

Topical Review

A reappraisal of the blood glucose homeostat which comprehensively explains the type 2 diabetes mellitus–syndrome X complex

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Blood glucose concentrations are unaffected by exercise despite very high rates of glucose flux. The plasma ionised calcium levels are even more tightly controlled after meals and during lactation. This implies 'integral control'. However, pairs of integral counterregulatory controllers (e.g. insulin and glucagon, or calcitonin and parathyroid hormone) cannot operate on the same controlled variable, unless there is some form of mutual inhibition. Flip-flop functional coupling between pancreatic α - and β -cells via gap junctions may provide such a mechanism. Secretion of a common inhibitory chromogranin by the parathyroids and the thyroidal C-cells provides another. Here we describe how the insulin:glucagon flip-flop controller can be complemented by growth hormone, despite both being integral controllers. Homeostatic conflict is prevented by somatostatin-28 secretion from both the hypothalamus and the pancreatic islets. Our synthesis of the information pertaining to the glucose homeostat that has accumulated in the literature predicts that disruption of the flip-flop mechanism by the accumulation of amyloid in the pancreatic islets in type 2 diabetes mellitus will lead to hyperglucagonaemia, hyperinsulinaemia, insulin resistance, glucose intolerance and impaired insulin responsiveness to elevated blood glucose levels. It explains syndrome X (or metabolic syndrome) as incipient type 2 diabetes in which the glucose control system, while impaired, can still maintain blood glucose at the desired level. It also explains why it is characterised by high plasma insulin levels and low plasma growth hormone levels, despite normoglycaemia, and how this leads to central obesity, dyslipidaemia and cardiovascular disease in both syndrome X and type 2 diabetes.

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While the actions and physiological effects of insulin and glucagon are known in great detail, the homeostat to which these hormones belong is poorly understood. Why is there normally no hint of hypoglycaemia during exercise (Ahlborg, 1969; Felig & Wahren, 1975; Zinman *et al.* 1977; Winder *et al.* 1979; Koeslag *et al.* 1980, 1982; Kjaer *et al.* 1991; Roy *et al.* 1991; Coggan *et al.* 1995), when a glucose drink causes moderate hyperglycaemia? Why is there apparently no syndrome associated with a deficiency of the hormone which so effectively prevents hypoglycaemia during exercise, when absence of the slower responding insulin is life threatening? In fact, why are there two counterregulatory hormones – a system also used to regulate, amongst other things, the plasma ionised calcium level – when one would appear to be enough? What is the stimulus for the hyperinsulinaemia of syndrome X (Reaven, 2002; Meigs, 2002; Hayden, 2002) when the

blood sugar concentration is still normal? What is the purpose of the functional syncytiality between the α -, β - and D-cells of the Islets of Langerhans? What are the roles of GABA, pancreastatin and somatostatin in these islets? How can the accumulation of amyloid in these islets (a characteristic feature of type 2 diabetes mellitus) cause peripheral insulin resistance? Here we review the data pertaining to these questions and present a synthesis which not only answers these questions, but also suggests new therapeutic approaches to syndrome X and type 2 diabetes.

Zero steady state error

A standard negative feedback system responds only when the sensor detects an error in the value of the controlled variable. The greater the error the greater the response (Cannon, 1960; Riggs, 1963, 1970; Milsum, 1966; Guyton & Hall, 1996). Such a system is said to exhibit proportional

control. In particular, a zero error always produces a zero response. Thus glucagon levels could only be raised, and the insulin levels lowered during exercise, if the blood sugar concentration is lower than normal. Yet the blood sugar concentration remains remarkably constant (i.e. the same as at rest) or may even be slightly higher than normal during exercise (Ahlborg, 1969; Felig & Wahren, 1975; Zinman *et al.* 1977; Winder *et al.* 1979; Koeslag *et al.* 1980, 1982, 1985; Kjaer *et al.* 1991; Roy *et al.* 1991; Coggan *et al.* 1995) despite very high rates of glucose utilisation.

This suggests integral control (Milsum, 1966; Koeslag *et al.* 1997, 1999; Saunders *et al.* 1998, 2000), for which direct experimental evidence (summarised in Fig. 1) is provided by a variety of hyperglycaemic clamp studies (Grodsky, 1972; Gerich *et al.* 1974; Bolaffi *et al.* 1986; Tsuchiyama *et al.* 1992). An integral controller responds not simply to the static error in the value of the controlled variable, but to the error multiplied by the time it persists (i.e. the time integral of the error). It is the only known type of homeostat that is capable of eliminating the error altogether, rather

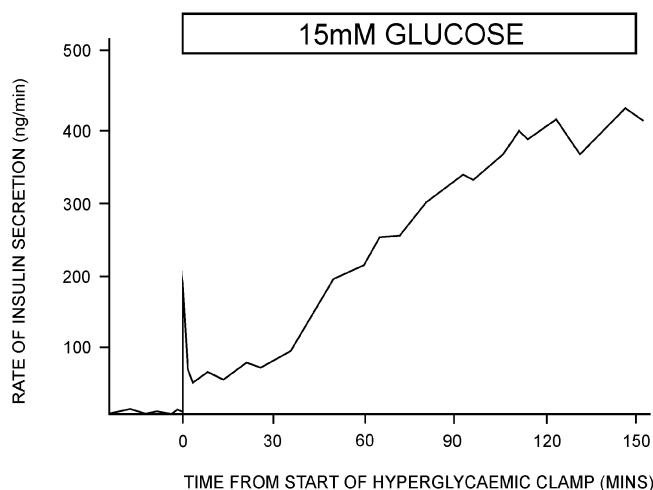


Figure 1. The result of an archetypical hyperglycaemic clamp experiment (Grodsky, 1972; Gerich *et al.* 1974; Bolaffi *et al.* 1986; Tsuchiyama *et al.* 1992)

When the endocrine pancreas is exposed to an abrupt hyperglycaemic clamp (e.g. 15 mmol l^{-1}), there is an initial spike of insulin release lasting less than 10 min. This is followed by a prolonged 'second phase' of progressively increasing rates of insulin secretion for more than 2 h. This 'second phase' is characteristic of an integral response, in which the rate of insulin release is not determined by the static error in the blood sugar concentration (in this case, $15 - 5 = 10 \text{ mmol l}^{-1}$), but by the error multiplied by the time that the error persists (i.e. the 'time integral of the error'). Since the error in a hyperglycaemic clamp experiment remains constant, the time integral of the error increases with time. The rate of insulin secretion therefore increases as a function of time, as shown. An abrupt hypoglycaemic clamp (1 mmol l^{-1}) also produces a spike in glucagon secretion, similarly followed by a gradual rise in glucagon secretion (Weir *et al.* 1974). Reproduced by kind permission of the Society for Endocrinology from Koeslag *et al.* (1997).

than merely reducing it (as is the case with a standard controller), thus achieving what is known as zero steady state error (ZSSE) (Milsum, 1966).

