. As insulin became more widely used, it also appeared that in many patients whom today we characterize as typical type 2 diabetic patients, achieving metabolic control with insulin treatment was difficult if not impossible. Thus, half a century before the insulin receptor and its signaling pathways were identified, the British clinician HP Himsworth[2,3] wrote '…I would suggest the possibility of the existence of a type of diabetes due not to diminished secretion of insulin by the pancreas, but to a greater or less impairment of the organism's susceptibility to insulin'. In other words, the idea that diabetes might be the result of insulin resistance

was aired already in the 1930s. Indeed, the presence of obesity in the vast majority of patients with type 2 diabetes makes it a reasonable assumption that insulin resistance must exist in this disorder.

Strikingly different is the approach that has dominated the last decades of the 20th century, viewing insulin resistance as the main, often sole, etiological factor in type 2 diabetes, negating any role of deranged insulin secretion. Summarizing this view, the *Journal of Clinical Investigation* as recently as 2000 published a 'Perspective' series entitled 'On diabetes: insulin resistance'[4]. It is difficult to understand how insulin deficiency could so widely be ignored; already in the early 1960s it was shown by several investigators that the insulin responses to glucose challenge is markedly reduced in type 2 diabetes, including in normoglycemic subjects with glucose intolerance only (IGT)[5–9]. Fortunately, over the years, in the face of dogmatic-monolithic positions, balanced views have also been presented[10–12] that point to the fact that the biology of type 2 diabetes is not simple; pure β-cell deficiency or exclusive insulin resistance are rare events, because in reality, insulin secretion and insulin action are interconnected, as would be expected in any feedback regulatory loop.

Also, clinical evidence points to the fact that in many type 2 diabetic patients the metabolic state cannot be ascribed to insuperable insulin resistance. Indeed, although conventional insulin

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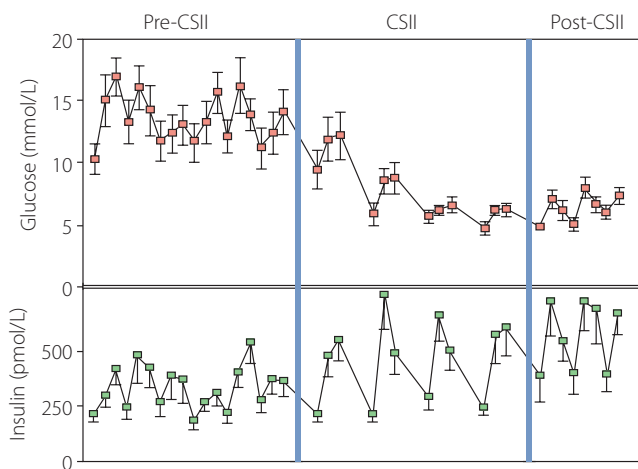E-mail address: erol.cerasi@huji.ac.il

Figure 1 | Induction of near-normoglycemia in type 2 diabetes patients by continuous subcutaneous infusion of insulin (CSII). A total of 12 patients were treated with CSII for 2 weeks; towards the end of the period, daily blood glucose excursions were markedly reduced and approached normal values. The data shown are part of a larger study, presented by Ilkova et al.[14]

therapy in type 2 diabetes often fails or gives suboptimal results, intensified insulin treatment [multiple dose administration or continuous subcutaneous infusion of insulin (CSII)] is able to near-normalize blood glucose in many patients. Thus, in pilot studies in 23 type 2 diabetic patients, we could achieve fasting and postprandial normoglycemia with a mean daily CSII insulin dose of 0.55 U/kg bodyweight[13,14]; this is shown in Figure 1. Recently, Retnakaran et al.[15] achieved similar results with a multiple injection protocol administering a mean daily insulin dose of 0.65 U/kg. Finally, in contrast to the aforementioned studies that were carried out mainly in Caucasian patients, Weng et al.[16] achieved near-normoglycemia with CSII in a large group of Chinese type 2 diabetic patients with a daily insulin dose of 0.68 U/kg. The interest of these studies is not only that excellent metabolic control could be achieved in type 2 diabetic patients with exogenous insulin, but that the daily insulin requirement was not substantially different from that used as replacement therapy in insulin-deficient type 1 diabetic patients. We certainly recognize the important role of insulin resistance in the pathophysiology of type 2 diabetes, but conclude nevertheless that type 2 diabetes is first of all a disorder of insulin deficit; the input of insulin resistance to its pathogenesis increases with the severity of obesity, acting as a magnifier of insulin deficiency.

