GLYCOGEN STORAGE IN THE LIVER IN DIABETES MELLITUS

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In this clinic, where 800 patients are seen, we have noted what appears to be a new syndrome. It occurs in young, severe, "brittle" diabetics, and is characterized by two predominant features—an enlarged liver found to be full of glycogen and a constant tendency to hypoglycaemia while taking insulin. The enlarged liver, not unusual in diabetes mellitus and commonly supposed to be due to deposition of fat, was in each case found by repeated biopsy to be distended with masses of glycogen, as in von Gierke's disease.

A survey of the literature indicates that the general consensus of opinion regarding the amount of glycogen in the liver in diabetes mellitus is that in this condition the amount is decreased (Mirsky, 1942; Korenberg, 1943), and observations on experimentally produced diabetes in animals by many authors (Cruickshank, 1913, to Bodo, Co Tui, and Farber, 1933) support these findings. Pemberton (1925) records the low figure of 0.04% glycogen in the liver in a severe diabetic dying of insulin hypoglycaemia. Vallance-Owen (1952), on the other hand, describes a young diabetic dying in severe ketosis and coma, whose liver at necropsy was distended with glycogen.

The use of "glucagon," the hyperglycaemic glycogenolytic factor said to be liberated in the alpha cells in the pancreas and the gastric mucosa (Sutherland and de Duve, 1948), has led to some interesting speculations. For instance, Kirtley, Waife, and Peck (1953a) and Kirtley, Waife, Helmer, and Peck (1953b) suggest that the smallness of the rise in the blood sugar (after the exhibition of "glucagon") may be due to lack of liver glycogen in a patient with little or no endogenous insulin, i.e., the young, severe diabetic. Our own results in diabetic patients whose livers are full of glycogen are not in keeping with these observations.

We have found no evidence in the literature of a syndrome such as we describe, whereby severe diabetic patients exhibit hepatomegaly due to glycogen storage and yet are prone to hypoglycaemic

attacks, presumably due to the fact that glycogen is not available to them for gluconeogenesis. In fact in a series of 50 cases of diabetes mellitus with homologous serum jaundice, liver biopsy was done during the acute phase and one or two years afterwards in order to determine any abnormal liver histology, and no comparable amounts of glycogen were found in either series. The distribution of glycogen in these livers and in our present series was identified and visually assessed by using the periodic-acid-Schiff reaction (P.A.S.) before and after digestion with diastase (Pearse, 1953).

In this respect our cases resemble von Gierke's disease, the important difference being the fact that patients in this series are proven severe or brittle diabetics and do not fulfil the criteria for the recognition of von Gierke's disease, as laid down by Ellis and Payne (1936). An attempt has been made to mobilize the excess liver glycogen by administering glucagon to our cases. Hubble (1954) has already demonstrated that this substance is unable to mobilize glycogen from the liver in von Gierke's disease, and our own analyses, which have followed closely the technique previously published by Kirtley *et al.* (1953a and b), suggest that for some reason, at present unknown, glycogenolysis is impaired in these patients—a feature already suggested by their tendency to hypoglycaemia.

Case Records

Case 1.—A. W. was a female nursery assistant, aged 25, who had had diabetes since the age of 13 (1941). Stabilization had always been difficult owing to recurrent attacks of hypoglycaemia; she was finally admitted to this hospital in August, 1953, complaining of recurrent swelling of the parotid glands, repeated attacks of hypoglycaemia, and amenorrhoea. While in hospital she noticed increasing abdominal distension and a feeling of epigastric discomfort; examination of the abdomen revealed a smooth, soft enlargement of the liver, three fingerbreadths below the right costal margin. Biopsy showed the hepatic cells everywhere to be distended with glycogen. Pain, swelling, and tenderness

over both parotid glands were at first thought to be mumps (there had been a recent history of contact in the nursery). However, these glands have fluctuated in size up to the present time and there appears to be some relationship to the hypoglycaemic attacks.

The diabetes may be regarded as brittle in type because with a morning fasting blood sugar of 500 mg.% the patient tended to develop typical hypoglycaemia (blood sugar less than 50 mg.%) later the same afternoon. The hypoglycaemia was always relieved by the administration of oral or intravenous glucose. For 10 months she had had amenorrhoea, but pelvic examination and curettage revealed no abnormality and in fact her menstrual cycle became normal later. Radiographs of the gastrointestinal tract, chest, skull, and parotid glands were normal.

The hypoglycaemia became gradually more severe and abdominal pain and distension increased, and after surgical consultation it was decided to explore the abdomen on April 23, 1954, in the remote chance of finding a gross pancreatic lesion, although in a recent case of insulin adenoma seen by us the liver glycogen has been found to be microscopically normal. A smooth enlargement of the liver was confirmed, but nothing macroscopically abnormal was found in the pancreas although, as is shown below, the islets were small and scanty.

Case 2.—R. T. was a schoolboy aged 14, who had had diabetes since the age of 8 (1948). Stabilization presented the same difficulties as in Case 1 and he was finally admitted to this hospital in July, 1953, because of the sudden onset of abdominal distension, which was found to be due to enlargement of the liver, and in addition attacks of hypoglycaemia were now frequent. He also had well-marked insulin atrophy.