If, in response to a constant stress, a controller progressively increases its negative feedback activity in proportion to the error multiplied by its duration (see Fig. 1), it will only cease *escalating* its negative feedback activity when the error has returned to zero. At this stage the effectors will be operating at very different levels from those before the onset of the stress. Superficially it may therefore appear that there is no stimulus for this different level of effector activity, as the controlled variable is at set point.

For example, a motorist maintaining a constant speed on a road over undulating terrain, applies integral speed control via the throttle. At the first indication of a reduction in speed on an up-hill gradient the motorist opens the throttle and *continues opening it further and further* until the desired speed is re-attained. The accelerator pedal is then held in this new position till the road levels off.

Integral control is thus able to achieve the very close control that is observed in the case of blood glucose and plasma ionised calcium levels. What is more, while for the engineer integral control is generally more complicated to build into a system than is proportional control, this is not the case in physiological systems because it arises naturally when regulation involves chemical rate equations (Saunders *et al.* 2000).

There is, however, a problem. In many cases, including the regulation of the blood glucose level and plasma ionised calcium concentration, there are two controllers: the insulin-mediated and the glucagon-mediated homeostats in the former and the calcitonin and parathyroid hormone (PTH) systems in the latter. One element of the pair acts to increase the substance in question, the other acts to decrease it. They cannot simply be acting as mutual backups, for, if they were, type 1 diabetes mellitus and hypoparathyroidism would occur only if both controllers were to fail. These diseases would therefore be expected to be very rare. But since they are not, the two mechanisms must be combining in some way that does not involve backup.

Clynes (1969) has described how a pair of counteracting controllers can act together and has given the name 'rein control' to the resulting mechanism. Rein control solves the problem caused by the fact that the rate of secretion of a hormone can be rapidly increased or decreased, but its removal from the blood cannot be similarly adjusted. Thus, the blood insulin concentration might remain inappropriately high if, after a carbohydrate meal, the absorbed glucose is cleared from the blood faster than expected. The rapid release of a counterregulatory hormone (in this case glucagon) would then prevent the otherwise inevitable reactive hypoglycaemia.

While such a combination works well if the controllers are proportional, things are different if they are integral. An integral controller will escalate its negative feedback activity until its set point has been reached. Because the set points of two integral controllers are unlikely ever to be exactly the same, at least one of the controllers will always register an error. It will respond by attempting to move the system towards its set point. This will bring the other member of the pair into action, which will force the first one to increase its activity and so on. This homeostatic conflict eventually results in both controllers making large and unnecessary responses, and homeostasis breaking down.

'Integral rein control' offers a solution (Koeslag *et al.* 1997, 1999; Saunders *et al.* 1998, 2000). If the two integral controllers are linked in such a way that an increase in the activity of one induces a decrease in the activity of the other, the runaway effect is prevented. (Examples are described below). Loss of one member of such a linked pair spells disaster, accounting for diseases such as type 1 diabetes mellitus and hypoparathyroidism. In contrast, the set point of a pair of counterregulatory proportional controllers would hardly be affected by the loss of one of the hormones.

The regulation of blood glucose

The blood glucose concentration is regulated by two counteracting controllers, insulin and glucagon. If these two hormones are parts of an integral rein control system then they must be linked.

The human islets of Langerhans contain glucagon-secreting α -cells, insulin-secreting β -cells and somatostatin-secreting D-cells. These cells are characterised by membrane specialisations involving tight junctions, desmosomes and gap junctions (Orci *et al.* 1973, 1975; Bonner-Weir, 1991; Genuth, 1993). Molecules smaller than 1000 Da can move from the cytoplasm of one cell to that of another through the gap junctions without entering the intercellular space. Such junctions have been found between α - and β -cells (Orci & Unger, 1975; Orci *et al.* 1975; Orci, 1976; Meda *et al.* 1982), as well as between D-cells and α - or β -cells (Raskin *et al.* 1976; Unger *et al.* 1978; Michaels & Sheridan, 1981; Meda *et al.* 1982; Bonner-Weir, 1991). Thus gap junctions make functional syncytia of small groups of homologous and heterologous islet cells (Meda *et al.* 1982; Palti *et al.* 1996). Each islet consists of a collection of such syncytial aggregates.

It is now well established that the β -cells are glucose sensitive. Intense investigation into the molecular mechanism by which this is accomplished indicates that it is the rate of glucose metabolism, and consequent cytosolic ATP concentration in the β -cells, which is responsible for generating the signal for insulin secretion (Ashcroft *et al.* 1994; Dunne *et al.* 1994; Bell *et al.* 1996; Schuit *et al.* 2001).

Briefly, pancreatic β -cells express GLUT2 glucose transporters whose relatively low affinity for glucose ensures that the rate of glucose entry into the cell is proportional to the extracellular glucose concentration, at least in the physiological range. At low blood glucose concentrations, when very little glucose enters the cell, very little of that glucose is phosphorylated (Schuit *et al.* 1999), probably because of the low expression in these cells of any high affinity hexokinase isoforms (hexokinase I, hexokinase II, or hexokinase III). At blood glucose levels above 2.5 mmol l⁻¹ β -cells phosphorylate glucose via glucokinase (hexokinase IV), which, since it is not inhibited by glucose-6-phosphate, ensures a proportionality between the intracellular glucose-6-phosphate concentrations and the extracellular glucose concentration (Schuit *et al.* 2001), particularly in the physiological range. Differential usage of two alternative glucokinase gene promoters keeps enzyme expression levels much lower in pancreatic β -cells than in liver parenchymal cells (Magnuson & Shelton, 1989), making glucokinase activity in β -cells the rate limiting step for further glucose metabolism (Bell *et al.* 1996; Matschinsky *et al.* 1998; Schuit *et al.* 2001).

Unlike most other mammalian cell-types, pancreatic β -cells express low lactate dehydrogenase levels (Sekine *et al.* 1994; Schuit *et al.* 1997) and high pyruvate carboxylase levels (Schuit *et al.* 1997). This means that most of the products of glycolysis enter the mitochondria, and that there is therefore tight coupling between glucose-6-phosphate formation and ATP production (Prentki *et al.* 1997; Wollheim, 2000). The ATP thus formed acts as an intracellular ligand for ATP-sensitive K⁺ (K_{ATP}) membrane channels (Aguilar-Bryan & Bryan, 1999). ATP binding to these channels causes their closure, with consequent depolarisation of the cell membrane. This, in turn, opens L-type voltage-dependent calcium channels, leading to the exocytosis of insulin-containing granules (Prentki & Matschinsky, 1987; Ashcroft *et al.* 1994; Berggren & Larsson, 1994; Dunne *et al.* 1994).

