The pancreas and diabetes mellitus

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Few theories about diabetes gain universal acceptance, and the role attributed to the pancreas varies from a major to a very minor one in the opinions of different groups of workers. Two facts that are generally accepted are that diabetes can be caused by pancreatectomy, and that its symptoms can be alleviated by insulin administration. The relative lack of insulin which is implied by these two statements is, however, not necessarily of pancreatic origin in every case of diabetes. It could be caused by a defect at any point along the pathway followed by insulin between its production by the β cell and its final effect on peripheral cell metabolism. In discussing the abnormality in insulin-carbohydrate interaction, the following points should be considered:

- (1) The islet cell.
- (2) Insulin structure.
- (3) Insulin transfer.
- (4) Insulin destruction.
- (5) Insulin antagonism.
- (6) Endorgan responsiveness.
- (7) β cell control.

We should like to discuss the pathogenesis of the relative insulin lack found in diabetes mellitus under these headings, laying more stress on those factors which directly concern the pancreas.

(1) The islet cell

(a) Normal β cell mass

β cell mass has been studied directly by morphometric techniques, and indirectly by measuring pancreatic insulin content. Derivation of the β cell mass from histological observations is hindered by the variation in islet content in different regions, by the recognition of β cells in different stages of granulation and by the assumptions involved in calculating volume from two-dimensional measurements of irregular structures.

Measurement of total insulin content of the pancreas is less complicated, and has been shown to correlate well with β cell staining (Hartroft & Wrenshall, 1955). However, the important functional parameter is insulin production, not content or cell mass, particularly as the most active β cells are probably degranulated, containing no insulin, and not recognizable histologically. Despite these difficulties and the lack of dynamic observations, some conclusions can be drawn from the numerous static measurements made.

Morphological measurements have been made by numerous workers, and despite criticism (Tejning, 1947) of the method used, Ogilvie's work represents a major contribution. He found in controls (Ogilvie, 1937) that the total islet cell mass increases on average from 0.12 g at birth to 1.07 g at 21 years, and thereafter remains constant. Wrenshall, Bogoch & Ritchie (1952), in a careful autopsy study, showed that the extractable insulin correlated well with body surface area and that total insulin content increased during childhood, reached adult levels between the years of 12 and 17, thereafter only increasing slightly up to the age of 50 years.

(b) The islet cell in adult and juvenile onset diabetes

In adult onset diabetes the classical lesions of islet hyalinization and vacuolar degeneration have received much attention in the past. The quantitative changes in the islets are, however, probably of more importance than the qualitative. The two most comprehensive quantitative studies of the pancreatic islets in diabetes are those of MacLean & Ogilvie (1955) and Gepts (1957). The islets are of approximately normal diameter, but are reduced in number. The total islet weight, according to Gepts, is reduced to 56% of normal. The $\alpha:\beta$ cell ratio is increased but this is due to a reduction in the number of β cells; the total α cell weight remains constant. The estimates of total β cell mass given by the two authors are in good agreement; MacLean & Ogilvie finding that the diabetics have a β cell mass of 0.22 g compared with a control figure of 0.64 g, while Gepts' figures are 0.30 and 0.75 g respectively. While these figures represent at least a 60% drop in total β cell mass, there is a considerable overlap with control figures in the individual measurements. Wrenshall *et al.* (1952) found a slight reduction in the amount of extractable insulin in pancreases from adult onset diabetics, again with a considerable overlap with control figures.

The qualitative changes in the islets have been comprehensively reviewed by Warren & LeCompte (1952), and Gepts (1957) has added a careful personal study of fifty-one diabetics. Hyalinization of the islets is the most consistently described abnormality, occurring in 40-45% of diabetics, and 2-4% of controls (Bell, 1952; Warren & LeCompte, 1952; Gepts, 1957). The amorphous eosinophilic hyaline material which accumulates

between the capillary wall and the islet cell gives the staining reaction of amyloid, and has also recently been shown to have its electron microscopic characteristics (Lacy, 1964). Amyloid has also been demonstrated in the stroma of islet cell tumours (Porta, Yerry & Scott, 1962), and because of this, as well as the variability in its incidence, hyalinization is likely to be a secondary phenomenon.