PLASMA INSULIN IN TYPE 2 DIABETES

It is important to remember that both insulin response to glucose and peripheral sensitivity to insulin show a remarkably wide range of variation in non-diabetic lean subjects as well as obese subjects, with a continuous distribution of the parameters; that is, without evidence of population segregation[17–20]. This is shown in Figure 2 with data selected from two studies as
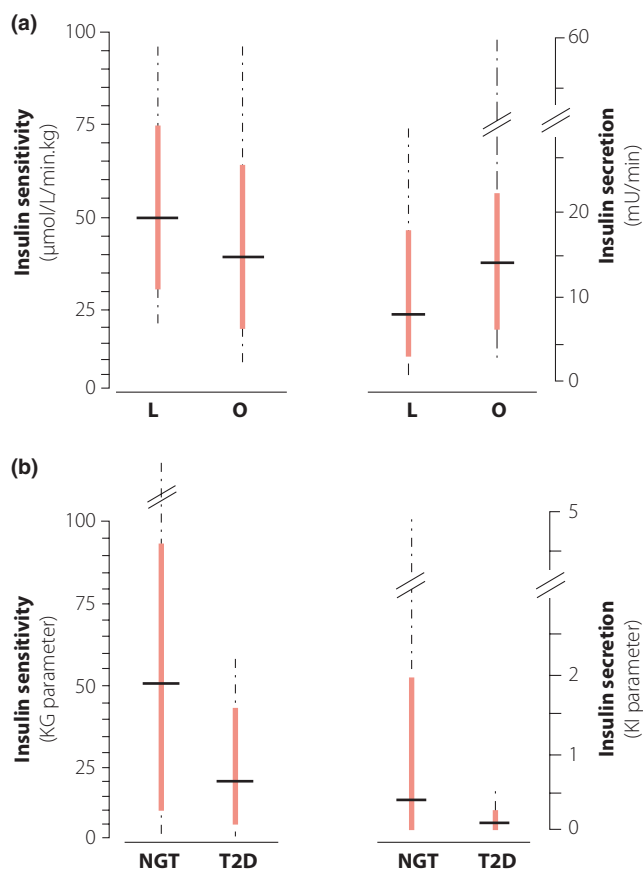

Figure 2 | The wide variability of insulin sensitivity and insulin secretion. (a) Whole body insulin-mediated glucose uptake (left panel) and basal post-hepatic insulin delivery rate (right panel) were assessed in 608 lean (L) and 538 obese (O) non-diabetic subjects with euglycemic hyperinsulinemic clamps by Ferrannini et al.[20]; the data shown here were recalculated from the original publication. The thick vertical bars show the results between the 5th and 95th percentile, whereas the dotted lines show the data range; the short horizontal bars denote the median. (b) Whole body glucose clearance stimulated by endogenous insulin (insulin sensitivity, KG, left panel) and sensitivity of insulin secretion to glucose (insulin secretion, KI, right panel) were assessed by modeling of data generated with a glucose infusion test in 226 lean normal glucose tolerant controls (NGT) and 25 lean type 2 diabetes patients (T2D)[17]. The results of Efendic et al.[17] have been replotted here in a similar manner to those of Figure 2a. Note the markedly skewed distribution of the values; the range of insulin sensitivities in NGT subjects exceeded the limits of the figure (166.0 KG units). Note also the markedly lower insulin response of the type 2 diabetes patients, and their more modestly reduced insulin sensitivity.