For any given β -cell, insulin biosynthesis appears to be all or none (Schuit *et al.* 1988; Pipeleers, 1992; Pipeleers *et al.* 1994). Glucose is a well-known stimulus of proinsulin biosynthesis. In purified β -cells, the presence of glucose produces a 25-fold increase in the synthesis of immunoreactive proinsulin over a 60 min incubation period (Schuit *et al.* 1988). Autoradiographic analysis of individual cells shows, however, that this effect is achieved via a dose-dependent recruitment of pancreatic β -cells to biosynthetic activity. Recruitment of β -cells is also seen in isolated islets exposed to glucose. The sigmoidal dose-response curve for glucose-induced proinsulin biosynthesis thus reflects a heterogeneous responsiveness of pancreatic β -cells, rather than progressively increasing activity of a population of fundamentally homogeneous cells (Schuit *et al.* 1988; Pipeleers *et al.* 1994). A similar

β -cell heterogeneity is seen with respect to the intracellular Ca^{2+} -induced insulin secretory pulses (Hellman *et al.* 1994). Glucose produces sudden transitions, in individual β -cells, between basal and raised intracellular Ca^{2+} concentrations. The thresholds at which these transitions occur differ in different cells. It is thus concluded that glucose stimulation of insulin release is determined by the number of cells that have entered into a state of Ca^{2+} -induced secretory pulses, rather than by a uniform graded response of the entire β -cell population (Hellman *et al.* 1994; Pipeleers *et al.* 1994).

The incretin hormones, glucose-dependent insulinotropic peptide (GIP) and glucagon-like peptide 1 (GLP-1), amplify glucose-induced insulin secretion, probably by increasing the cytosolic cAMP concentrations of β -cells (Schmidt *et al.* 1985; Pipeleers *et al.* 1994; Schuit *et al.* 2001). Since cAMP does not increase the rate of glucose oxidation of these cells, it might be involved in the exocytosis of secretory granules (Ämmälä *et al.* 1993). GABA stimulates insulin secretion and inhibits glucagon secretion (Gerber & Hare, 1980; Garry *et al.* 1986; Rorsman *et al.* 1989, 1991; Adeghate & Ponery, 2002). It is known to bind to, and open, chloride channels (Rorsman *et al.* 1989), but the downstream molecular events within the α - and β -cells are unknown. Somatostatin-28 and, to a lesser extent, somatostatin-14 produced by the pancreatic D-cells inhibit pancreatic islet insulin and glucagon release by binding to somatostatin-specific SSTR5 and SSTR2 receptors, respectively (Zambre *et al.* 1999; Kimura *et al.* 2001). These are linked via guanine nucleotide binding proteins (G-proteins) to multiple cellular effector systems, which mediate, amongst others, reduction in the conductance of voltage-dependent Ca^{2+} channels (Reisine & Bell, 1995).

The mechanisms for the stimulus–secretion coupling in the α -cells are unclear (Schuit *et al.* 2001). Although rat islet α -cells express the β -cell form of glucokinase (Heimberg *et al.* 1996), glucose uptake occurs considerably more slowly via GLUT1 transporters (instead of GLUT2 transporters), and anaplerosis (the entry of pyruvate into the mitochondrion via the pyruvate carboxylase reaction) is much more restricted in α -cells than in β -cells (Heimberg *et al.* 1995; Schuit *et al.* 1997). Variations in glucose concentrations from 1 to 10 mmol l⁻¹ therefore do not result in changes in ADP/ATP ratio in fluorescence-activated cell-sorter-purified α -cells (Heimberg *et al.* 1995; Detimary *et al.* 1998). Glucose inhibition is therefore unlikely to be mediated by ATP binding to ATP-sensitive membrane channels. Although *in vitro* studies have indicated that purified α -cells are to some extent glucose sensitive (Wang & McDaniel, 1990; Sumida *et al.* 1994), it is unclear whether variations in glucose concentration in the physiological range have a direct effect on α -cells *in vivo* (Asplin *et al.* 1981; Rorsman & Hellman, 1988; Rorsman *et al.* 1989, 1991).

The most powerful inhibitory effects in the physiological range seem to originate from the islet environment. Such inhibitory influences include somatostatin-28 and -14 originating from neighbouring D-cells (Schuit *et al.* 1989; Brunnicardi *et al.* 2001), GABA secretion from β -cells (Gerber & Hare, 1980; Garry *et al.* 1986; Rorsman *et al.* 1989, 1991; Adeghate & Ponery, 2002), and insulin (Samols & Stagner, 1988; Van Schravendijk *et al.* 1987; Ito *et al.* 1995). Another important possibility is that α -cells receive their glycaemic cues directly from the β -cells, via their common gap junctions (Orci *et al.* 1975; Michaels & Sheridan, 1981; Meda *et al.* 1982, 1986; Palti *et al.* 1996; Koeslag *et al.* 1997). Indeed, the α -cells of patients with insulin-dependent diabetes mellitus continue to secrete glucagon at normal resting rates despite severe hyperglycaemia (Unger *et al.* 1970; Raskin *et al.* 1976; Liu *et al.* 1991; Powell *et al.* 1993; Smismans *et al.* 1997). These patients also do not increase glucagon secretion during hypoglycaemia (Asplin *et al.* 1981; Bolli *et al.* 1983; Rorsman & Hellman, 1988; Powell *et al.* 1993). The absence of β -cells, therefore, renders the α -cells glucose insensitive. Indeed, Ishihara *et al.* (2003) have shown that although α -cells possess some inherent capacity to respond to blood nutrient levels, secretion from α -cells is normally suppressed by simultaneous activity of β -cells. Zinc released from active β -cells is believed to be implicated in this suppression (Ishihara *et al.* 2003).

Since the β -cells operate as on–off units (Hellman *et al.* 1994; Pipeleers *et al.* 1994) and are functionally linked to α -cells via gap junctions, then each syncytial unit probably operates as a flip-flop mechanism, which secretes either insulin or glucagon (Ishihara *et al.* 2003). Either hormone would, according to this suggestion, be secreted, by a given syncytial unit, maximally or not at all. Only under ancillary circumstances would a syncytial unit be modulated via autonomic nerves, the gut hormones, or other influences, to secrete both hormones simultaneously, or, under the influence of, particularly, somatostatin-28 to shut down completely (Kawai *et al.* 1982; Pipeleers *et al.* 1985; Rorsman *et al.* 1989; Zambre *et al.* 1999; Laedtke *et al.* 2000; Brunnicardi *et al.* 2001; Kimura *et al.* 2001).

Pipeleers *et al.* (1985) have demonstrated that at extracellular glucose concentrations of 1.4 mmol l⁻¹ isolated β -cells secrete 2–3 times more insulin than do β -cells in undissociated islet tissue. At hyperglycaemic glucose concentrations (20 mmol l⁻¹), isolated cells secreted 4–5 times less insulin than β -cells in intact islets, despite identical rates of glucose transport and oxidation in the purified cells and intact islets. The responses of isolated cells to somatostatin, leucine and adrenaline were also blunted compared to cells in intact islets. The response to glucagon was, however, exaggerated. Pipeleers *et al.* (1985) present evidence that the poor glucose-induced insulin release from single β -cells was unlikely to have been due to

damage sustained during the purification process. Importantly, the secretory defect could be fully restored by recombining the separated islet cells, particularly by the addition of glucagon-containing α -cells. This strongly suggests that it is the α - β -cell syncytium that constitutes the fundamental functional unit of the endocrine pancreas, and not its individual cell types (Palti *et al.* 1996). We denote this functional unit by the letters AB, and its two most important functional states as Ab (glucagon-secreting) and aB (insulin-secreting).