The incidence of hydropic degeneration of the β cell varies from 4% to 53% of diabetics in different series (Gepts, 1957). Similar clear vacuoles have been described in the β cell cytoplasm in experimental diabetes, and these have been shown to be due to glycogen accumulation, and to be reversible with insulin treatment (Toreson, 1951). These changes, like the glycogen accumulation in hepatic cells, are also probably secondary to diabetes, and not causative. Inflammatory infiltration of the islets is very uncommon in adult onset diabetes; the only other common lesion is fibrosis; this will be discussed later.

In juvenile diabetes both Gepts (1965) and MacLean & Ogilvie (1959) find that while the total mass of islet tissue is moderately reduced, the number of large islets is increased. The number of β cells present is very greatly reduced in recently diagnosed juvenile diabetics (Gepts, 1965). The β cells that are present are often severely degranulated (Bell, 1953) and may be large with nuclear abnormalities. In view of these findings, it is not surprising that Wrenshall *et al.* (1952) found a low total pancreatic insulin content. In twenty-one cases of recent onset diabetes, Gepts (1965) found lymphoid infiltration in and around the islets in fifteen. In long-standing diabetes of juvenile onset, lymphocytic infiltration was absent, insular fibrosis more common, and β cells very greatly reduced or completely absent. Hyalinization of the islets is rarely seen in juvenile diabetes (Gepts, 1957). Gepts concludes from the histological findings that an extra-pancreatic factor has exerted a strong stimulatory action on the islet tissue. His observations that the number of β cells continued to decline and that an inflammatory infiltrate was frequently present in and around the islets suggest to us that an active destructive process was also present.

(c) *The islet cell in diabetes secondary to other diseases*

It has been known for many years that total pancreatectomy leads to diabetes (von Mering & Minkowski, 1890) and in animals partial pancreatectomy has been used in the study of experimental diabetes since the early work of Sandmeyer (1892-93). In man subtotal pancreatectomy is a variable and uncommon operation. However, destruction of the pancreas or specifically of the

islets may occur with a variety of diseases, some of which are discussed below.

(i) *Pancreatic carcinoma.* The incidence of abnormal carbohydrate metabolism in patients with carcinoma of the pancreas is known to be higher than in the general population. Bell (1957), in a retrospective study of a large series of cases of carcinoma of the pancreas, found that 14.4% had glycosuria. There was no correlation between the degree of pancreatic destruction and the development of glycosuria, nor was there significant degranulation of the β cells in these patients. He suggested that the tumour cells may interfere with the release of insulin. Green, Baggenstoss & Sprague (1958) found that 15% of patients with pancreatic carcinoma had hyperglycaemia; there was no correlation with the cell type, grade of malignancy or site of the tumour. Thirty-seven per cent of the patients with carcinoma of the pancreas studied by Murphy & Smith (1963) showed an abnormal carbohydrate metabolism. These authors found that the sex ratio of patients with carcinoma of the pancreas and diabetes was similar to that of patients with pancreatic carcinoma, and different from that of patients with idiopathic diabetes mellitus of the same age group. Although the mechanism is not clear, it must be concluded that carcinoma of the pancreas can lead to the development of diabetes mellitus.

(ii) *Acute inflammation.* Well-documented reports of patients with diabetes mellitus which can be proven to be due to an acute infection of the pancreas are rare. One recognized infective agent which may lead to diabetes is the mumps virus. Hinden (1962) reported a case, and in a study of the world literature found only eighteen others. Acute haemorrhagic pancreatitis is an uncommon cause of diabetes, perhaps in part because the disease is associated with a high mortality (Hughes, 1961). However, Mathiesen & Rasmussen (1965) found that 7% of subjects followed up 12 years or more had diabetes mellitus. Warren & LeCompte (1952) considered that acute pancreatitis was a causal factor in less than 1% of their large series of diabetics.

(iii) *Chronic inflammation.* A considerable proportion of patients suffering from chronic pancreatitis develop diabetes, although this condition is only present in a small percentage of any large series of cases of diabetes. Comfort, Gambill & Baggenstoss (1946) found that one quarter of their small but well-studied series of cases of chronic pancreatitis had diabetes. While clinical evidence of chronic pancreatitis—such as pancreatic calculi—is rare in unselected diabetic patients, an increased amount of periductular and interacinar fibrosis is often described in pathological studies, and is considered by some authors to represent a burnt-out

pancreatitis. Lazarus & Volk (1962) found some degree of pancreatic fibrosis in 58% of their series of adult onset diabetics. However, they also found abnormal pancreatic fibrosis in 42% of a control series with cardiovascular or renal disease. Thus while the diabetic patients certainly show slightly more fibrosis than controls, the fibrosis alone cannot be considered the causative lesion in the majority of cases with diabetes. Blumenthal, Probst & Berns (1963), using rather stricter criteria for diagnosing pancreatitis, found an incidence of 11.2% in patients with diabetes, while in the control autopsy series the incidence was 5.3%.

