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Review

On the role of uncoupling protein-2 in pancreatic beta cells

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ABSTRACT

Pancreatic beta cells secrete insulin when blood glucose levels are high. Dysfunction of this glucose-stimulated insulin secretion (GSIS) is partly responsible for the manifestation of type 2 diabetes, a metabolic disorder that is rapidly becoming a global pandemic. Mitochondria play a central role in GSIS by coupling glucose oxidation to production of ATP, a signal that triggers a series of events that ultimately leads to insulin release. Beta cells express a mitochondrial uncoupling protein, UCP2, which is rather surprising as activity of such a protein is anticipated to lower the efficiency of oxidative phosphorylation, and hence to impair GSIS. The mounting evidence demonstrating that insulin secretion is indeed blunted by UCP2 agrees with this prediction, and has provoked the idea that UCP2 activity contributes to beta cell pathogenesis and development of type 2 diabetes. Although this notion may be correct, the evolved function of UCP2 remains unclear. With this paper we aim to provide a brief account of the present state of affairs in this field, suggest a physiological role for UCP2, and highlight some of our own recent results.

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1. Introduction

Pancreatic beta cells are important in the maintenance of blood glucose homeostasis. When plasma glucose levels are high (>5 mM), beta cells secrete insulin, which is a signal for peripheral tissues such as skeletal muscle and liver to take up or store glucose, respectively. Mitochondria play a pivotal role in glucose-stimulated insulin secretion (GSIS): transported glucose is oxidised by beta cells, which leads to an increased mitochondrial protonmotive force and cytoplasmic ATP/ADP ratio, closure of ATP-sensitive potassium channels, depolarization of the plasma membrane potential, opening of voltage-sensitive calcium channels, influx of calcium, and the eventual exocytosis of insulin-containing granules ([1]; Fig. 1). In this canonical model of events, it is important that mitochondrial electron transfer is coupled tightly to ATP synthesis, as cytoplasmic ATP/ADP is the key GSIS signal. In addition to the 'triggering' GSIS pathway shown in Fig. 1, beta cells exploit several non-canonical 'amplifying' pathways that facilitate optimal coordination of insulin secretion in response to fluctuating extra-cellular signals [2].

From a bioenergetic point of view, pancreatic beta cells are interesting for several reasons. For example, the responsiveness of the cytoplasmic ATP/ADP ratio in beta cells to glucose implies that this ratio is controlled predominantly by ATP supply, which is fundamentally different from the situation in most other cell types where ATP/ADP is governed mainly by ATP demand [3]. This specific control distribution is generally attributed to the particular hexokinase isozyme found in beta cells, 'glucokinase' [4]. As a consequence of its high control over glycolysis, glucokinase is thought to control ATP/ADP and insulin secretion almost exclusively [5]. Although the importance of this glycolytic enzyme is evident from the control it exerts over blood glucose homeostasis [4,6], this notion largely ignores the potential constraints that are imposed on GSIS by mitochondrial metabolism [7]. One such constraint may be (in)efficient oxidative phosphorylation. In this respect, another interesting bioenergetic feature of beta cells is the presence of a mitochondrial uncoupling protein, UCP2 [8]. The expression of this protein in beta cells is rather paradoxical as one would expect intuitively that UCP2 activity would uncouple mitochondrial substrate oxidation from ATP synthesis, and hence affect GSIS unfavourably. A steadily increasing body of experimental evidence indeed suggests that UCP2 impairs GSIS and as such is detrimental to beta cell function. The question therefore arises as to why the beta cell UCP2 has not landed on the evolutionary scrap heap. In other words, how is UCP2 activity beneficial to beta cell physiology?

The purpose of this paper is to provide a brief overview of research on UCP2 in beta cells, which includes whole-animal studies as well as experiments on isolated pancreatic islets and clonal beta cells. In this respect, it should be noted that cultured and primary beta cells exhibit different insulin secretion mechanisms. We put forward our thoughts on the elusive physiological function of beta cell UCP2, but first discuss some of our own recent results that highlight the importance of mitochondrial proton leak in general.