Note that, if the α - and β -cells form syncytia that can flip-flop between the Ab and aB modes of activity, this provides the link that is required for integral rein control.

According to our model (Fig. 2), Ab \rightarrow aB and aB \rightarrow Ab transitions occur spontaneously, but are influenced by paracrine secretions (probably pancreastatin and GABA) from neighbouring units (Koeslag *et al.* 1997). GABA, which is cosecreted with insulin, stimulates insulin secretion, and strongly inhibits glucagon secretion (Gerber & Hare, 1980; Garry *et al.* 1986; Rorsman *et al.* 1989, 1991; Adegate & Ponerly, 2002). It would thus be the agent which promotes Ab \rightarrow aB transitions. Pancreastatin, a powerful paracrine inhibitor of insulin secretion (Schmidt *et al.* 1988; Winkler & Fischer-Colbrie, 1992; Aunis & Metz-Boutigue, 2000), we propose, does the opposite. Under normoglycaemic circumstances GABA and pancreastatin are equally effective at promoting Ab \rightarrow aB and aB \rightarrow Ab transitions, respectively, thereby ensuring that the insulin:glucagon ratio in the blood remains unchanged. Low blood sugar levels, we propose, potentiate the effectiveness of pancreastatin (relative to that of GABA) causing a greater number of aB \rightarrow Ab transitions than Ab \rightarrow aB transitions; whereas high blood sugar concentrations promote the effectiveness of GABA (relative to that of pancreastatin) causing the rate of Ab \rightarrow aB transitions to exceed that of the aB \rightarrow Ab transitions.

The lowered insulin and raised glucagon levels that characterise prolonged exercise, despite normal blood glucose concentrations (Ahlborg, 1969; Felig & Wahren, 1975; Zinman *et al.* 1977; Winder *et al.* 1979; Koeslag *et al.* 1980, 1982, 1985; Kjaer *et al.* 1991; Roy *et al.* 1991; Coggan *et al.* 1995), would, we propose, come about as follows. Assuming, for the sake of simplicity, no feedforward or anticipatory influences, a sudden increase in the rate of glucose utilisation by the exercising muscles will cause an initial small reduction in the blood glucose level. This will enhance pancreastatin's paracrine actions over those of GABA. The number of aB \rightarrow Ab transitions in the pancreatic islets will therefore exceed the number of Ab \rightarrow aB transitions. The blood glucagon concentration therefore rises, and that of insulin falls. The number of Ab units will continue to increase (at the expense of the aB units) for as long as the blood sugar level is below set point, which could, during exercise, have been set at a slightly higher

level than that of the resting state, via the autonomic nervous system or circulating catecholamine levels. The blood insulin level therefore continues to fall, and the glucagon level to rise, until the set point blood sugar level has been re-attained. At this stage the majority of islet syncytial units will be in the Ab (glucagon-secreting) mode. Very few will be in the aB (insulin-secreting) mode. This situation persists for as long as there is no change in the rate of glucose utilisation, and the blood glucose level remains at set point.

At set point, pancreastatin and GABA are intrinsically equally effective at promoting their aB \rightarrow Ab and Ab \rightarrow aB transitions, respectively. However, during exercise the islets contain more pancreastatin than GABA. (Pancreastatin is secreted by the majority Ab units, and GABA by the minority aB units.) Thus, if, for example, 90 out of 100 syncytial units are in the Ab mode, and 10 are in the aB mode, then there will be 9 times more pancreastatin in the islets than GABA. This means that Ab units have only a 10% probability of flipping to the aB state, while the aB units have a 90% likelihood of flipping to the Ab state. Thus, during a given arbitrary time interval, 9 of the 90 Ab units will flip to the aB state, while 9 out of the original 10 aB units will flip to the Ab state. This flip-flopping therefore does not change the 90:10 ratio of Ab to aB units while the blood glucose level is at set point. This contrasts

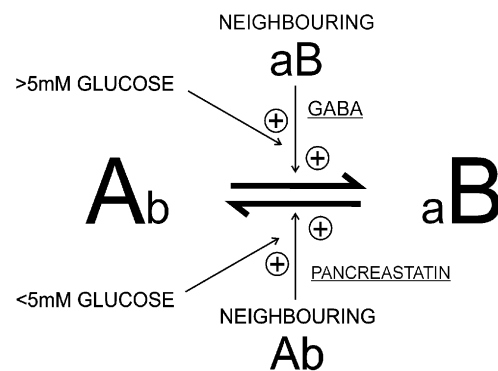


Figure 2. The proposed model of pancreatic islet function

The functional units of the endocrine pancreas are suggested to be sub-islet heterologous syncytial aggregates of electrically coupled α - and β -cells which operate as flip-flop mechanisms. They are either in the Ab (glucagon-secreting) or aB (insulin-secreting) mode. Ab \rightarrow aB and aB \rightarrow Ab transitions occur spontaneously, but are influenced by paracrine secretions from neighbouring units. The relative effectivenesses of the two types of paracrine secretion are determined by the blood sugar concentration. A rise in the blood sugar level above 5 mmol l⁻¹ promotes the effectiveness of GABA relative to that of pancreastatin, whereas a blood sugar level below 5 mmol l⁻¹ promotes pancreastatin's effectiveness. A fall in the blood sugar level below 5 mmol l⁻¹ will therefore produce a progressive increase in the number of Ab units (at the expense of aB units), which comes to a halt (i.e. no further increase in the number of Ab units) only when the blood sugar concentration normalises. Reproduced by kind permission of the Society for Endocrinology from Koeslag *et al.* (1997).

with the maintenance of the blood glucose level at rest (also at set point) when the islet $\text{Ab}:\text{aB}$ ratio remains stable at, say, 50:50, causing a markedly different ratio of glucagon:insulin output from the pancreas. At the end of exercise, when the rate of glucose utilisation starts to fall, any deviation of the blood glucose level above set point will immediately lead to a greater number of $\text{Ab} \rightarrow \text{aB}$ than $\text{aB} \rightarrow \text{Ab}$ transitions. The resting situation is thus gradually restored, with the beginning and end blood glucose levels not differing from one another. Even the intermediate blood glucose levels need hardly deviate from set point, if changes in the rate of glucose utilisation are not too abrupt. This is ZSSE control.