(iv) *Haemochromatosis*. Diabetes is considered by some to be an essential part of this disease; however, in many studies the diagnosis has been made on a clinicopathological basis without diabetes necessarily being present. The incidence of diabetes in these series varies from about 30% to 50% (Kleckner, Baggenstoss & Weir, 1954; Becker & Mills, 1960; MacDonald & Mallory, 1960). Extensive iron deposition is invariably seen in the pancreas; the deposits are usually present in both exocrine and endocrine tissue, and are accompanied by a variable degree of diffuse pancreatic fibrosis (Kleckner *et al.*, 1954; MacDonald & Mallory, 1960). The latter authors failed to find any correlation between the degree of iron deposition in the islets and the development of diabetes. Bell (1955) described degranulation of the β cells in haemochromatosis with diabetes but considered that the degree of pigmentation of the islets was insufficient to interfere with their function. Despite these observations, and the lack of blood glucose abnormalities in experimental haemochromatosis in rats (Macdonald & Pechet, 1965), it is difficult to avoid the conclusion that the iron deposition in the islets is likely to be the factor precipitating diabetes in these patients.

(v) *Vascular disease*. Vascular disease of the pancreas was suggested as a cause of diabetes before the discovery of insulin (O'Hare, 1920), and it remains a possible mechanism for reduction in β cell mass. Moschowitz (1956) studied the islets in seventy cases of essential hypertension, and found abnormalities in eight of them, while finding no lesions in sixty-six cases of peptic ulcer. Lazarus & Volk (1961) found that thickening and hyalinization of the pancreatic arteries was nearly twice as common in diabetics as in a control group with comparable generalized arterial disease. However, direct evidence that vascular disease in man causes reduction in β cell mass is not available.

(2) Insulin structure

The amino acid sequence of human insulin is known (Brown, Sanger & Kitai, 1955; Sanger, 1960)

and recently it has become possible to synthesize insulin (Katsoyannis, 1966). Little direct information is available on the structure of insulin from human diabetics. It has been shown by electron microscope observations that the β cell granules in the pancreas of adult onset diabetes appear normal (Lacy, 1964). Elliott, O'Brien & Roy (1965) found that the insulin extracted from the serum of juvenile diabetics was more resistant to destruction by insulinase than insulin from normal serum. Their method of extraction involved complexing the insulin with guinea-pig anti-insulin serum with later separation. They conclude that the likeliest cause is a genetically determined structural difference between the insulin of patients with juvenile diabetes and controls.

(3) Insulin transfer

The normal mechanism of insulin transport in blood is a matter of considerable debate. Using the conditions described by Prout *et al.* (1963) insulin has the same electrophoretic mobility as α_1 -globulin, and these authors describe an insulin carrying protein.

Antoniades *et al.* (1965), using complex extraction procedures, have separated insulin-like activity from serum into two main components; bound insulin, possibly a complex of insulin and a substance of large molecular weight, and free insulin. In the fasting state, most of the insulin exists in the bound form. They have shown that following an intravenous injection of glucose, free insulin predominates and have suggested that in diabetic subjects conversion of the bound to the free form may be abnormal (Antoniades *et al.*, 1962).

Slater *et al.* (1961), using the isolated rat fat pad, found that a large proportion of insulin-like activity of human serum had identical properties with those of crystalline insulin. However, unlike crystalline insulin it could not be neutralized by insulin anti-serum. This atypical insulin activity was present in normal serum but unlike typical insulin remained constant in concentration during a glucose tolerance test. Samaan, Fraser & Dempster (1963) have shown in dogs that the liver may be responsible for converting typical insulin to atypical insulin.

As the identification and preparation of these factors depend on the use of widely differing techniques, it is not at present possible to correlate the separation of insulin-like activity into 'bound and free insulin' with its separation into 'typical and atypical insulin.'

The carriage of insulin in blood is not the only factor concerned with its transfer from the site of production to the site of action. As Lacy (1964) has pointed out, insulin after leaving the storage granule has to cross the islet cell basement membrane, two capillary basement membranes as well as endothelial

cytoplasm and plasma membranes before it can be effective. Theoretically vascular wall abnormalities could restrict insulin movement. Camerini-Davalos *et al.* (1963) have described minor vascular changes in 'prediabetes,' but no direct evidence for abnormalities in insulin transfer across vessels has been produced.