2. High proton leak in beta cells

As alluded to above, glucose-dependent fluctuations in the mitochondrial protonmotive force and cytoplasmic ATP/ADP ratio are

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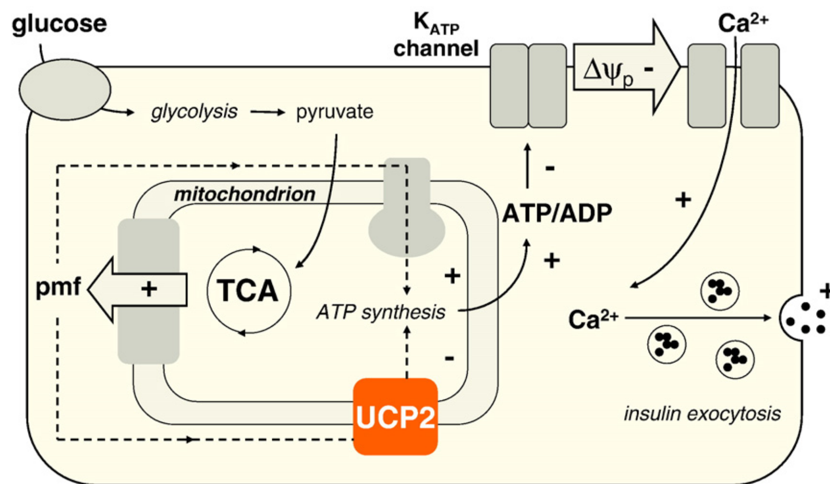

Fig. 1. Canonical model of GSIS in pancreatic beta cells. Glucose is taken up by beta cells and catabolised glycolytically. The formed pyruvate is metabolised by mitochondria, which leads to an increased mitochondrial protonmotive force (pmf) and rate of ATP synthesis. Consequently, the cytoplasmic ATP/ADP ratio rises, which causes closure of ATP-sensitive potassium channels, depolarization of the plasma membrane potential, opening of voltage-sensitive calcium channels, influx of calcium, and the eventual exocytosis of insulin-containing granules. Beta cell mitochondria contain an uncoupling protein (UCP2) that dissipates the pmf as heat thus decreasing the efficiency of oxidative phosphorylation.

observed in beta cells but not in other cell types such as muscle, where regulatory feedback loops ensure these bioenergetic parameters are relatively unresponsive to substrate supply [3]. A comparative metabolic control analysis of oxidative phosphorylation in mitochondria isolated from clonal beta cells (INS-1E) and rat skeletal muscle revealed that this difference in flexibility arises partly from mitochondrial peculiarities [9]. In particular, it appears that mitochondrial proton leak exerts, in absolute terms, approximately 7.5 times stronger control over ATP/ADP in INS-1E than muscle mitochondria (Fig. 2A). Furthermore, the control of leak over membrane potential and respiration is considerably higher in INS-1E than muscle mitochondria, roughly 13 and 8 times in absolute terms, respectively. Based on a comparative modular-kinetic analysis, we predicted that the relative strength of beta cell leak is a direct consequence of its comparably large magnitude: the contribution of leak to overall respiratory activity is nearly 3-fold higher in mitochondria from INS-1E cells than in those from rat skeletal muscle [9]. Similarly, the proton leak activity exhibited by myoblast mitochondria is at least 5 times lower, at all measured membrane potentials, than that observed in INS-1E mitochondria [10]. These substantial differences are manifested at the cellular level too, which demonstrates that they do not result from an organelle-isolation artefact. When approximated as the intracellular mitochondrial respiratory rate (i.e. myxothiazol-sensitive respiration) that is resistant to oligomycin, it appeared that the leak was nearly 4-fold higher in INS-1E cells than in myoblasts [10]. This difference is reflected in the mitochondrial coupling efficiency (i.e. the proportion of respiratory activity that is used to make ATP), which is about 25% in INS-1E cells and 90% in myoblasts (Fig. 2B). This implies that the fraction of INS-1E respiration used to drive 'futile' proton leak is roughly 75%, which is high compared to the 20–30% observed generally in other cells types [10].