examples. In obese subjects, although mean insulin sensitivity is reduced and insulin response augmented, the wide variation of these parameters leads to a major overlap with the levels of lean subjects. Although less marked, the variability of insulin sensitivity is also important in type 2 diabetic patients (Figure 2b). In substantial numbers of subjects with either markedly low insulin response or low sensitivity to insulin, normal glucose tolerance

is retained; furthermore, clinical experience shows that more than two out of three obese subjects never develop impaired glucose tolerance (IGT), despite insulin resistance. In type 2 diabetic patients, except in its very severe forms, fasting and postprandial plasma insulin levels are normal or higher than normal. This observation provides the rationale for insulin resistance in diabetes; it is, however, a static view of a highly dynamic regulatory system, confusing cause and effect: what degree of hyperinsulinemia is adequate for a given degree of hyperglycemia? An example of the fallacy of the argument is provided by our study in 15 mildly obese type 2 diabetic patients treated with a sulfonylurea for 6 months in whom, with the normalization of blood glucose, the initially high fasting plasma insulin levels fell to the normal range, despite the use of the β-cell stimulating sulfonylurea[13]. Thus, fasting insulin is also under the control of blood glucose. Obviously, similar arguments might (and should) be applied to postprandial insulin levels in type 2 diabetes. For example, the bell-shaped insulin curve used to describe changes in β-cell function during the transition from normal to IGT and type 2 diabetes is an artefact as a result of the use of 120-min plasma insulin values in the oral glucose tolerance test (OGTT); the higher glucose levels in patients with IGT amplify the secretion of insulin, resulting in a typical late insulin peak. When earlier (e.g. 30-min) time-points are chosen, the insulin response to OGTT shows a continuous fall from normal over IGT to type 2 diabetes[21–23]. We wish to reiterate with emphasis that, provided plasma insulin data are interpreted with full reference to the physiology of regulated insulin secretion, β-cell responsiveness to glucose is lower than normal in IGT, and more so in type 2 diabetes. The data presented in Figure 2b show this point; the glucose responsiveness of β-cells assessed by computer simulation was dramatically lower in mild type 2 diabetic patients than in control subjects.

When glucose tolerance is impaired, the earliest modifications of the insulin response to glucose to be detected concern the so-called first-phase insulin response, which is rapidly reduced and then lost, and the disruption of the oscillatory character of insulin release[5,6,8,24,25]. Assessment of insulin oscillations is technically demanding and therefore seldom utilized in larger clinical studies. In contrast, the early insulin response to glucose can be measured during oral or i.v. glucose tolerance tests, whereas glucose clamps allow detailed definition of the insulin response kinetics. The first-phase response is markedly reduced in subjects with IGT, and further diminishes with the advent of fasting hyperglycemia. The later or second-phase insulin response to glucose is retained in early or moderately advanced type 2 diabetes, but as the disease progresses this phase of insulin secretion also collapses (Figure 3). Some studies suggest that type 2 diabetic patients might also present changes in incretin secretion and action, with consequences for the remaining β-cell function. For extensive reading, please see a recent review in this journal[26].

Low first-phase insulin response is also found in a proportion of subjects with normal glucose tolerance[5,6] (Figure 2b). Several
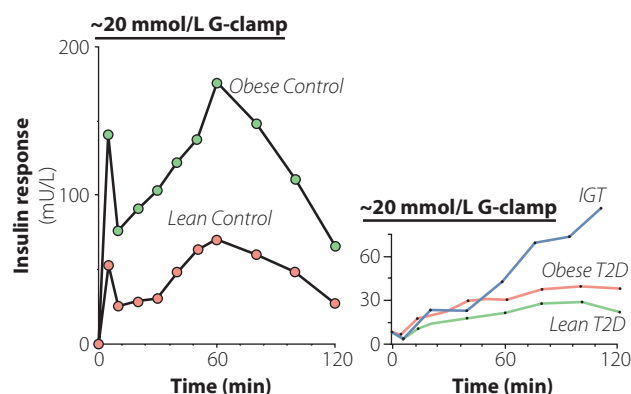

Figure 3 | Insulin response to glucose infusion. Typical examples of the insulin response in lean and obese normal glucose tolerant (left panel) and type 2 diabetes subjects at different stages of the disease (right panel). Note the loss of the first-phase insulin response already at the stage of impaired glucose tolerance (IGT).

studies over past years have shown that a low insulin response is a predictor of future glucose intolerance and type 2 diabetes, in lean as well as obese subjects belonging to various ethnic groups[27–29]. To give an example, we followed a large group of lean and physically active Swedish subjects with normal glucose tolerance for a mean period of 25 years; the initially-measured first-phase insulin response corrected for insulin sensitivity (disposition index) was significantly correlated to later glucose tolerance, low values predicting IGT and type 2 diabetes[27].