(4) Insulin destruction

The rate of destruction of insulin is obviously relevant to its biological activity. Insulinase activity is present in more than one form and has been found in variable amounts in almost all tissues except plasma (Mirsky, 1963).

The significance of the particularly high insulinase activity of placental tissue will be discussed later. Thyroxine has been shown to increase the rate of insulin breakdown and the half life of insulin is prolonged in hypothyroid rats (Elgee & Williams, 1955).

The level of insulinase activity is probably not a significant factor in idiopathic diabetes, as Welsh *et al.* (1956) have shown that the half life of labelled insulin is the same in untreated diabetes as in controls.

(5) and (6) Insulin antagonism and endorgan responsiveness

Very similar effects may be attained either from a circulating antagonist or by a tissue change which reduces its responsiveness to insulin—hence these two disorders will be considered together.

(a) Growth hormone

The physiological effects of growth hormone have been reviewed recently by Hartog (1964). The effect of excess growth hormone on glucose metabolism may be due to its peripheral action as an antagonist to insulin. In a study of forearm metabolism in six acromegalic patients Zierler & Rabinowitz (1963) found a normal basal glucose uptake but an impaired insulin response. Butterfield, Garratt & Whichelow (1963) also studied forearm metabolism in acromegalics and, using a different technique, found that insulin fixation and glucose uptake were within the control range. The results of animal work offer more conclusive evidence in support of the peripheral action of growth hormone. Milman, DeMoor & Lukens (1951) found that the insulin requirement of depancreatized cats increased considerably when they were treated with growth hormone. Despite conflicting earlier reports, more recent studies of a small number of cases of acromegaly have shown no significant changes in the islets of Langerhans (Lazarus & Volk, 1962).

The diabetogenic effect of growth hormone led Young (1950) to suggest that some cases of idio-

pathic diabetes might be due to prolonged slight overproduction of growth hormone, but recent work (Yalow *et al.*, 1965) has not shown any increase in growth hormone levels in diabetic sera. A more detailed study (Hunter, Clarke & Duncan, 1966) measuring growth hormone in untreated diabetics supports previous evidence that growth hormone does not play a significant role in the pathogenesis of diabetes. However, patients with acromegaly may develop diabetes although the proportion who do varies widely from series to series. Lazarus & Volk (1962) reviewed thirteen papers in which the incidence of diabetes in acromegalics ranged between 7.5% and 80.7%, the mean being 25%. This is almost exactly the proportion found in a group of seventy-five acromegalics investigated at the Hammersmith Hospital (Joplin & Wright, 1966).

(b) Corticosteroids

The incidence of diabetes in Cushing's syndrome is similar to its incidence in acromegaly. Ross, Marshall-Jones & Freedman (1966) found frank diabetes in seven out of fifty cases of Cushing's syndrome. In Cushing's syndrome and in steroid treated patients, acute pancreatitis and peripancreatic fat necrosis may be present (Carone & Liebow, 1957). Lukens, Flippen & Thigpen (1937) considered that the islets were qualitatively normal in patients with Cushing's syndrome and glycosuria, but we know of no quantitative studies. Lazarus & Volk (1962) reviewed the pancreatic findings in Cushing's syndrome, and concluded that no consistent islet changes had been described. The peripheral diabetogenic action of steroids has been described by many authors, including Lazarus & Volk (1962) who showed that steroids slowed the rate of fall of blood sugar following insulin injection.

(c) Adrenaline

The effect of adrenaline on blood sugar level is complex, as it leads to both increased glycogenolysis and decreased peripheral glucose uptake. Recent work has shown that adrenaline also inhibits insulin release both *in vitro* (Coore & Randle, 1964) and *in vivo* (Porte *et al.*, 1966). In view of the experimental observations it is not surprising that hyperglycaemia is frequently recorded in patients with pheochromocytoma. Hume (1960) in a comprehensive review of the literature quoted several series in which glucose metabolism had been investigated. From these reports it seems that the fasting blood sugar is elevated in approximately 50% of patients with pheochromocytoma. Graham (1951) in his review found frank diabetes in twenty-one of eighty-eight cases.