Proton leak activity in isolated mitochondria is mainly constitutive and depends largely on the amount (but not activity) of the adenine nucleotide translocase [11]. This basal leak, however, is unlikely to explain the observed leak difference between INS-1E cells and myoblasts, since the adenine nucleotide translocase content is the same in these systems (C. Affourtit and M. D. Brand, unpublished observation). Some mitochondrial proton leak activity is inducible, which allows modulation of the energy transduction coupling efficiency [12]. This inducible proton conductance can be catalysed by the adenine nucleotide translocase and by specific uncoupling proteins. Because of its prominent presence in beta cells [8], it is conceivable that UCP2 accounts for the high leak activity observed in INS-1E cells. Our RNA interference experiments to test this notion directly are discussed below in relation to work reported by others.

3. UCP2 regulation

Expression of the *Ucp2* gene is upregulated by several transcription factors, including the sterol-regulatory-element-binding-protein-1c [13] and the peroxisome-proliferator-activated receptors PPARα [14] and PPARγ [15,16], as well as by the PPARγ coactivator PGC-1α [17,18]. Repression of the gene can be effected by Sirt1, a mammalian homologue of the yeast silencing information regulator-2 [19]. Furthermore, *Ucp2* is a transcriptional target of Foxa1, one of the forkhead transcription factors, which represses expression too, although it is not understood at present whether this is a direct or indirect effect [20]. Various environmental factors modulate *Ucp2* transcription: message levels are increased when beta cells are exposed to cold [17], elevated glucose or non-esterified fatty acid levels [15] and hydrogen peroxide [21], whilst it is decreased upon exposure to interleukin-1ß [22]. Most of this information on transcriptional regulation has been obtained from research on rodents, but it is becoming clear that the principal molecular mechanisms are similar in humans, despite a lack of sequence homology within the regulatory region of the respective genes [18].

It is important to realise that UCP2 mRNA and protein levels are not necessarily proportional in beta cells: a low *in vivo* protein content contrasts for example with a high amount of message, and protein increases may occur without changes in mRNA [23]. Such observations suggest that some UCP2 regulation occurs at the translational level [23], but could in principle also be explained by a high and variable rate of protein degradation. In strong favour of translational regulation, however, is the finding that a single-base substitution in the 3′ region of an upstream open reading frame within the UCP2 mRNA alters translation significantly [24]. It is this open reading frame that is indeed required for stimulation of UCP2 expression by glutamine [25], an amino acid that has been implicated in insulin secretion [26].

Generally, UCP2 and UCP3 increase the proton conductance of isolated mitochondria, but only when they are activated by superoxide, reactive oxygen species derivatives such as hydroxynonenal, or by other reactive alkenals [27]. The activity of these relatively novel carriers is inhibited by purine nucleoside di- and triphosphates [27], similar to the activity of the archetypal uncoupling protein, UCP1 [28].