Integral rein control of the blood sugar level also explains why 'insulin resistance' (whatever the cause) results in hyperinsulinaemia even when the blood sugar levels are normal. Blood sugar levels that, as a result of insulin resistance, remain elevated longer than usual after a meal will promote an excessive number of $\text{Ab} \rightarrow \text{aB}$ transitions, which continue to predominate over $\text{aB} \rightarrow \text{Ab}$ transitions until the blood sugar level normalises. The $\text{aB}:\text{Ab}$ ratio at the point when the blood sugar level normalises depends on the degree of insulin resistance. If normoglycaemia can only be achieved by a higher than normal resting plasma insulin level, then the integral rein controller will only come to rest at the islet $\text{aB}:\text{Ab}$ ratio that provides that degree of hyperinsulinaemia. That $\text{aB}:\text{Ab}$ ratio (and its corresponding insulin:glucagon output from the pancreas) will then persist for as long as the insulin resistance remains unchanged. The standard model of glucose homeostasis (Schuit *et al.* 2001) would predict that since the blood sugar level is now normal, β -cell ATP production will also normalise, with the result that the K_{ATP} membrane channels will open, and the excessive insulin secretion will cease. Since all the other glucose sensors in the body would respond in the same way, there is no mechanism, in the standard model, whereby insulin resistance would not inevitably be accompanied by hyperglycaemia.

Note that the term 'insulin resistance' is generally used to describe any situation in which more insulin than normal is required to control the blood sugar level. The implication is that insulin's target tissues have become less sensitive to insulin, by, for instance, the down-regulation of insulin receptors. This we would term true insulin resistance. Clinical insulin resistance could, however, also result from the need for extra insulin to counter the metabolic effects of the hyperglucagonaemia which characterises syndrome X and type 2 diabetes mellitus (Reaven *et al.* 1987; Baron *et al.* 1987; Rorsman *et al.* 1989; Larrson & Ahrén, 2000), as well as type 1 diabetes mellitus (Unger *et al.* 1970; Raskin *et al.* 1976; Asplin *et al.* 1981; Bolli *et al.* 1983; Rorsman & Hellman, 1988; Liu *et al.* 1991; Powell *et al.* 1993; Smismans *et al.* 1997; Adegate *et al.* 2000). Our proposed glucose homeostat responds to both

forms of 'insulin resistance', as well as to those seen in acromegaly, pregnancy and Cushing's syndrome, in the same manner.

If GABA is involved in pancreatic islet function in the manner suggested, then a lack of pancreatic GABA would be expected to result in type 1 diabetes mellitus. In fact, autoantibodies against glutamic acid decarboxylase (or GAD, the enzyme which catalyses the production of GABA from glutamic acid) are present in over 80% of newly diagnosed patients with type 1 diabetes, often before other autoantibodies, such as insulin autoantibodies and islet cell antibodies become detectable (Baekkeskov *et al.* 1990; Yokota & Shima, 1998; Adegate & Ponery, 2002). It is, however, not known how autoantibodies against this intracellular enzyme develop, nor whether such antibodies affect the enzymes within intact β -cells (Baekkeskov *et al.* 1990; Adegate & Ponery, 2002). The finding of GAD antibodies in type 1 diabetes mellitus might therefore mean nothing more than that β -cells have been damaged. On the other hand, pancreatic β -cells express major histocompatibility complex (MHC) class I molecules which present self-peptides to the immune system (Baekkeskov *et al.* 1990). This suggests that some, as yet unknown, problem with the GAD enzyme might lead to the production of antibodies against it. The abnormality in the enzyme might then also cause reduced GABA synthesis. Anti-GAD antibodies would then aggravate this situation, producing type 1 diabetes mellitus before all the β -cells have been destroyed. Thus, it has been shown that GABA receptor agonists are capable of increasing plasma insulin levels even in diabetic rats (Gomez *et al.* 1999), strongly suggesting that in at least some forms of type 1 diabetes mellitus the inability to secrete insulin, particularly in the early stages, is due to an absence of pancreatic GABA, supporting the notion that GABA is important for normal flip-flop activity in the pancreatic islets (Koeslag *et al.* 1997).

The set point of an integral rein control system is determined dynamically, at the point at which the two controllers are balanced. Consequently, if one of the controllers fails, as in type 1 diabetes, there is no longer a set point. In fact, the remaining mechanism actively drives the system to an equilibrium that is far removed from the physiological set point (Koeslag *et al.* 1997; Saunders *et al.* 1998). This is why individuals with type 1 diabetes find it difficult to achieve precise control by injecting insulin, even on a regimen of regular diet and exercise. Their problem is not simply that they are not secreting insulin, but that the destruction of the β -cells also causes inappropriate, glucose-insensitive glucagon secretion (Unger *et al.* 1970; Raskin *et al.* 1976; Asplin *et al.* 1981; Bolli *et al.* 1983; Rorsman & Hellman, 1988; Liu *et al.* 1991; Powell *et al.* 1993; Adegate *et al.* 2000). Apart from driving the blood glucose to very high levels, this causes, additionally, a form of insulin resistance, while, simultaneously, providing

no protection against hypoglycaemia (Asplin *et al.* 1981; Bolli *et al.* 1983; Powell *et al.* 1993). Their only defence against hypoglycaemia is emergency adrenaline secretion, which appears to operate as a 'threshold regulator' (i.e. is triggered only when the blood sugar level falls to dangerously low levels), and therefore does not contribute to the maintenance of the blood sugar at its normal set point of about 5 mmol l⁻¹.

Anatomically remote glucose sensors and homeostatic conflict

Human growth hormone (hGH) is secreted from the adenohypophysis in response to, amongst other stimuli, exercise, fasting and hypoglycaemia (Koeslag *et al.* 1980, 1982; Koeslag, 1982). It has many actions on glucose metabolism which are similar to those of glucagon, and the two tend to rise to similar extents in response to exercise, fasting and hypoglycaemia, as well as amino acid ingestion (Koeslag *et al.* 1980, 1982, 1985). The blood glucose sensors responsible for hGH release are located in the hypothalamus, but their mechanism of glucose responsiveness has not yet been fully elucidated (Schuit *et al.* 2001). Importantly, however, they monitor the blood glucose concentration independently from the β -cells (Schuit *et al.* 2001). This is a recipe for homeostatic conflict.

A similar problem afflicts the plasma ionised calcium homeostat. The counterregulatory hormones, PTH and calcitonin, are secreted by anatomically remote endocrine tissues each with its own calcium sensors. We have proposed that homeostatic conflict, in this instance, is probably prevented by both glands co-secreting a species of chromogranin that strongly inhibits the endocrine activity of both tissues (Koeslag *et al.* 1999; Saunders *et al.* 2000; Helle *et al.* 2001). Thus increased activity in the one gland is then always at the expense of activity of its counterregulatory partner, creating an integral rein controller, capable of ZSSE control (Koeslag *et al.* 1999; Saunders *et al.* 2000). It relies on the same fundamental operating principles as the insulin:glucagon integral rein controller.