(d) Glucagon

For some years it has been suggested that glucagon might play an important part in the development of

diabetes, and this has been correlated with an increased $\alpha : \beta$ cell ratio in the islets (Ferner, 1952). However, the conclusions drawn from this and earlier work are invalid, as Gepts (1957) has shown that the increase in α cells is relative and not absolute. Also Hellman and co-workers in a series of articles have clearly separated two types of α cells. Studies on the distribution of these two types and correlation with glucagon content suggests that the non-argyrophilic α cell is responsible for glucagon production (Hellman, Wallgren & Hellerstrom, 1962). As many of the earlier workers used silver techniques in their study of α cells in diabetes, no conclusions can be drawn about glucagon from their work. It appears unlikely that glucagon plays a major role in the pathogenesis of idiopathic diabetes mellitus. It may be of importance in rare cases; and a single example of a glucagon secreting α cell carcinoma of the pancreas associated with mild diabetes has recently been reported (McGavran *et al.*, 1966).

(e) *Synalbumin*

Synalbumin is the name given by Vallance-Owen to a substance which he found antagonized the effect of insulin on the isolated rat diaphragm preparation (Vallance-Owen, Dennes & Campbell, 1958). Synalbumin is prepared from serum using an ethanol-trichloroacetic acid extraction procedure, and it is found in both diabetic and control sera, although it is consistently present at a high concentration in diabetes. While Keen (1963) was unable to confirm these findings, the essentials of Vallance-Owen's work have been substantiated by several groups (Lowy, Blanshard & Phear, 1961; Alp & Recant, 1965). A high level of synalbumin activity is found in some relatives of diabetics, and it has been suggested that this high level may be inherited as a dominant character (Vallance-Owen, 1964). While it is surprising that if synalbumin is the antagonist responsible for diabetes, its activity cannot be demonstrated in whole diabetic sera, the observation is too important to be dismissed for this reason alone. Vallance-Owen and co-workers (Ensink, Mahler & Vallance-Owen, 1965) have shown that the B chain of insulin has an effect similar to synalbumin, and suggest that abnormal degradation of insulin leading to excessive levels of B chain may be the basic abnormality in diabetes.

(f) *Free fatty acids*

Free fatty acids (FFA), the form in which fats are released from the depots, have been shown to act as antagonists to insulin *in vitro* (Randle *et al.*, 1963). Insulin is known to inhibit FFA release and hence lower circulating FFA *in vivo* (Bierman, Schwartz & Dole, 1957; Rabinowitz & Zierler, 1965). Randle and his group (Hales & Randle, 1963) have also

shown that in both diabetics and control subjects on a low carbohydrate diet, serum FFA levels are raised despite high serum insulin values. They have suggested that an excessive production of FFA with a consequent increased insulin resistance may be the primary factor in the development of diabetes in many patients.

(g) *Other antagonists*

Field, Stetten & Woodson (1956) found insulin antagonism in the sera of patients with diabetic ketosis. The antagonism was transient, and was probably related to the metabolic abnormalities associated with ketosis. In support of this work, *in vitro* experiments (Ottaway, 1961) have shown that ketone bodies decrease glucose uptake of the isolated rat diaphragm. Bornstein (1953) described pituitary dependent activity in the serum lipoprotein fraction of alloxan diabetic rats which depressed the glucose uptake of the isolated rat diaphragm. No clear evidence of the importance of these observations in human diabetes has been produced. Insulin antagonism due to antibody formation following treatment with exogenous insulin is well known. Berson & Yalow (1964), in a study of a large series of untreated diabetics, failed to find insulin antibodies, so this is probably not an important mechanism in the pathogenesis of diabetes.

(h) *Obesity*

The incidence of diabetes in obese subjects is much greater than in the general population. Obesity with or without diabetes appears to be associated with a decreased endorgan responsiveness to insulin. The evidence for this is derived from the common finding that obese diabetics are usually relatively resistant to insulin, and from the observation that the augmented insulin tolerance test is abnormal in obese non-diabetic patients (Fraser *et al.*, 1962). Also it has been found in studies of forearm metabolism of obese non-diabetic patients that the glucose uptake after insulin infusion was lower than that of controls (Rabinowitz & Zierler, 1962).