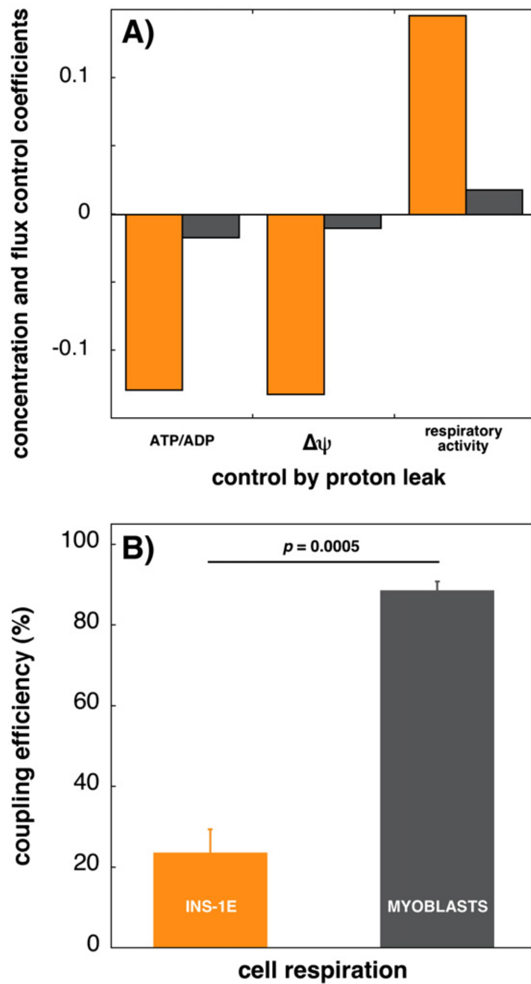

Fig. 2. High proton leak exerts relatively strong control over oxidative phosphorylation in INS-1E cells. A: coefficients quantifying control by proton leak over respiratory activity, membrane potential (Δψ), and extra-mitochondrial ATP/ADP in INS-1E and rat skeletal muscle mitochondria (orange and grey bars, respectively) were sampled from data reported in [9]. B: the coupling efficiency of INS-1E cells and C2C12 myoblasts was calculated from data reported in [10] as the percentage myxothiazol-sensitive (i.e. mitochondrial) respiratory activity that was inhibited by oligomycin ($n=4$).

Mitochondria from cultured beta cells exhibit proton leak that is stimulated considerably by superoxide in a GDP-sensitive manner [29]. Moreover, removal of endogenous superoxide causes a time-dependent increase in mitochondrial membrane potential in pancreatic islets from wild type but not *Ucp2*-ablated mice, which shows that UCP2 activity is stimulated by superoxide [30]. Together, these results demonstrate clearly that as well as the concentration, the activity of UCP2 is subject to regulation in beta cells.

It may be evident from this section that beta cell UCP2 is extensively regulated at many levels ranging from gene transcription to catalysis. One area of potential regulation that has not been explored in any detail to date, is protein degradation. In this respect, it is worth noticing that UCP2 protein in spleen, lung, and the duodenum is turned over exceptionally rapidly, exhibiting a half-life of roughly 30 min [31]. In INS-1E insulinoma cells, we find a UCP2 half-life of the same order of magnitude, approximately 60 min (V. Azzu, C. Affourtit, E. P. Breen, N. Parker and M. D. Brand, submitted for publication).

4. UCP2 controls GSIS

Because UCP2 dissipates the mitochondrial protonmotive force as heat (Fig. 1), it is intuitively likely that its activity in beta cells will blunt the response of the cytoplasmic ATP/ADP ratio to glucose, and thus attenuate insulin secretion. Evidence to support the notion that GSIS is indeed modulated by UCP2 is increasing steadily. Adenoviral overexpression of UCP2 attenuates GSIS in pancreatic islets [32] and cultured insulinoma cells [33]. Although overexpression of UCPs is generally prone to 'false positives' – because misfolded protein will increase proton conductance non-specifically [27] – these results underpin the principal idea that GSIS is impaired when oxidative phosphorylation is uncoupled. Studies involving *Ucp2*-ablated mice have shown more convincingly that UCP2 attenuates GSIS under conditions of (patho)physiological relevance [8,30,34,35]. In pancreatic islets from *Ucp2*-knockout mice, ATP levels are higher than control islets, and GSIS is increased [8]. In wild type islets, fat feeding and fatty acids lower glucose-induced increases in mitochondrial membrane potential, reactive oxygen species (ROS) production, cytosolic ATP/ADP and calcium, and insulin release. These attenuating effects of fatty acids are abolished in *Ucp2*-knockouts, which demonstrates they are mediated by this carrier [34,35]. Furthermore, GSIS is enhanced when endogenous superoxide is diminished by a superoxide dismutase mimetic, and impaired during hyperglycaemia through a mechanism involving superoxide. Both these observations are exclusive to wild type islets, again supporting a signal-mediating role for UCP2 [30].

In general, the interpretation of experiments involving *Ucp2*-knockout animals could be complicated by potentially complex developmental adaptations to the life-long absence of the *Ucp2* gene. Additionally, the observed changes in islet behaviour may have in part resulted from the absence of UCP2 in other tissues such as the brain. Of importance in this respect is the recently reported work on glucose-excitable pro-opiomelanocortin neurons that are found in the arcuate nucleus of the hypothalamus [36]. Glucose-sensing by these neurons was shown to play a role in the maintenance of blood glucose homeostasis, and to be regulated negatively by UCP2 [36]. These results illustrate nicely that beta cell behaviour may well be affected by distal UCP2 effects.