GENETIC CAUSES OF REDUCED INSULIN SECRETION

Extensive studies over past decades in family members of diabetic patients and control subjects, including monozygotic twin pairs, have shown that many aspects of the plasma insulin response to glucose administration in humans are under strong genetic control[30–33]. However, it is only recently that data have emerged that allow some insight into the possible cellular mechanisms responsible for the decrease of β-cell function in subjects at risk of developing diabetes. Indeed, numerous whole genome association studies carried out over the past decade have identified allelic variants of several genes that collectively participate in the risk of type 2 diabetes development. The majority of these genes are involved in β-cell development, function and survival[34]. As the number of risk alleles that a subject carries increases, several aspects of β-cell function deteriorate; most pertinently, the insulin response to oral or i.v. glucose decreases in proportion to the number of risk alleles[35,36]. By which cellular mechanisms these risk alleles impair insulin secretion is not known. However, recent data suggests that, at least regarding the highest-risk gene transcription factor 7-like 2 (TCF7L2), distal steps in exocytosis, including insulin granule connection with voltage-gated calcium channels in the β-cell, might be involved, thus reducing the efficiency of the insulin exocytotic machinery, although other mechanisms have also been proposed[37–40]. It can be expected that within a short time the molecular mechanisms

of the low insulin response to glucose, which is a strong risk factor for type 2 diabetes, will be fully clarified at the molecular level, as has been done for the various forms of monogenic diabetes[41–43].

PROGRESSIVE DETERIORATION OF β-CELL FUNCTION IN TYPE 2 DIABETES

It has become axiomatic that type 2 diabetes is a progressive disease; it is indeed a common clinical experience that with prolonged duration, the severity of diabetes increases and requires augmenting numbers and doses of anti-diabetic drugs. This experience has been confirmed and extended by the United Kingdom Prospective Diabetes Study (UKPDS) investigators; whatever the treatment modality chosen, the level of hemoglobin A_{1c} (HbA$_{1c}$) increases with time[44]. A very important question is whether type 2 diabetes is an inherently progressive disorder as a result of the molecular nature of its pathogenesis, or whether progression is secondary to the metabolic state including hyperglycemia. It is indeed nearly impossible to obtain normoglycemia throughout the day over the life of a diabetic patient, whichever treatment modality is chosen. Notwithstanding its molecular cause, the progressive deterioration of metabolism in type 2 diabetes is accompanied by a progressive decline in β-cell function[44]. This decline might be as a result of either a reduction in the function of individual β-cells or a reduction in the number of β-cells, that is, β-cell mass (or both).

Is β-Cell Mass Reduced in Type 2 Diabetes?

Studies with classical pathology methods carried out more than 30–40 years ago established the existence of structural abnormalities in islets and some reduction in β-cell mass in diabetic patients[45–47]. This concept has recently been revived and is strongly advocated by Butler et al.[48], whose studies suggest that β-cell mass is already markedly reduced at the stage of IGT, a further deficit being apparent in overt diabetes, even when treated by diet alone. Contrasting with these dramatic data showing 50–60% reduction in β-cell mass independent of the severity of diabetes, other studies in Europe and Asia have found considerably less reduction in β-cell mass in type 2 diabetes[49–52]. Of special interest is the study by Rahier et al.[52], where *absolute* β-cell mass was estimated (Butler et al.[48] measured β-cell area, which gives only an approximation of β-cell mass). Most importantly, Rahier et al.[52] clearly showed the extraordinarily wide range of β-cell masses that exist both in the diabetic and non-diabetic populations, with a major overlap between the hyperglycemic and normoglycemic subjects (Figure 4). These observations make it difficult to ascribe a determining role to reduced β-cell mass in the etiology of hyperglycemia in type 2 diabetes. Nevertheless, it might be questioned why the high blood glucose of the patients did not stimulate β-cell mass to *increase* as a compensatory mechanism. In a physiological situation of insulin resistance, pregnancy, β-cell mass is indeed augmented by approximately 40%[53]. To our knowledge, no study has reported increased β-cell mass in type 2 diabetes.
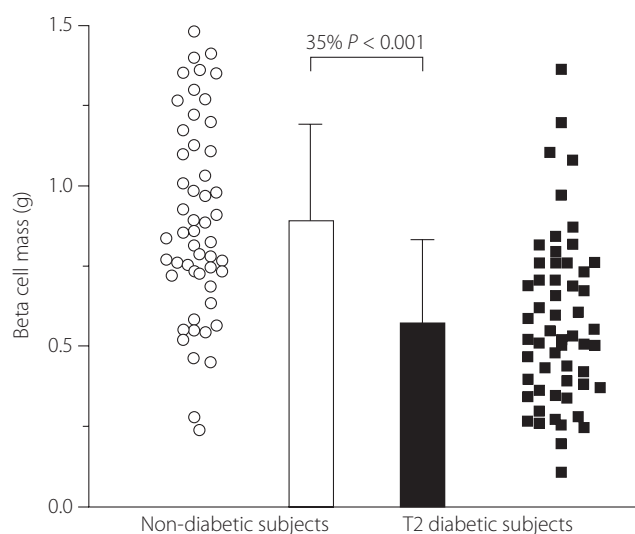