Biopsy showed the liver cells to be distended with glycogen. The abdominal distension persisted, some pyrexia and vomiting developed, and the diagnosis of a tuberculous peritonitis was entertained, especially as the Mantoux test was positive 1 in 10,000. Streptomycin and P.A.S. were given for a time but discontinued when the meteorism and pyrexia had subsided. A repeat biopsy showed that the liver remained full of glycogen and that it was not altered by the administration of thyroid extract, although the blood sugar remained at a higher level while on this glycogenolytic drug.

Case 3.—J. C. was a schoolgirl aged 13, who had had diabetes since the age of 11 (1951). From the first she had been remarkably unstable and required large doses of insulin. She was admitted to this hospital in November, 1953, with tonsillitis and her mother related recent attacks of hypoglycaemia as a new feature. On examination follicular tonsillitis, a distended abdomen, and an enlarged liver were found. Biopsy again showed a normal liver pattern with hepatic cells swollen with glycogen.

Stabilization was attempted with "lente" and "ultra lente" insulins, but repeated attacks of hypoglycaemia necessitated the use of another glycogenolytic agent—adrenaline—with a rise in blood sugar and partial control. Further liver biopsy after adrenaline revealed that the liver cells were still distended with glycogen.

Case 4.—K. C. was a schoolgirl aged 13, who had had diabetes since the age of 3½ (1942). She was admitted to hospital in January, 1954, because of hypoglycaemic attacks, distension of the abdomen, and enlargement of the liver. Liver biopsy showed architecture of the normal pattern, but distension of the cells with glycogen. Recurrent abdominal distension and epigastric pain were again prominent features as in the other cases. The use of adrenaline brought about some partial control of the hypoglycaemia as in the previous case, but liver biopsy once again revealed that the appearance of the glycogen-distended liver cells had not altered. An E.C.G. showed no abnormality.

Table I summarizes the histological and biochemical investigations which were performed on these patients. From the histological point of view it can be seen that, although the livers are full of glycogen, their architecture is normal. From the

TABLE I
HISTOLOGICAL AND BIOCHEMICAL FINDINGS

Investigations	Case 1 A. W.	Case 2 R. T.	Case 3 J. C.	Case 4 K. C.
No. of biopsies ..	2	4	2	3
Glycogen in liver ..	Excess	Excess	Excess	Excess
Liver pattern ..	Normal	Normal	Normal	Normal
Glycogen in muscle ..	—	—	—	—
Pancreas ..	Small and scanty islets	—	—	—
Gastric analysis ..	Hyperchlorhydria	—	Normal	Normal
Serum cholesterol (in mg.%)	250	—	220	300
17-Ketosteroids (mg.)	17	—	—	—
Glucose tolerance test (in mg.%)	490, 650, 500, 550, 650	100, 194, 352, 420, 364	510, 520, 540, 600, 540	444, 744, 696, 558, 508
Lactose tolerance test (in mg.%)	390, 420, 510, 590, 580	—	640, 660, 720, 740, 700	400, 570, 600, 620, 600
Insulin tolerance test	Insensitive	—	Normal	Insensitive
Glucagon sensitivity (intravenous drip, mg.%)	425, 460, 475, 510	420, 440, 420, 420	—	450, 420, 480, 485
Serum inorganic phosphate with insulin (in mg.%) ..	Fell 4-3.6	Fell 2.3-2.38-2.43-1.48	2.3	Fell 5.3-5.1

fall in serum inorganic phosphate levels it is to be inferred that insulin activity is normal. The serum cholesterol levels and the lactose tolerance test were done to exclude von Gierke's disease. The insulin tolerance test was done to exclude insulin adenoma for which, in fact, a very careful visual search was made in Case 1.

Discussion

When we first recognized the existence of this syndrome we were greatly puzzled by the apparently conflicting nature of its component parts. The

four patients described in this paper were obviously unique when compared with the other diabetics, numbering about 800, at present attending the Clinic. Such features as hepatomegaly due to glycogen storage, associated with severe hypoglycaemic tendencies, in patients who were also severe or brittle diabetics constituted a problem for which we have as yet found no entirely satisfactory solution. Enlargement of the liver constitutes one of the most significant features of the syndrome. We are, of course, familiar with enlargement of the liver in diabetic patients associated with deposition of fat or haemochromatosis, but the discovery of glycogen as the cause of hepatomegaly in these patients is very difficult to explain. We suggest that there are two possible explanations.

(1) All our cases were exceptionally severe or brittle diabetics in whom stabilization had been virtually impossible. For years their blood sugar had been elevated well above the normal levels, interrupted at frequent intervals by repeated attacks of hypoglycaemia. Bouckaert and de Duve (1947) have stated that the normal regulator of sugar metabolism is the blood sugar itself and that upon it the effect of insulin depends. Apparently glycogen storage in the liver will occur in the depancreatized animal as a result of hyperglycaemia (Bodo *et al.*, 1933) and thus it would appear that insulin acts as a catalyst in carbohydrate metabolism, allowing the tissues to achieve at normal blood sugar levels that for which high levels are required in its absence (Vallance-Owen, 1952). By virtue of the severity of the diabetes in the patients whom we have described, the accumulation of glycogen may have resulted from prolonged periods of hyperglycaemia.