Thus, a possible reason why glucagon and hGH can combine in the integral control of the blood glucose level without causing homeostatic conflict is that the pancreatic islets and the hypothalamic-pituitary axis responsible for hGH secretion share a common endocrine statin, somatostatin-28.

The roles of somatostatin and hGH

Pancreatic islets have one of the highest levels of somatostatin-like immunoreactivity in the body (Arimura *et al.* 1975; Unger *et al.* 1978; Rorsman *et al.* 1989; Zambre *et al.* 1999). It is present at the capillary poles of the D-cells (Unger *et al.* 1978; Zambre *et al.* 1999). Release of somatostatin-28 and, to a lesser extent, somatostatin-14 into the pancreatic vein is stimulated by perfusion of the

pancreas with high concentrations of glucose (Schauder *et al.* 1976; Ipp *et al.* 1977; Weir *et al.* 1977; Berts *et al.* 1996) and other insulin secretagogues (Unger *et al.* 1978). Like the other islet hormones, somatostatin-28 secretion is pulsatile (Stagner *et al.* 1980). These pulses are in phase with the insulin pulses (Stagner *et al.* 1980; Jaspan *et al.* 1986; Matthews *et al.* 1987). Since groups of β - and D-cells are functionally interconnected by gap junctions (Unger *et al.* 1978; Michaels & Sheridan, 1981; Meda *et al.* 1982; Bonner-Weir, 1991), it is highly probable that somatostatin-28 secretion is physically linked to insulin secretion (Unger *et al.* 1978). This therefore suggests that somatostatin-28 is playing a role similar to that proposed for the chromogranins in plasma ionised calcium homeostasis (Koeslag *et al.* 1999; Helle & Aunis, 2000; Helle *et al.* 2001).

Somatostatin-28 is a potent circulating inhibitor of insulin, glucagon and hGH secretion (Srikant & Patel, 1981; Tannenbaum *et al.* 1982; Kawai *et al.* 1982; Pipeleers *et al.* 1985; Schuit *et al.* 1989; Rorsman *et al.* 1989; Zambre *et al.* 1999; Laedtke *et al.* 2000; Brunicardi *et al.* 2001; Kimura *et al.* 2001). Mole for mole it is ten times more powerful at inhibiting insulin, glucagon and hGH secretion than somatostatin-14 which is believed to have primarily a paracrine function in the pancreas (Mandarino *et al.* 1981; Tannenbaum *et al.* 1982; Genuth, 1993). Since α - and β -cells are equally sensitive to somatostatin-28 (Schuit *et al.* 1989), it effectively determines how many pancreatic AB units are active at any one time. There is no evidence to suggest that it changes the relative rates of $\text{Ab} \rightarrow \text{aB}$ and $\text{aB} \rightarrow \text{Ab}$ transitions. In other words, high plasma somatostatin-28 levels decrease the blood concentrations of both insulin and glucagon by reducing the overall functional size of the endocrine pancreas (Matthews *et al.* 1987), creating a family of 'ab' (i.e. inactive) units, without influencing the concentration of glucose at which the remaining active units flip equally in the one direction ($\text{Ab} \rightarrow \text{aB}$) as in the other ($\text{aB} \rightarrow \text{Ab}$). But this is precisely the set point of the integral rein controller. So the steady state concentration of blood glucose is independent of the somatostatin-28 concentration (Fig. 3). A high somatostatin-28 concentration in the blood merely reduces the pancreas' rate of response to a change in glucose flux, as well as its capacity to respond to large long-term changes in glucose flux.

The hypothalamus reduces hGH secretion by secreting more somatostatin-28, and increases it by putting out less. (There is also a hypothalamic hGH releasing factor, but, since it is not believed to affect the pancreas directly, it is unlikely to be relevant in the present context.) Changes in the rate of hGH secretion are therefore associated with, but in the opposite direction to, changes in the rate of somatostatin-28 secretion.

The complete insulin-glucagon-hGH homeostat is diagrammatically described in Fig. 4.

If the blood sugar level falls, then the rate of insulin secretion immediately decreases, while those of glucagon and hGH increase. The fall in insulin secretion and the rise in hGH secretion are both accompanied by a reduction in the blood somatostatin-28 concentration. This immediately recruits active syncytial units from the inactive 'ab' pool (see Fig. 3). These will all tend to flip to the Ab mode, as a result of the low blood sugar concentration. This somatostatopenically augmented response will rapidly raise the blood sugar level. But as it rises, the inhibition of insulin secretion is lifted, while the opposite happens to hGH. The blood somatostatin-28 concentration therefore rises again, until equilibrium is restored at the blood glucose set point.

If the challenge to the blood sugar level is due to a longer-term perturbation, for example prolonged exercise, the levels of the three hormones, insulin, glucagon and hGH will stabilise at values very different from those under

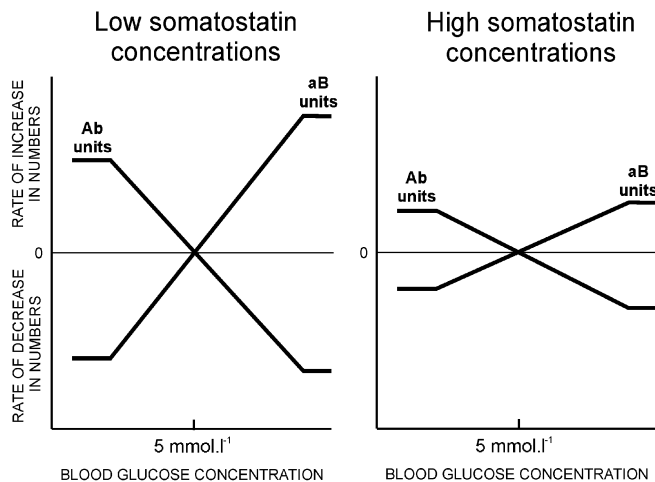


Figure 3. A diagrammatic representation of the effect of the plasma glucose and somatostatin-28 concentrations on the mechanism described in Fig. 2

Ab \rightarrow aB and aB \rightarrow Ab transitions occur at all blood glucose concentrations. Since the system operates as a flip-flop mechanism, the point at which the number of Ab \rightarrow aB transitions equals the number of aB \rightarrow Ab transitions is also the point at which the rate of increase of both is zero. Since this is the only equilibrium point, it defines the homeostat's set point (5 mmol glucose l⁻¹). At blood glucose levels below 5 mmol l⁻¹ Ab units increase in number at the expense of aB units; at blood glucose levels above 5 mmol l⁻¹ the opposite happens. Somatostatin-28 inhibits both insulin and glucagon secretion. It probably does so by inactivating a proportion of the α - β -cell syncytial units, thereby reducing the overall functional size of the endocrine pancreas. At high plasma somatostatin-28 concentrations there are, therefore, fewer flip-flopping units, resulting in the graph on the right. The point at which the number of Ab \rightarrow aB transitions equals the number of aB \rightarrow Ab transitions is, however, not affected by the plasma somatostatin-28 concentration. (Although we assume that somatostatin inhibits insulin and glucagon secretion by reducing the number of active syncytial units, all forms of inhibition produce the graph on the right.)

resting conditions, but the blood glucose level will always return to the same ZSSE set point, as will the plasma somatostatin-28 level (Fig. 4).