To test whether or not the high proton leak activity exhibited by INS-1E cells [10] is accounted for by UCP2, we followed an RNA interference approach to reduce its level by over 80%, and then measured consequent effects on respiration and GSIS. UCP2 knockdown lowered the intracellular mitochondrial respiratory activity that was resistant to oligomycin by approximately 30% [10]. Since we estimated that roughly 75% of INS-1E respiration is used to drive proton leak, this number suggests that UCP2 is responsible for about 20% (i.e. 0.3×0.75) of the resting respiratory activity. As a result, UCP2 knockdown significantly increases the efficiency by which INS-1E cells couple respiration to ATP synthesis (Fig. 3A). The drop in UCP2 protein did not appear to affect insulin secretion at 2 mM glucose, but increased it considerably at 30 mM (Fig. 3B). KCl-induced insulin release, which does not rely on mitochondrial metabolism, was not affected by UCP2 knockdown. These observations are in qualitative agreement with the results obtained from experiments with *Ucp2*-ablated mice, and demonstrate that GSIS is improved upon *acute* removal of UCP2.

Acute pharmacological interference with UCP2 activity also affects beta cell physiology. The aforementioned UCP2-mediated effect of endogenous superoxide scavenging on mitochondrial membrane potential and GSIS is a good example of such immediate control by UCP2 [30]. Additional support for acute UCP2 effects on islet behaviour comes from studies with genipin, a cell-permeant compound that is reported to inhibit UCP2-mediated proton leak in isolated kidney mitochondria [37]. In pancreatic islets from wild type but not *Ucp2*-knockout mice, genipin increases the mitochondrial membrane potential and cytosolic ATP, closes K_{ATP} channels, and stimulates insulin secretion [37]. Analogously, inhibition of UCP2 by genipin results in immediate excitation of pro-opiomelanocortin neurons [36].

5. Involvement of UCP2 in type 2 diabetes

Type 2 diabetes is a common metabolic disorder that is reaching pandemic proportions [38]. The disease is polygenic, influenced strongly

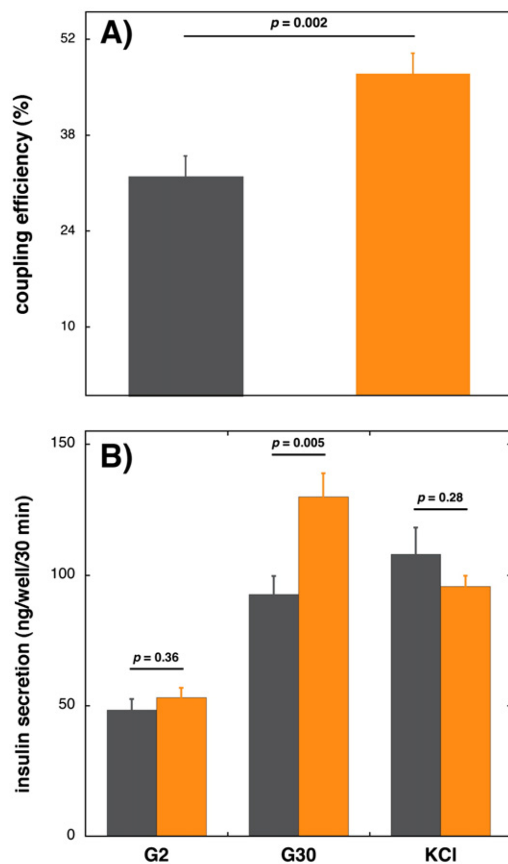

Fig. 3. UCP2 knockdown increases coupling efficiency and enhances GSIS. These data are an expansion of results reported in [10] and were obtained from experiments with INS-1E cells transfected with scrambled (grey bars) or UCP2 siRNA (orange bars). A: coupling efficiencies (*cf.* Fig. 2B) are means ± S.E.M. of twelve (scrambled siRNA) and twenty-two (pooled siRNAs targeted at three separate *Ucp2* exons) experiments. B: insulin secretion was measured at 2 and 30 mM glucose (G2 and G30) and 2 mM glucose + 30 mM KCl (KCl). Data are means ± S.E.M. of nine (G2 and G30) and fifteen (KCl) experiments with each condition assayed four to eight times.