(2) The only other condition in which glycogen is stored in the liver in comparable amounts to that in this syndrome is von Gierke's disease (glycogen storage disease). This is a congenital disturbance possibly inherited as a Mendelian recessive character, in which there appears to be a defect of the glycogen-splitting enzyme, which produces an abnormal stability of glycogen. According to Crawford (1946), in von Gierke's disease the normal reaction glucose $\rightarrow$ glycogen $\rightarrow$ glucose is replaced by the sequence glucose $\rightarrow$ fat $\rightarrow$ ketone bodies and glucose, presumably due to the lack of the normal enzyme reaction. We wish to make it quite clear that our cases are not examples of von Gierke's disease. We have utilized the criteria laid down for the diagnosis of this disease by Ellis and Payne (1936) and have demonstrated that our patients do not satisfy these requirements (Table I). Briefly, our patients do not have a low fasting blood sugar level, they are

confirmed severe diabetics, and their hypoglycaemic tendencies are partially controlled by the administration of thyroid extract or adrenaline. Further, the blood cholesterol levels are not significantly raised. Nevertheless, hypoglycaemic tendencies and an enlarged liver are features closely resembling two major components of von Gierke's glycogen storage syndrome. In this connexion we may find the explanation for the recurrent attacks of hypoglycaemia which all our patients have shown. Because they are all severe diabetics, they require daily insulin to stave off diabetic pre-coma. If we postulate that they lack the glycogen-splitting enzyme and that their liver glycogen is abnormally stable, as in von Gierke's disease, the recurrent hypoglycaemic episodes from which all of them suffer may be due to the virtually unopposed action of the daily insulin injections. Some support for this theory is found in Case 1 (A.W.), who became hypoglycaemic at regular intervals twice daily, and on each occasion it could be inferred that insulin activity was maximal at the time. Conversely her morning fasting blood sugar was invariably in the region of 500 mg. %.

To establish this syndrome as a separate entity, we have pointed out that, although it bears a certain resemblance to von Gierke's disease, our patients do not satisfy the diagnostic criteria already indicated. It is important also to stress that the syndrome differs greatly from the more easily defined picture which we refer to as diabetes mellitus. The ordinary diabetic patient, however severely affected, can be stabilized by appropriate diet and insulin therapy. Diabetic neuropathy, renal lesions, retinopathy, are all recognized associated features of the diabetic syndrome, and liver enlargement, when it occurs, is always due to fatty change or occasionally haemochromatosis. In the present series of patients all attempts at stabilization failed despite frequent variations of diet and the use of all types of insulin therapy. Further, the association of acute attacks of abdominal pain, meteorism, and liver enlargement (due to glycogen storage), together with recurrent attacks of hypoglycaemia which cannot be controlled by modification of insulin therapy, are never found in the ordinary diabetic patient. The response of our patients to "glucagon" (H.G.F.) has greatly interested us. From Table I it may be seen that the rise in blood sugar following an intravenous drip containing this substance was surprisingly small, when it is recalled that the liver cells of all these patients are distended with masses of glycogen. This apparent inability of "glucagon" to mobilize glycogen from the liver in any significant amount is similar to results published

by Hubble (1954) in von Gierke's disease. Alternatively, Kirtley *et al.* (1953a and b) suggest that in the ordinary severe diabetic the smallness in the rise of blood sugar following "glucagon" is due to lack of liver glycogen in these patients. We can only assume that in our series of cases glycogenolysis is impaired, a feature already suggested by their tendency to hypoglycaemia.

In conclusion we would refer to Case 1 (A. W.) in which, after prolonged discussion, laparotomy was considered justified. Although biochemical investigation did not suggest the presence of an insulin adenoma, we were of the opinion that in such a bizarre syndrome as this only direct inspection of the pancreas would satisfy us that no gross pancreatic lesion existed. As stated above, the pancreas appeared normal on inspection, but histologically we observed that the islets were remarkably small and extremely scanty. These changes could be secondary to prolonged hyperglycaemia (Vallance-Owen, 1952), as the patient had never achieved stabilization during a diabetic life of 12 years.

We hope that in this paper we have justified the introduction of a new syndrome, which should be sought in any young, brittle diabetic patient with enlargement of the liver.

Summary

In a diabetic clinic of 800 patients a new syndrome has been described in four cases having a brittle type of diabetes mellitus, associated with enlargement of the liver due to glycogen storage and recurrent attacks of hypoglycaemia.

The cases are described from both clinical and biochemical aspects.

The nature of these findings is discussed with particular reference to von Gierke's disease, the use of "glucagon" (H.G.F.), and the more common syndrome of diabetes mellitus.

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