Serum levels of insulin in obese patients, both diabetic and non-diabetic, are known to be above normal. Yalow *et al.* (1965) investigated serum insulin levels during a glucose tolerance test in diabetic and control patients, with a wide range of body weight. They found that raised insulin levels occurred particularly in the diabetic patients. Karem, Grodsky & Forsham (1965b) in a similar investigation found that raised serum levels correlated better with the degree of obesity than with diabetes. A wide spectrum of insulin levels may be seen in both obese diabetic and obese non-diabetic patients. There is no logical reason why patients with obesity should be regarded as a homogenous

group, and anomalous results are to be expected until the pathogenesis of both diabetes and obesity is more fully understood.

(i) *Pregnancy*

Circulating insulin is slightly raised in the normal pregnancy (Bleicher, O'Sullivan & Freinkel, 1964) and there is a diminished response to injected insulin in the third trimester (Burt, 1956). While several other hormones show an increased serum binding in pregnancy, there is no direct evidence that this is true for insulin. It has been suggested that these findings are related to a change in endorgan responsiveness, but both these observations and the well-known increase in insulin requirements in the pregnant diabetic patient could be due to more rapid insulin destruction. Direct evidence that this may occur has been provided by Freinkel & Goodner (1962) who have shown that the human placenta is very rich in insulinase, and that the half life of ^{131}I insulin in the pregnant rat is greatly reduced.

Diabetes is relatively first diagnosed during pregnancy; this is partly due to routine urine testing of otherwise healthy subjects and partly to a real increase in its incidence (O'Sullivan, 1961). O'Sullivan also showed that in a considerable proportion of pregnant women, the abnormal glucose tolerance tests reverted to normal after delivery. However, on retesting some of these more than 5 years later, about two-thirds were found to be diabetic. While it has been claimed that multiparous women have a higher incidence of diabetes than nulliparae, the survey of the population of Bedford did not show any increase in diabetes among women with three or more children as compared to those with fewer children (Keen, 1964). This observation, however, was based on a small sample and previously diagnosed diabetics were excluded. Both Fitzgerald *et al.* (1961) and Walker (1964) claim that multiparity increases the risk of developing diabetes, but the interpretation of this observation is made difficult by the correlation between parity, obesity and increasing age (O'Sullivan, Gellis & Tenney, 1966).

(7) β cell control

The major factor in controlling the release of insulin from the pancreas is generally considered to be the blood glucose level. This is supported by work in which the amount of insulin released from the isolated pancreas was shown to depend on the glucose concentration of the incubating medium (Grodsky *et al.*, 1963). More recently, two groups of workers (McIntyre, Holdsworth & Turner, 1964; Elrick *et al.*, 1964) have shown that oral glucose leads to a higher serum insulin level than an equivalent amount of intravenous glucose, reviving the suggestion that an intestinal factor might be in-

involved in insulin release. Other factors known to lead to increased insulin secretion include sugars other than glucose (Karam *et al.*, 1965a), glucagon, and certain amino acids, notably leucine. The response of the insulin releasing mechanism can be varied by certain factors. For instance, growth hormone increases the amount of insulin released in response to a glucose load (Cerasi & Luft, 1964); and chlorpropamide potentiates leucine induced insulin release (Fajans *et al.*, 1963). Chlorpropamide and tolbutamide are two of the sulphonylurea group of drugs, the hypoglycaemic effect of which was first shown to depend on an intact pancreas by Loubatières (1944). They are known to stimulate insulin secretion whatever the blood sugar level, providing the islets are not already under maximal stimulation.

Direct evidence for an abnormality in feedback control of the pancreatic islets in diabetes mellitus is hindered by the lack of insulin secretion rate studies. However, an abnormality in the mechanism controlling islet function can be inferred in the majority of maturity onset diabetics, as treatment with tolbutamide demonstrates that their islets can still be stimulated to produce more insulin.

The control of β cell growth is more difficult to study than the control of function. In endocrine organs like the thyroid and the adrenal, a stimulus to increased function is accompanied by a stimulus to growth, and after partial ablation, regeneration occurs. Martin *et al.* (1963) commented on earlier work in which diabetes was produced by partial pancreatectomy. They pointed out the importance of species variation, and found that in the rat hyperglycaemia stimulated islet regeneration. By contrast, in dogs and cats, hyperglycaemia led to an increased incidence of diabetes after partial pancreatectomy. From this work, and from the evidence reviewed by Lazarus & Volk (1962), it seems that regeneration of the islets may occur in adult rat and rabbit, but probably does not occur in cats and dogs. Work on regeneration of the human islet is limited for obvious reasons, but in the adult the capacity for regeneration seems to be limited.

