oxytocin induction and the knowledge that decreased deformability leads to accelerated haemolysis[9-12] indicate the pathway of haemolysis in the induced group, and only the cause of the reduced deformability remains to be proved.

That it was oxytocin crossing the placenta that affected deformability was shown in the in-vitro studies, with the finding of both a time-related and dose-related effect of oxytocin on erythrocyte deformability in the range of oxytocin concentrations found in maternal blood during induction of labour with oxytocin.[16] These in-vitro results also help to explain the clinical observations that the hyperbilirubinaemia after induction of labour is related to the dose and the duration of oxytocin administration.[3 4] The lowered plasma osmolality after induced labour agreed with the findings of Singhi and Singh[8] and suggests that the vasopressin-like action of oxytocin[17] causes activation of electrolyte and water transport across the erythrocyte membrane with consequent osmotic swelling, which is a well-recognised cause of reduced erythrocyte deformability[9] and leads to more rapid erythrocyte destruction. In the neonate, whose hepatic enzymes are unable to cope with the increased bilirubin production, clinical hyperbilirubinaemia ensues.

Oxytocin is an important therapeutic agent in obstetrics and probably its effect on erythrocytes cannot be prevented other than by keeping the total dose used to a minimum. The use of prenatal drug treatment with either phenobarbitone[18] or antipyrene[19] to activate fetal hepatic glucuronyltransferase and so increase the neonate's ability to eliminate bilirubin has been suggested, but it would be more logical to prevent the hyperbilirubinaemia by reducing the dose of oxytocin rather than treat it with potentially toxic drugs.

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Role of residual insulin secretion in protecting against ketoacidosis in insulin-dependent diabetes

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Summary and conclusions

The role of preserved beta-cell function in preventing ketoacidosis in type I insulin-dependent diabetes was assessed in eight patients with and seven patients without residual beta-cell function as determined from C-peptide concentrations. After 12 hours of insulin deprivation blood glucose, ketone-body, non-esterified fatty-acid, and glycerol concentrations were all significantly higher in patients without beta-cell function than in those with residual secretion. Mean blood glucose concentrations reached 17·2±SE of mean 1·3 mmol/l (310±23 mg/100 ml) in the first group compared with 8·8±1·4 mmol/l (159±25 mg/100 ml) in the second ($P<0·01$), while 3-hydroxybutyrate concentrations rose to 5·5±0·5 mmol/l (57±5 mg/100 ml) and 1·4±0·3 mmol/l (15±3 mg/100 ml) in the two groups respectively ($P<0·01$). Individual mean C-peptide concentrations showed a significant inverse correlation with the final blood glucose values ($r=-0·91$; $P<0·02$).

These findings strongly suggest that even minimal residual insulin secretion is important for metabolic wellbeing in diabetes and may prevent the development of severe ketoacidosis when insulin delivery is inadequate.

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Introduction

Increased blood ketone-body concentrations and a tendency to develop severe ketoacidosis are characteristic of insulin-dependent type I diabetes mellitus. Nevertheless, patients vary widely in their susceptibility to ketoacidosis. Changes in catabolic hormone concentrations certainly influence the

development of ketoacidosis,[1-4] but insulin deficiency per se must remain a key factor. Studies of endogenous insulin secretion based on plasma C-peptide measurements show that almost all insulin-dependent patients have demonstrable residual beta-cell function during the first years of the disease,[5 6] while even after 10 years some 15% may show residual function.[5]

We decided to assess the role of this preserved beta-cell function in preventing the development of ketoacidosis in insulin-dependent diabetes.

Patients and methods

Fifteen diabetic patients were studied. All were considered to be insulin-dependent because of proneness to ketosis and weight loss when not given exogenous insulin at the onset or within the first year of the disease. The table gives their clinical characteristics. Seven patients were selected because no endogenous insulin secretion could be detected by means of C-peptide measurements in plasma before and six minutes after an intravenous injection of 1 mg glucagon.[7] The eight other patients had a mean fasting plasma C-peptide concentration of 0·15 (range 0·05-0·24) nmol/l, which increased to 0·23 (0·06-0·43) nmol/l six minutes after an intravenous injection of 1 mg glucagon. Corresponding values in 18 normal people were 0·35 (0·25-0·63) and 1·20 (0·86-1·88) nmol/l respectively.[8] All the patients were well educated in self-adjustment of their insulin dosage and diet according to frequent testing of preprandial urine specimens with Clinitest and Ketostix. In accordance with the Helsinki declaration, we carefully informed all patients about the purpose and design of the study before obtaining their consent.

Clinical characteristics of patients with and without beta-cell function

	Patients with beta-cell function (n=8)		Patients without beta-cell function (n=7)		P
	Mean	Range	Mean	Range	
Age (years)	26·5	17–33	23·7	17–31	NS
Duration of diabetes (years)	3·2	1–6	6·3	1–11	NS
% of ideal body weight	94	87–103	94	79–116	NS
Insulin dose (IU/day)	32	19–42	49	36–58	0·05
Insulin binding to IgG (U/l)	0·112	0–0·529	0·500	0·015–1·531	NS

NS = Not significant.