by environmental factors such as diet and exercise, and results from defects in the function of both peripheral tissues and pancreatic beta cells. Beta cell dysfunction is widely believed to be secondary to prolonged exposure to high glucose and lipid levels [39,40], conditions that are often associated with obesity and insulin-resistance. Although the mechanism of beta cell glucotoxicity and lipotoxicity is not fully understood, reactive oxygen species are likely to be involved [41,42], as such species are increased during both hyperglycaemia [30,43] and hyperlipidaemia [35,44,45]. Superoxide production is increased accordingly in diabetic animal models [30,43]. Interestingly, the presence and activity of UCP2 appear to correlate rather closely with these phenomena. UCP2 expression is upregulated by high glucose [15,30] and fatty acid [15,32,34,46] levels, and is relatively high in mouse models of type 2 diabetes [8,47–49] and lipotoxicity [50,51]. In addition, UCP2 activity is stimulated by superoxide [30]. From these observations, the idea has emerged that UCP2 plays an important role in beta cell pathology and the development of type 2 diabetes [38,52–55].

The putative involvement of UCP2 in the aetiology of type 2 diabetes is supported by experiments that show ablation of UCP2 activity rescues the impaired beta cell function of various diabetic mouse models. For example, leptin-deficient *ob/ob* mice exhibit obesity-induced diabetic symptoms that are ameliorated upon cross-breeding with *Ucp2*-ablated animals: first-phase insulin secretion is restored, serum insulin levels are increased, whilst blood glucose levels are greatly decreased [8,30]. Type 2 diabetes can also be modelled by subjecting pancreatic islets chronically to high glucose levels or by feeding mice a high-fat diet. The impaired GSIS that results from such treatments is greatly diminished in *Ucp2* knockout mice ([30] and [34,35], respectively). Similarly, beta cell dysfunction caused by hyperglycaemia or leptin-deficiency is reversed acutely upon pharmacological UCP2 inhibition by genipin [37]. Acute knockdown of UCP2 by RNAi partially restores GSIS in a beta cell lipotoxicity mouse model that overexpresses the sterol-regulatory-element-binding-protein-1c [51]. In *ob/ob* mice, short-term RNAi-mediated UCP2 knockdown does not improve GSIS, which seems discrepant with the *Ucp2* knockout data, although GSIS is enhanced in wild type control animals [49]. In a different study, however, the hyperglycaemic syndrome exhibited by *ob/ob* mice *was* improved by administration of *Ucp2* antisense oligonucleotides [56]. This metabolic benefit, which was also observed in mice rendered diabetic through high-fat feeding, appears not only to be due to increased insulin secretion by beta cells, but also to enhanced insulin action on peripheral tissues [56]. In this respect, it is interesting to note that pro-opiomelanocortin neurons lose their ability to sense glucose as a result of a high-fat diet, and that this loss is restored by acute UCP2 inhibition, or prevented by genetic *Ucp2*-ablation [36].

The corroborative evidence for the suggested role of UCP2 in beta cell pathogenesis is extensive. It may nonetheless be clear that the evolved function of UCP2 is unlikely to be pathological. From an evolutionary perspective, UCP2 activity in beta cells must have a physiological outcome that is beneficial to the organism under certain conditions.

6. The elusive physiological role of UCP2

Regarding the physiological relevance of novel mitochondrial UCPs, it has been proposed that they have an ancestral function of protection against superoxide production and consequent damage [57,58]. This 'mild uncoupling' hypothesis is based on the observations that matrix superoxide production from complex I exhibits a steep dependence on the protonmotive force, and that matrix superoxide stimulates UCP-mediated proton conductance. Activation of UCPs thus provides a negative feedback loop attenuating further superoxide production, at the expense of a slightly lowered energy transduction efficiency. It is predicted by this model that UCP-specific mild uncoupling occurs under conditions that allow endogenous superoxide production and, conversely, that impaired UCP activity increases the mitochondrial membrane potential, ROS production, and oxidative damage. In pancreatic islets, *Ucp2*-ablation increases the membrane potential as well as superoxide production; the effect of genetic knockout on membrane potential is mimicked by conditions under which superoxide levels are lowered with a cell-permeant superoxide dismutase mimetic [30]. These results are consistent with the notion that UCP2 performs a protective task against oxidative damage in beta cells. In this scenario, UCP2-mediated beta cell dysfunction and development of type 2 diabetes are pathological side effects of a damage limitation mechanism that is induced during prolonged periods of hyperglycaemia and hyperlipidaemia. A defensive role of UCP2 against ROS may be necessitated by the striking lack of expression and activity of antioxidant enzymes in beta cells [59–61]. However, such a function does not agree with observations that *Ucp2* knockout mice exhibit improved glucose homeostasis for an extended period of time and do not suffer beta cell loss, despite having elevated ROS levels [30].