Figure 4 | β-Cell mass in type 2 diabetes. β-Cell mass in absolute terms (mg/pancreas) was measured in autopsy material from 52 non-diabetic control cases and 57 typical type 2 diabetes patients. Note that despite a statistically significant reduction of mean β-cell mass, the very large variation of the results in both groups leads to a major overlap between the diabetic and non-diabetic pancreases. Minimally modified from Rahier et al.[52], with permission.

There exist several problems that must be overcome when ascribing a role to β-cell mass changes in the development of type 2 diabetes. In addition to the technical problems relating to the quality of the pancreas obtained post-mortem, the information generated is only cross-sectional and static, and thus does not reflect the dynamics of β-cell turnover during the development of type 2 diabetes. Presently, intensive effort is being made to develop non-invasive β-cell imaging techniques; however, these are as yet in their infancy[54]. Therefore, to gain insight into the β-cell mass dynamics in type 2 diabetes, animal models of diabetes are the only tools presently available that permit detailed longitudinal observations on the pancreas. We studied this in an animal model of nutrition-dependent type 2 diabetes, the gerbil *Psammomys obesus*. These animals have an inborn insulin resistance, but retain normal glucose tolerance under caloric restriction; when given a diet with approximately 40% higher calories and low fibre content they rapidly become hyperglycemic[55]. Figure 5 shows that as the animals develop hyperglycemia, they rapidly lose their pancreatic insulin stores because the β-cells are forced to discharge all their insulin granules in the face of the unrelenting hyperglycemic stimulation. Nevertheless, β-cell mass remains normal for a considerable period; it is even slightly increased as a result of increased β-cell proliferation induced by the high glucose levels[55]. β-Cell mass collapses only after prolonged diabetes duration, with severe worsening of hyperglycemia (so-called end-stage diabetes). Thus, in this model, possibly in analogy with European and Asian type 2 diabetic patients, physiologically significant β-cell mass
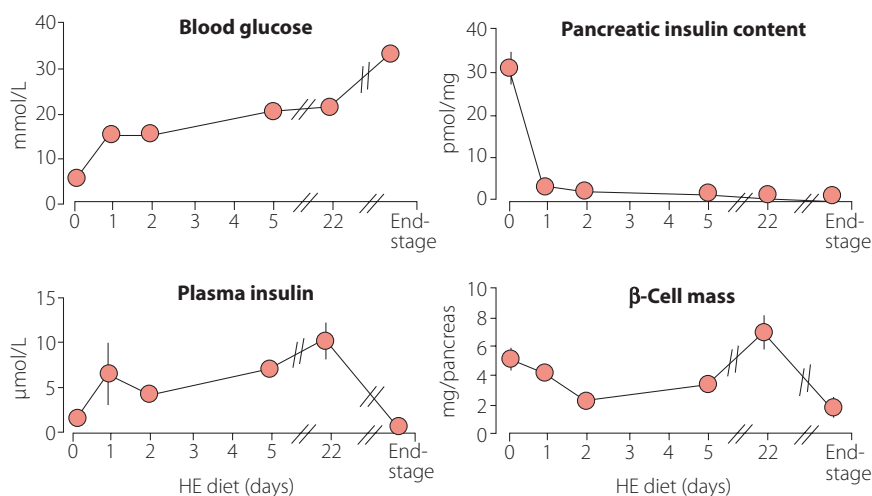
Figure 5 | Evolution of metabolic parameters in diabetic *Psammomys obesus*. Normoglycemic animals were switched from a low-energy diet to a high-energy (HE) diet at day 0. After several weeks on a HE diet, animals developed severe diabetes (end-stage). Vertical bars denote SEM; when not shown, SEM was smaller than the symbol for the mean value.