If an individual is incapable of producing hGH, the glucagon–insulin system will still be capable of maintaining the desired set point of 5 mmol glucose l⁻¹ (Fig. 3), though the response to hypoglycaemia may be slower than normal. If, on the other hand, the production of glucagon is impaired, then hGH can combine with insulin to produce an alternative integral rein control system, using somatostatin-28 as the intermediary that establishes the required link, similar to the proposed chromogranin link between PTH and calcitonin secretion (Koeslag *et al.* 1999).

Note that the intact glucose homeostat responds far more vigorously to hypoglycaemia than to hyperglycaemia. This results from the unusual relationship between somatostatin-28 secretion and hGH secretion on the one hand, and insulin secretion on the other. If somatostatin-28 was co-secreted with both hormones, in the way that chromo-

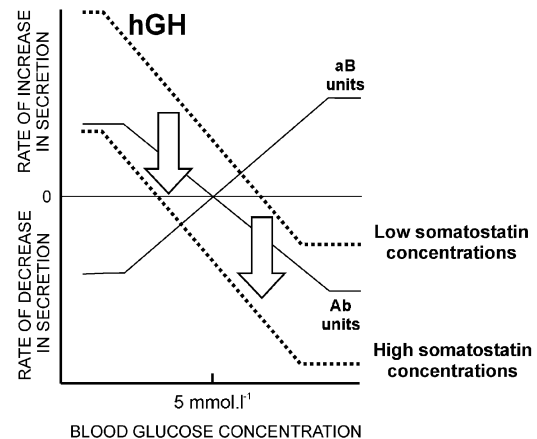


Figure 4. A schematic representation of the effect of the plasma somatostatin-28 concentration on the rate at which growth hormone (hGH) secretion increases or decreases at different blood glucose concentrations

The response of the endocrine pancreas to different blood glucose concentrations (Fig. 3) is depicted in the background. Somatostatin-28 inhibits hGH secretion. It therefore depresses the hGH response curve in the manner indicated by the arrows: a given hGH response requires a much lower blood glucose level when the somatostatin-28 concentration is high than when it is low. From the graph it is clear that there is a unique intermediate somatostatin-28 concentration that causes the hGH response curve to cross the zero rate of change line at exactly the same point as the pancreatic Ab–aB cross-over point. At this unique somatostatin-28 concentration the entire insulin–glucagon–hGH homeostat is in equilibrium. It determines the blood glucose set point, which, in the diagram, is indicated to be 5 mmol glucose l⁻¹. Any stressor that causes a deviation of the blood sugar level away from set point elicits a disequilibrium that has as its effect the return of the blood sugar concentration to set point. If the stress persists (e.g. prolonged exercise) the blood sugar and plasma somatostatin levels will always return to their equilibrium values, but the plasma levels at which insulin, glucagon and hGH stabilise will be different from those at rest.

granin is co-secreted with both PTH and calcitonin (Helle & Aunis, 2000), then the responses to hypo- and hyperglycaemia would be symmetrical.

Supplementary controllers

Any controller, whether proportional or integral, can be supplemented with feedforward and threshold regulators without risking homeostatic conflict. Feedforward regulators produce anticipatory responses usually to non-glycaemic cues. Thus the stimulus for the secretion of the incretin hormones GIP and GLP-1 is food in the gastrointestinal tract. This anticipates and enhances the insulin response to glucose absorbed from the gut. (The mechanism of action is described above.) Any under- or over-shoot of such an anticipatory response (which is, by definition, temporary) is soon corrected by the integral rein controller.

The suppression of insulin secretion, and stimulation of glucagon secretion by the sympathetic nervous system in anticipation of, and during, intense muscular activity (Schuit & Pipeleers, 1986; Havel *et al.* 1991; Hamilton-Wessler *et al.* 1994; Adeghate *et al.* 2000) is similarly a feedforward response which temporarily overrides the homeostat, ensuring that exercise is not accompanied by an initial (albeit slight) dip in the blood glucose level.

High circulating adrenaline levels, in addition to suppressing insulin secretion (Schuit & Pipeleers, 1986; Hamilton-Wessler *et al.* 1994; Adeghate *et al.* 2000), have a potent anti-insulin effect on the metabolism which is independent of that induced by glucagon and hGH. Adrenaline is released into the circulation in response to many non-glycaemic cues. None of these cause homeostatic conflict. At worst, chronically high blood adrenaline levels cause 'insulin resistance' (Rösen & Rösen, 1998), which is readily accommodated by an intact homeostat. There are, however, independent glucoreceptors located particularly in the portal vein and liver (Sawchenko & Friedman, 1979; Nijijima, 1986; Donovan *et al.* 1991; Berthoud *et al.* 1992; Hamilton-Wessler *et al.* 1994), which, when they detect hypoglycaemia, cause a vigorous sympathetic response, including adrenaline release (Hamilton-Wessler *et al.* 1994). If this reflex is triggered only in response to a rapid fall in the blood glucose level, then it constitutes an emergency system (or 'threshold regulator'), which supplements the integral rein controller, without hindering its normal operation.

A new explanation of type 2 diabetes mellitus

Type 2 diabetes mellitus, or non-insulin-dependent diabetes mellitus (NIDDM), is characterised by glucose intolerance, insulin resistance and impaired insulin responsiveness to elevated blood glucose levels. Despite its uncertain, and apparently confusing pathogenesis, the most substantial and uniform morphological aspect of the disease with respect to the islets of Langerhans is the deposition of amyloid (Westermarck & Wilander, 1978; Cooper *et al.* 1987; Kahn, 2000; Hayden, 2002). Such deposits, exclusively

limited to the islets of Langerhans, occur in over 90% of patients with NIDDM (of the maturity onset type). The severity of the diabetic state appears to be directly related to the amount of amyloid deposited in the islets of Langerhans (Kahn, 2000). Islet amyloid also sometimes occurs in elderly 'normal' subjects, but since this is always a post-mortem finding, mild or undiagnosed diabetes cannot be excluded retrospectively. In contrast, it is extremely rare in type 1 diabetes mellitus, and in the enzyme-deficiency forms of type 2 diabetes mellitus (Bell *et al.* 1996).