The intrinsically low antioxidant levels in beta cells may facilitate a role for ROS in cell signalling. It is becoming increasingly clear in this respect that physiological, non-damaging ROS levels are important in apoptosis, kinase activation, immune response, calcium signalling and gene expression (listed in [62]). It seems therefore reasonable to postulate that ROS are involved in the regulation of beta cell physiology [63]. For example, a fluctuating superoxide level could modulate UCP2 activity, and thereby provide a signal that could be relevant under both pathological and physiological conditions. Our recent results concerning the relatively high contribution of UCP2 to INS-1E respiration, its attenuating effect on GSIS ([10]; *cf.* Fig. 3), and the significant control exerted

by proton leak over the cytoplasmic ATP/ADP ratio ([9]; *cf*. Fig. 2A), are consistent with this role. We suggest that the relatively high leak exhibited by beta cells (Fig. 2B) amplifies the effect of physiological regulators of UCP2 proton conductance (i.e. superoxide) on the ATP/ADP ratio and hence on GSIS.

UCP2 activity may be important to coordinate the physiological response of beta cells to fluctuating nutrient supply. A signalling role of UCP2 could be important to restrict insulin secretion when blood glucose levels are low, for example during sleep (Fig. 4A). The circulation of non-esterified fatty acids is increased under such conditions, which will cause both UCP2 expression [15] and activation by superoxide [30] that is formed upon fatty acid oxidation [45,64]. Despite the availability of an adequate substrate for ATP production, insulin secretion will consequently decrease, which prevents the occurrence of hypoglycaemia in a fasting state [55,57]. After a meal, on the other hand, blood glucose and lipid concentrations will rise considerably, again causing expression [15] and superoxide activation of UCP2 [30,45]. Under these conditions, insulin release needs to be increased markedly. Such an increase will indeed happen as an immediate, triggering response to the elevated glucose level, although it may not be as pronounced as intuitively expected because of seemingly undesirable UCP2 activity. However, to sustain a boosted insulin secretion rate for as long as it may take to deal appropriately with the postprandial nutrient load, signals additional to an increased ATP/ADP are required to amplify the initial GSIS [2,65]. These potentiating signals may include NADPH, acetyl-CoA, malonyl-CoA, and α-ketoglutarate, the production of which depends on TCA cycle activity [66]. When substrate supply is high, turnover of this cycle is likely to be limited by the mitochondrial protonmotive force. UCP2 activity may be important during periods of nutrient abundance to alleviate this backpressure and thus allow TCA cycle turnover to occur uncoupled from ATP synthesis (Fig. 4B). In this view of events, GSIS is sustained at the expense of blunted first-phase insulin secretion. The degree of this initial attenuation is not anticipated to be high, however, since nutrient-induced UCP2 expression and subsequent activation are likely to be slower events than first-phase GSIS, which lasts only a few min [2]. This potential role of UCP2 to facilitate unrestricted electron transfer for metabolic purposes (i.e. to produce GSIS amplifiers) is not without precedent, as the plant UCP performs a similar function, allowing mitochondrial metabolism when required during photorespiration in *Arabidopsis thaliana* [67].