reductions occur only in long-standing and advanced type 2 diabetes. In earlier stages, the β-cell deficiency seems to be more of a functional nature. Therefore, the term 'functional β-cell mass' should be preferentially used to denote the globally insufficient insulin delivery state in type 2 diabetes, until we gain access to *in vivo* imaging techniques with the ability to determine β-cell mass *in situ*.

THE β-CELL IN CHRONIC HYPERGLYCEMIA

The close tie between hyperglycemia and β-cell dysfunction raises the possibility that elevated glucose itself is deleterious to the β-cell (so-called glucotoxicity). Near-normalization of blood glucose by short-term insulin treatment of newly diagnosed diabetic patients or of patients with secondary failure to oral hypoglycemic agents improved β-cell function[14], and a recent multicenter study showed that intensive insulin treatment in newly-diagnosed type 2 diabetic patients enhanced β-cell function, resulting in prolonged diabetes remission in approximately 50% of the patients[16]. These findings support the hypothesis that glucotoxicity plays an important role in maintaining the hyperglycemic state in type 2 diabetes. In *Psammomys obesus*, hyperglycemia is associated with a marked depletion of pancreatic insulin content, increased proinsulin/insulin ratio and β-cell apoptosis[55]. Strikingly, normalization of blood glucose using the glucosuric drug phlorizin or by changing the diabetogenic diet to a low-energy diet rapidly reversed these β-cell abnormalities[55]. This further emphasizes the importance of glucotoxicity for the β-cell dysfunction of diabetes.

The mechanisms underlying glucotoxicity are not clear. Gene expression analysis in islet preparations of patients with type 2 diabetes identified multiple changes in the expression of genes known to be important for β-cell function, including a major decrease in β-cell transcription factors, the insulin receptor and

its downstream effectors, as well as in several genes involved in glucose metabolism[56]. However, a recent study on β-cell-enriched tissue obtained by laser capture microdissection from type 2 diabetic patients failed to confirm these findings[57]. A limitation of these studies is the considerable heterogeneity in β-cell purity, and differences in degree of ischemia and stress between tissue preparations. Furthermore, the level of hyperglycemia and the response to metabolic stress can vary substantially between pancreas donors. Several mechanisms have been implicated in glucotoxicity, including inflammation, endoplasmic reticulum (ER) stress and oxidative stress[58,59]. Intricate interactions exist between the various stress pathways, which culminate in an impairment of β-cell function and survival. In humans, the β-cell turnover rate is slow[60]; however, over the years, small but persistent β-cell loss might eventually decrease the β-cell mass[48]. Cytokines, such as interleukin-1β (IL-1β), are probably involved in hyperglycemia-induced β-cell dysfunction, as shown in animal models and diabetic patients[61,62]. Notably, treatment of uncontrolled type 2 diabetic patients with IL-1β receptor antagonist improved insulin secretion and ameliorated diabetes[62]. Thus, protection against inflammatory stress might become a therapeutic strategy in diabetes.

ER Stress and β-Cell Glucotoxicity

The role of ER stress in glucose-induced β-cell dysfunction is controversial. Accumulation of misfolded proteins in the ER stimulates a signaling pathway called the unfolded protein response (UPR), which protects the cell by translational attenuation, induction of chaperone synthesis and ER-associated protein degradation (ERAD). Severe β-cell ER stress leading to strong activation of the UPR might cause apoptosis, which is mediated by stress kinases and transcription factors, such as Jun N-terminal kinase (JNK) and C/Ebp-homologous protein (CHOP).