The islet amyloid observed in NIDDM contains the 37-amino acid islet amyloid polypeptide (IAPP), or amylin (Westermarck *et al.* 1987; Cooper *et al.* 1987). Amylin is a normal secretory product of the β -cell that is co-secreted with insulin. The reason why this normal secretory product forms insoluble amyloid fibrils in patients with NIDDM but not in healthy subjects is not known (Kahn, 2000). However, its deposition round the β -cells destroys many of these cells (Westermarck & Wilander, 1978; Kahn, 2000), and disrupts the interaction of the surviving cells with the other islet cells. The regular rhythmic pulses of insulin secretion every 8–15 min of normal persons, are replaced by an irregular pattern of basal insulin output from the pancreas (Lang *et al.* 1981; O'Rahilly *et al.* 1988; O'Meara *et al.* 1993; Laedtke *et al.* 2000; Nyholm *et al.* 2000). Since the normal pulses occur in isolated pancreases exposed to a constant glucose concentration (Stagner *et al.* 1980; Matthews *et al.* 1987), there must be not only intra-islet but also inter-islet β -cell communication to co-ordinate the rhythm over the whole pancreas. This is clearly disturbed in NIDDM. Impaired pulsatile secretion of insulin has also been described in first degree relatives of patients with NIDDM, and in normoglycaemic patients with impaired glucose tolerance (Lang *et al.* 1981; O'Rahilly *et al.* 1988; O'Meara *et al.* 1993; Schmitz *et al.* 1997). Since these people are predisposed to NIDDM, these findings suggest that amyloid deposition leading to disruptions in intercellular communication in the pancreatic islets might be a very early or even the primary defect in this disease.

Other pathophysiological characteristics of NIDDM are: impaired stimulation of insulin secretion and suppression of glucagon by glucose (Kipnis 1969; Unger *et al.* 1970; Dimitriadis *et al.* 1985; Rorsman *et al.* 1989; Powell *et al.* 1993), impaired stimulation of glucagon secretion by hypoglycaemia (Bolli *et al.* 1984; Dimitriadis *et al.* 1985), hyperglucagonaemia throughout the day (Starke *et al.* 1984; Reaven *et al.* 1987; Rorsman *et al.* 1989; Solerte *et al.* 1999; Larrson & Ahrén, 1996, 2000), and low plasma hGH concentrations (Kjeldsen *et al.* 1975; Reaven *et al.* 1987; Lee *et al.* 1999).

These features can all be explained in terms of our model if it is assumed that the accumulation of amyloid in the islets of Langerhans causes disruption of not only inter- β -cell

communication but also of the communication between β -cells and α -cells, and β -cells and D-cells. Those α -cells that lose their functional syncytiality with β -cells would be expected to become glucose insensitive (Unger *et al.* 1970; Raskin *et al.* 1976; Powell *et al.* 1993). Not only would they not respond to hypoglycaemia, they would also continue secreting glucagon inappropriately at normal and high blood glucose concentrations. This would have an unrelenting anti-insulin effect on metabolism, which, it is now recognised, is an important factor in the pathogenesis of the insulin resistance that clinically characterises NIDDM (Reaven *et al.* 1987; Baron *et al.* 1987; Rorsman *et al.* 1989; Larrson & Ahrén, 2000). In the early stages of the disease, while there are still adequate numbers of functional α -, β - and D-cell syncytial units, normoglycaemia can be maintained, but only with higher than expected plasma insulin levels. However, since amyloid accumulation reduces total β -cell mass (Westermarck & Wilander, 1978; Kahn, 2000), and the rate of glucagon secretion cannot be reduced in the normal manner, the patient will also be glucose intolerant. The glucagon response to hypoglycaemia is similarly impaired (Bolli *et al.* 1984; Dimitriadis *et al.* 1985), because it can involve only α -cells that are still functionally linked to β -cells; the autonomous ones are already secreting glucagon.

As the disease progresses, the proportion of autonomous α -cells will become so great that normoglycaemia can no longer be maintained on a normal diet. The surviving functional units will be predominantly in the aB mode, leading, in addition to the resting hyperinsulinaemia, to a markedly blunted insulin response to any further glycaemic challenge. The patient now has frank NIDDM.

The persistently high blood insulin levels operating on glucagon-insensitive tissues (e.g. muscle) will result in insulin-receptor down-regulation in these tissues, aggravating the insulin resistance of NIDDM. It is this component of the insulin resistance of NIDDM which is rapidly, though temporarily, alleviated by exercise.

In the pre-diabetic stages of the disease, also referred to as syndrome X, metabolic syndrome, or, simply, insulin resistance (Reaven, 2002; Meigs 2002; Hayden 2002), the hyperinsulinaemia would be accompanied by a high somatostatin-28 output from the pancreas. (Somatostatin-28 is functionally co-secreted with insulin.) This will inhibit hGH secretion from the adenohypophysis. Since hGH deficiency in adults is now recognised as a syndrome with distinct features, such as central adiposity, decreased lean body mass, reduced bone mineral density, disturbed lipoprotein metabolism, increased intimal thickening of the arteries and early signs of atherosclerosis (Markussis *et al.* 1992; Carroll *et al.* 1998), it is possible that when these features are seen as frequent accompaniments of syndrome X and NIDDM, they are the result of the somatostatin-

induced low blood hGH levels. When frank type 2 diabetes develops, the low blood hGH levels will be driven lower still by the persistent hyperglycaemia. This then accentuates the central obesity, dyslipidaemia and atherosclerosis.

DISCUSSION

The integral rein control model of blood glucose homeostasis is able to account for a number of phenomena which otherwise appear anomalous. It explains, for the first time, the physiological importance of the D-cells in the islets of Langerhans. The model parsimoniously provides a comprehensive explanation of most of the homeostat's known features and behaviour, and how loss of insulin-secreting capacity causes severe disease, whereas loss of either glucagon or hGH causes no apparent disturbance of blood glucose homeostasis. We have also proposed important roles for islet GABA and pancreastatin secretion, and the critical significance of the gap junctions between the α -, β - and D-cells, which, until now, has never been understood.

Our synthesis also suggests, for the first time, a unifying hypothesis for the origin and pathophysiology of syndrome X, and its progression into NIDDM, taking into account the accumulation of amyloid in the pancreatic islets, and how that leads to hyperglucagonaemia, insulin resistance, glucose intolerance, and impaired insulin responsiveness to elevated blood glucose levels. It explains why these conditions are frequently characterised by low plasma hGH levels, and how this may lead to central obesity, dyslipidaemia, and increased risk of cardiovascular disease. Finally, we note that Pepys *et al.* (2002) have reported that R-1-[6-[R-2-carboxypyrrolidin-1-yl]-6-oxo-hexanoyl]-pyrrolidine-2-carboxylic acid (CPHC) promotes amyloid regression. Our results suggest that if CPHC or a similar drug proves effective in the treatment of advanced NIDDM, it may also be an effective treatment in the early stages, hopefully preventing the disease from progressing to the later ones. Alternatively, a glucagon receptor blocker would be a rational therapeutic agent in syndrome X and NIDDM, as this would relieve the insulin resistance and probably prevent most of NIDDM's complications. Such a glucagon receptor blocker would be effective whether the disruption of intra- and inter-islet communication is caused by amyloid or any other agent. The proposed integral rein control mechanism of glucose homeostasis suggests that any disruption of intra- and inter-islet communication will lead to NIDDM.

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