If the exclusive physiological role of UCP2 in beta cells is indeed to lower GSIS during periods of fasting and, conversely, to allow sustained GSIS when nutrients levels are high, then its activity would need to be dampened rather promptly under intermediate conditions, i.e. between meals. This may be achieved by virtue of the exceptionally short half-life of UCP2 [31] that would ensure rapid protein degradation when glucose and lipid concentrations have reattained their resting level, and hence *de novo* UCP2 synthesis has stopped. UCP2 levels and activity would thus correlate with physiologically relevant nutrient fluctuations. In this functional model, the chronic exposure to hyperglycaemia and hyperlipidaemia that eventually leads to beta cell dysfunction may be explained by the *persistent* nature of the UCP2-mediated drop in ATP/ADP under pathological conditions, and perhaps also by a gradual deterioration of GSIS-amplifying mechanisms. The pronounced beneficial effect of *Ucp2*-ablation on glucose homeostasis in diabetic animal models is likely to be a combined effect of enhanced first-phase insulin secretion [8], increased insulin action on peripheral tissues [56], and improved behaviour of glucose-sensing neurons [36].

7. Concluding remarks

The evidence for a GSIS-regulatory role of UCP2 is extensive and convincing, which demonstrates that the efficiency of mitochondrial energy metabolism exerts significant control over GSIS, and weakens the notion that insulin secretion is controlled exclusively by glucokinase. Involvement of UCP2 in beta cell pathogenesis and type 2 diabetes is underpinned by strong experimental support, whereas its physiological role remains speculative. We think it would be sensible to establish the evolved function of UCP2 in beta cells, if this carrier is to fulfil its promise as a safe therapeutic target in the treatment or prevention of type 2 diabetes. In this respect, it is clear that our functional model requires

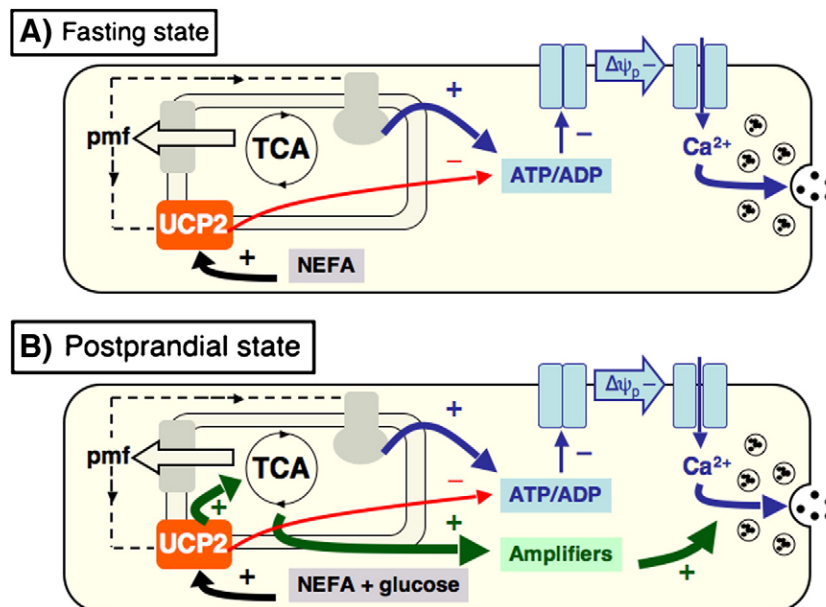

Fig. 4. Putative involvement of UCP2 in coordinating the response of beta cells to physiological fluctuations in nutrient supply. In the fasting state (A) blood glucose levels are low and circulation of non-esterified fatty acids (NEFA) rises. These free fatty acids increase UCP2 expression and stimulate its activity either directly (e.g. by nucleotide displacement) or indirectly by superoxide that is formed during beta oxidation. The mitochondrial protonmotive force (pmf) that results from NEFA oxidation is consequently blunted and canonical insulin secretion (blue route) attenuated, which protects against hypoglycaemia. After a meal (B) glucose and NEFA levels are high, which requires insulin release. UCP2 activity under these conditions would facilitate increased and sustained GSIS (green and blue routes) because UCP2 allows continued production of TCA cycle-derived amplifying signals even though the pmf is high in the abundant presence of nutrients.

rigorous experimental testing, which should reveal whether or not UCP2 regulates insulin secretion in response to physiologically relevant fluctuations in nutrient supply.

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