The presence of an islet growth promoting factor in the serum of diabetics can be inferred from post-mortem studies, demonstrating large islets in children born to diabetic mothers. Careful studies have shown that the total islet mass in these children is on average more than twice that of controls (D'Agostino & Bahn, 1963). Several authors (Helwig, 1940; D'Agostino & Bahn, 1963; Silverman, 1963) have noted that infants born to diabetic mothers often show an infiltration by eosinophil leucocytes in and around the pancreatic islets. Large islets are also found in infants of prediabetic mothers and in infants with rhesus incompatibility (Woelf & Jackson, 1957). The mechanism for these

islet changes remains unexplained, although it may be glucose itself that is the factor controlling islet growth. In support of this is the observation that a high carbohydrate diet leads to increase in islet size in rats (Tejning, 1947; Kuleshova, 1961).

Discussion

From this brief review, it can be seen that abnormalities have been found in each of the stages in insulin-carbohydrate interaction enumerated in the introduction, and many of these have been suggested as fundamental in the pathogenesis of diabetes mellitus. Clearly no step can be considered in isolation, and in discussing the significance of pancreatic change in diabetes mellitus, abnormalities in other stages, and particularly in the control of β cell mass and function must be considered. If the β cell mass is reduced or if insulin is destroyed by excess insulinase, the feedback mechanism should restore normality. However, the ability of the feedback mechanism to cope with an abnormal situation is limited by the maximum secretory capacity of each cell, and by the potential growth of the β cell mass.

In human juvenile diabetes, the fact that the β cell mass is reduced at onset and continues to fall implies islet destruction. However, as the proportion of large islets is much greater than that found in controls, it seems likely that some regeneration has been occurring. If we review briefly the various stages in the insulin carbohydrate cycle, the evidence for an abnormality in any other stage in juvenile diabetes is slender. The isolated observation suggesting an abnormal insulin structure in juvenile patients remains unconfirmed.

The clinical observation of relative insulin sensitivity in most juvenile diabetics suggests that there is no major abnormality in insulin transfer, antagonism, or in endorgan sensitivity. The finding of degranulated β cells and low insulin content suggests that the islets are responding to hyperglycaemia, and that the feed-back mechanism is functioning normally. The overall picture in juvenile diabetes is therefore one of maximal stimulation of a greatly reduced β cell mass, with inadequate total insulin production. Several mechanisms may be responsible for the loss of β cells in this condition. Gepts (1965) found lymphocytic infiltration in and around the islets in fifteen of twenty-one cases of juvenile diabetes of recent onset. The description of similar islet changes, in some instances with diabetes, in animals injected with homologous or heterologous insulin and Freund's adjuvant (Toreson *et al.*, 1964; Grodsky *et al.*, 1966; LeCompte *et al.*, 1966) support the possibility that the islet changes in juvenile diabetes may be due to an autoimmune disease. While circulating anti-islet antibodies have not as yet been demonstrated in juvenile diabetes,

indirect supporting evidence is available. Landing *et al.* (1963) found antithyroid antibodies in 13% of a large series of juvenile diabetics, a significant figure in this age group despite the absence of sex matched controls, and Moore & Neilson (1963) found abnormal complement fixation tests for both thyroid and gastric mucosa more commonly in a group of juvenile diabetics than in either adult onset diabetics or controls. While autoimmune destruction of the islet is a likely cause for some cases of diabetes, particularly those of juvenile onset, it is not proven, and other possible causes for the loss of β cells and islet inflammation in juvenile diabetes include viral infections and toxic agents.

The relationship between the pancreas and adult onset diabetes is perhaps more complicated, and certainly more confused. Many studies on adult onset diabetes, including those of islet histology and extractable insulin (Wrenshall *et al.*, 1952), unfortunately have not differentiated between obese and non-obese subjects, or between those who could be treated by diet alone, sulphonylurea or insulin.

In contrast to juvenile diabetes, the β cell mass in diabetes of adult onset is not greatly reduced. Insulin structure is normal as far as is known. Although most adult onset diabetics do not require insulin, the fact that they are relatively insulin insensitive suggests that there is an abnormality in insulin transfer, antagonism, breakdown or endorgan responsiveness. It is difficult to establish a single aetiology for idiopathic diabetes mellitus of adult onset as abnormalities are claimed for each of these stages.