Importantly, disruption of the CHOP gene was shown to inhibit β-cell apoptosis, expand β-cell mass and improve glycemic control in mouse models of diabetes, suggesting that the UPR plays an important role in mediating the β-cell dysfunction of diabetes[63]. We found that glucose moderately stimulates ER stress; however, high glucose levels synergize with fatty acids to stimulate UPR and JNK with β-cell apoptosis as a consequence[64]. Therefore, hyperglycemia-induced ER stress seems to become apparent mainly under conditions of glucolipotoxicity.

Increased expression of ER stress markers was observed in islets of patients with type 2 diabetes; however, the number of β-cells expressing stress markers was small[65]. Furthermore, most studies showing that ER stress is involved in β-cell dysfunction and apoptosis were carried out *in vitro*. Intriguingly, transplantation to mice of β-cell-enriched tissue derived from pancreases of non-diabetic subjects, conditions that exposed the human β-cells to mild to moderate hyperglycemia, increased the expression of UPR genes without stimulating pro-apoptotic genes[66]. Thus, UPR stimulation by hyperglycemia might be adaptive, rather than deleterious. Altogether, the conclusion that ER stress plays an important role in the β-cell dysfunction of type 2 diabetes should be drawn with caution.

Oxidative Stress and β-Cell Glucotoxicity

Chronic exposure to high glucose is expected to increase the metabolic flux in mitochondria and through the hexosamine pathway, leading to excess production of reactive oxygen species (ROS). The level of oxidative stress exerted on the β-cell depends on its capacity to scavenge ROS and other free radicals generated under conditions of glucotoxicity and glucolipotoxicity. It is widely believed that β-cells are particularly vulnerable to oxidative stress as a result of low expression of the main anti-oxidant enzymes, superoxide dismutase, catalase and glutathione peroxidase[67,68]. However, a recent report describing an effective adaptive response of diabetic GK rat islets to oxidative stress, with increased expression of anti-oxidants and high glutathione content, challenges this notion[69]. Furthermore, *in vitro* studies showed that ROS production in β-cells is maximal at low glucose concentrations when nicotinamide adenine dinucleotide phosphate (reduced; NADPH) is low, whereas it is markedly diminished by high glucose, which increases NADPH production[70]. Thus, it is possible that NADPH-dependent anti-oxidant systems operate well in β-cells and do protect the cells from oxidative stress and apoptosis under conditions of hyperglycemia.

Glutaredoxin and thioredoxin are the main redox acceptor proteins for NADPH electrons[71]. Located in distinct subcellular domains, they are highly expressed in β-cells[72]; their abundance argues against the dogma that β-cell anti-oxidant capacity is generally poor. Thioredoxin is emerging as an important anti-oxidant in the β-cell defence against oxidative stress. Thioredoxin partners with thioredoxin reductase and thioredoxin peroxidase to reduce oxidized proteins and scavenge free radicals[73]. In addition to its anti-oxidative function, thioredoxin inhibits apoptosis through interaction with signaling molecules and transcription factors, such as redox effector protein-1 (Ref-1), activator protein 1 (AP-1), nuclear factor κB (NF-κB) and apoptosis signal regulating kinase-1 (ASK1)[73].

Thioredoxin-interacting protein (TXNIP), also known as vitamin D3 upregulated protein-1 (VDUP-1) and thioredoxin binding protein-2 (TBP-2), is an endogenous inhibitor of thioredoxin, which, by binding to its redox-active cysteine residues, inhibits the anti-oxidative function of thioredoxin[74]. Under conditions of oxidative stress, TXNIP shuttles from the nucleus to the mitochondria, binds and oxidizes thioredoxin, thereby reducing its binding to ASK-1[75]. This in turn activates ASK-1 with subsequent induction of mitochondrial cell death. TXNIP expression is robustly induced by glucose in islets and β-cell lines[76–79]. Strikingly, islets derived from TXNIP-deficient mice are fully protected from glucose-induced β-cell apoptosis[80]. Such mice have increased β-cell mass and are resistant to streptozotocin-induced β-cell apoptosis and diabetes[81]. Furthermore, crossing the HcB-19 TXNIP-mutant mice with *ob/ob* mice protected against diabetes and β-cell apoptosis[81]. Similarly, we have shown that partial knockdown of TXNIP in insulinoma-1E (INS-1E) cells was sufficient to prevent β-cell apoptosis in response to high glucose[82]. This suggests that TXNIP is an important mediator of β-cell glucotoxicity.

Interestingly, TXNIP was shown to participate in the activation of nucleotide-binding oligomerization domain (NOD)-like receptor 3 (NLRP3) inflammasomes, leading to secretion of IL-1β[83]. Thus, TXNIP might provide a molecular link between hyperglycemia, oxidative stress and inflammation, and serve as an important mediator of β-cell damage in type 2 diabetes.

CONCLUDING REMARKS

The considerable clinical heterogeneity of type 2 diabetes, and the large number of genes that seem to be involved in this disease (more than 25[34]), clearly show that the pathogenesis of type 2 diabetes is not simple. That β-cell defects play a preponderant role and that clinically overt diabetes is not possible in the absence of impaired insulin production seems universally accepted nowadays. Nevertheless, β-cell deficiency in type 2 diabetic patients is modest compared with that of type 1 diabetics; it is therefore questionable whether type 2 diabetes would reach its present epidemic proportions without the concourse of additional factors; that is, an environment characterized by overfeeding and decreased exercise (an idea already discussed several decades ago[84]). Thus, type 2 diabetes is a classic example of gene–environment interaction. Indeed, what the numerous diabetes-related polymorphic alleles seem to do is to somewhat reduce the functional (and perhaps survival) capabilities of the β-cell; that is, place the cell in the lower-end of the normal variation in terms of its adaptation and resistance to the stress exerted by overfeeding and insulin resistance. By themselves, these characteristics do not define such a β-cell as abnormal or sick, nor can the aforementioned polymorphic alleles be strictly described as diabetes-causing gene variants, because subjects carrying them are free of disease unless exposed for a prolonged

time to the inappropriate environment (even then only a fraction of the population at risk develops type 2 diabetes). Indeed, it can be speculated that in a different context, for example, after World War 2 in the undernourished but physically very active populations of Europe and Asia, genome-wide association studies would probably fail to discover any of the diabetes-related gene variants hotly debated today. Our view of type 2 diabetes development is that, the greater the caloric intake and the lower the insulin sensitivity of a subject, the greater the need for the β-cells to increase their insulin output to maintain normal glucose homeostasis. To do so, the β-cells must have the capacity to continuously augment insulin secretion and proinsulin biosynthesis, and probably also expand the β-cell mass; failure to fine-tune the adaptation of means to needs ineluctably leads to impaired glucose homeostasis. It is therefore of utmost importance that the mechanisms that allow the full adaptation of β-cells to increased functional demands be well understood; enhancing these mechanisms will provide the means to prevent transition from normal glucose tolerance to IGT, that is, prevention of type 2 diabetes. Once IGT appears, glucolipotoxic events impair β-cell function and well-being, and accelerate the loss of glucose homeostasis. We are only starting to understand the mechanisms of glucolipotoxicity and to describe the molecular mechanisms involved in β-cell ER stress as well as oxidative stress. A picture is emerging ascribing to the UPR and to the TXNIP-thioredoxin couple key roles in determining whether β-cells exposed to the pressure of extended hyperglycemia succeed to adapt the functional β-cell mass to demand, or fail and undergo apoptotic cell death. However, we know nothing of gene polymorphisms that might render these stress responses adaptive or deleterious for the β-cell. This is indeed a major area of future research, with a great impact on our understanding of the pathophysiology of type 2 diabetes and on the discovery of a new generation of diabetes drugs.

As is apparent from the present discussion, most studies on the β-cell in the context of type 2 diabetes deal with various aspects of glucolipotoxicity. This is obviously relevant to β-cell fate in the diabetic environment, and thus to diabetes progression. However, it might be questioned whether the glucolipotoxicity mechanisms that are responsible for the deterioration of β-cell function and induction of β-cell death, discussed earlier, are also responsible for the initial events in the development of type 2 diabetes; that is, the transition from normal glucose tolerance to IGT. Expressed differently, the question is whether the β-cell stress mechanisms described earlier are only a secondary consequence of diabetes (glucolipotoxicity), which can be referred to as a complication of the disease, or whether similar mechanisms are also operative early in the pathogenic events that lead to the gradual loss of glucose homeostasis. Future research should answer this important question.

ACKNOWLEDGEMENTS

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