The high level of circulating insulin in some adult onset diabetics does not suggest an increased insulin breakdown and no direct evidence that exogenous insulin is transported in an abnormal way has been produced. Serum from untreated non-ketotic diabetics has not been shown to interfere with the action of added insulin, so it seems likely that in many obese diabetics some defect of endorgan responsiveness is present. This may well reflect the obesity present in many adult onset diabetics, rather than the diabetic state.

There is evidence for a second abnormality in insulin carbohydrate interaction in adult onset diabetics. The presence of a relatively normal β cell mass and pancreatic insulin content despite hyperglycaemia suggests that the feedback control of insulin release is not functioning normally, particularly as this situation can be rectified by the use of sulphonylureas. The situation in many cases of diabetes of adult onset can, therefore, be summarized as one in which a relatively normal pancreatic islet cell mass is responding inadequately to the stimulus of hyperglycaemia, the hyperglycaemia being caused in part by endorgan un-

responsiveness to insulin. We do not consider that the morphological abnormalities described in the islets in adult onset diabetes are likely to be causative; the evidence suggests that they are secondary.

The factors that may lead directly to the development of adult onset diabetes are many, and because they include conditions in which an increased load is put on the pancreatic islets, the concept of exhaustion of the β cells has been used. In other peripheral endocrine organs an increased work load is accompanied by an increase in gland size, and a greater functional capacity. In the adult human pancreas the size increase does not occur, and it may well be that an increased work load on the islet cell shortens its life, and, in the absence of replacements, diminishes the islet mass. However, in the early stages of diabetes of adult onset, the islets are certainly not exhausted, as they contain adequate insulin and are capable of responding to the stimulus of tolbutamide. In our view the commonest precipitating factor leading to adult onset diabetes mellitus is the development of insulin resistance associated with obesity.

In this discussion we have not yet commented on the hereditary factor. Both diabetes of juvenile and adult onset type may occur in the same family and yet the abnormalities in insulin-carbohydrate interaction are different in the two types. The most likely explanation is that there is an inherited liability to develop diabetes mellitus which is distinct from the factors which precipitate its onset and determine its type. Vallance-Owen & Ashton (1963) have found that 50% of relatives of diabetics are synalbumin positive, whereas 20% of controls are synalbumin positive. They find that all patients with idiopathic diabetes are synalbumin positive, whether of juvenile or adult onset type. The degree of synalbumin antagonism may well be an expression of the inherited liability to develop diabetes mellitus. Vallance-Owen's hypothesis that the synalbumin factor is the β chain of insulin offers interesting possibilities. An excess of β chain could imply an abnormality in manufacture or in breakdown of insulin, and could interfere with peripheral metabolism or with feedback.

Diabetes may occur secondarily to a variety of diseases which fall into two main groups, those affecting the pancreas directly, as in pancreatitis, haemochromatosis and pancreatic carcinoma and those affecting endorgan responsiveness — for example Cushing's syndrome and acromegaly. There is no clear-cut correlation between the severity of these diseases and the development of diabetes, and the presence or absence of the inherited 'diabetic factor' is likely to be an important variable. Coggeshall & Root (1940) found a higher incidence of a family history of diabetes in acromegalics who themselves develop diabetes than in acromegalics

without diabetes.

In conclusion we would like to stress once again that diabetes mellitus is a clinical syndrome not a pathogenetic entity. We prefer to use the term in the same sense as myxoedema, as a descriptive diagnosis, not implying a single pathogenesis. A clear parallel can be drawn between the various abnormalities which may lead to diabetes, and those leading to other endocrine disorders. Diabetes and Addison's disease may both be due to surgical resection, or to destruction of the gland by infection, or an autoimmune process. Indeed, an autoimmune process may affect several endocrines simultaneously and diabetes occurs with surprising frequency among patients with autoimmune myxoedema and Addison's disease (Carpenter *et al.*, 1964). The possibility that diabetes may be due to an abnormality in the production of insulin has its counterpart in myxoedema due to dyshormonogenesis, while endorgan unresponsiveness has been described for parathormone as well as insulin. The two features which distinguish pancreatic β cell hypofunction from thyroid, parathyroid and adrenal hypofunction are the limited capacity for growth of the adult human β cells, and the presence of the inherited factor as a substrate with which any of the numerous other factors leading to diabetes may interact.

Integrated clinical, biochemical and pathological investigations offer the best hope of elucidating the two factors concerned in most cases of idiopathic diabetes mellitus, the inherited factor that predisposes towards diabetes, and the precipitating factor which leads to its final appearance.

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