To ensure clearance of subcutaneous depots of intermediate-acting insulin[9] only Actrapid insulin was used during the two days before admission. Insulin was taken before main meals and at 2200 in doses adjusted according to urine test results. None showed ketonuria. Five patients experienced slight hypoglycaemic reactions during the first half-day. Patients took their last dose of insulin at 1700 on the evening of admission followed by their usual meal at 1730 and a snack at 2000. They were admitted to our metabolic ward between 2000 and 2200. None had ketonuria, and blood glucose concentrations in patients with and without beta-cell function ranged from 5·0 to 20·0 mmol/l (90 to 360 mg/100 ml) (mean 9·6 mmol/l; 173 mg/100 ml) and 7·2 to 19·0 mmol/l (130 to 342 mg/100 ml) (mean 12·1 mmol/l; 218 mg/100 ml) respectively.

An overnight insulin infusion (Actrapid) was begun between 2200 and 2400 at an initial rate of 2 IU/hour This was adjusted according to frequent blood glucose measurements and aimed at achieving a normal blood glucose concentration by 0800. Insulin was then withdrawn for 12 hours. The patients were fasted and confined to bed from the start of the infusion until the end of the insulin-deprivation period. The patients' clinical condition and blood glucose and urinary ketone-body concentrations were assessed at frequent intervals during insulin deprivation. None of the patients showed symptoms attributable to hyperglycaemia or ketoacidosis. Urinary ketone-body concentrations never exceeded ++ (Ketostix), and blood glucose concentration did not exceed 25 mmol/l (450 mg/100 ml) in any case. At the end of the deprivation period all patients were given 0·1 IU Actrapid/kg body weight intravenously as a bolus injection followed by frequent subcutaneous injections until euglycaemia was re-established. All patients were discharged next morning with good metabolic control.

Blood samples were taken one hour before, at zero time, and 1, 2, 3, 4, 5, 6, 8, 10, and 12 hours after the start of insulin deprivation via an indwelling cannula inserted into a brachial vein. Samples were cooled immediately, the plasma being separated within five minutes and kept below −20 C until analysis. An aliquot of blood (0·5 ml) was also placed in 2 ml ice-cold perchloric acid (5% v/v) and the supernatant stored at −20 C for subsequent analysis. Blood glucose was measured by a glucose oxidase method. Lactate, alanine, glycerol, 3-hydroxybutyrate, and non-esterified fatty acid concentrations were determined as described.[10 11] C-peptide was assayed with 1230 antibody,[7] a plasma C-peptide concentration exceeding 0·05 nmol/l indicating preserved beta-cell function.[7] Insulin binding to IgG was measured by radioimmunoelectrophoresis.[12]

The Mann-Whitney rank sum test or Wilcoxon's test was used to compare mean concentrations, and Spearman's rank correlation test to calculate the coefficient of correlation. The level of type I error (2α) was set at 0·05.

Results

Figures 1 and 2 show the mean changes in blood glucose, metabolite, and plasma C-peptide concentrations during insulin deprivation. At the beginning of insulin deprivation there were no significant differences in the concentration of any metabolite between patients with and without residual beta-cell function. Blood glucose values were slightly increased in both groups (6·0 mmol/l (range 5·3-7·2 mmol/l) and 7·2 mmol/l (5·6-9·7 mmol/l)—108 mg/100 ml (range 96-130 mg/100 ml) and 129 mg/100 ml (101-175 mg/100 ml) —respectively), but there was no significant difference between them (fig 1). All the metabolite concentrations were within normal limits.

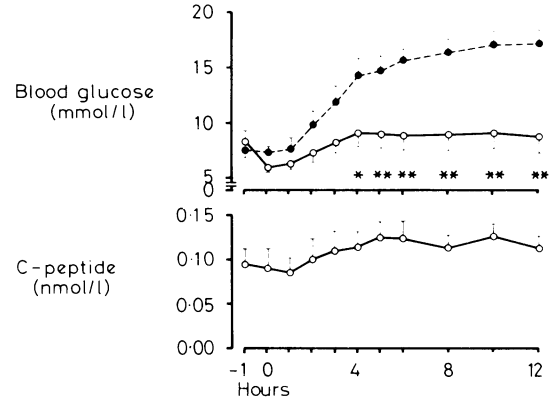

FIG 1—Mean (±SE of mean) changes in blood glucose and C-peptide concentrations after insulin deprivation. ○—○ Patients with beta-cell function. ●—● Patients without beta-cell function. *P < 0·05. **P < 0·01.
Conversion: SI to traditional units—Blood glucose: 1 mmol/l ≈ 18 mg/100 ml.

During the 12 hours of insulin deprivation blood glucose, 3-hydroxybutyrate, glycerol, and non-esterified fatty acids reached much higher concentrations in patients without residual beta-cell function than in patients with residual secretion (fig 2). Plasma alanine and lactate concentrations, however, were unchanged.

In both groups blood glucose concentrations were significantly ($P = 0·01$) above basal values after three hours of insulin deprivation. In patients with residual secretion blood glucose then remained constant at about 9 mmol/l (162 mg/100 ml). In the other patients values increased steadily to 17·2 mmol/l (310 mg/100 ml) (range 13·0-22·5 mmol/l; 234-405 mg/100 ml) after 12 hours of insulin deprivation and 24 hours of fasting.

3-Hydroxybutyrate concentrations were significantly higher than basal values after one hour in both groups. At the end of the study the mean 3-hydroxybutyrate concentration was 1·43 mmol/l (14·9 mg/100 ml) (range 0·40-2·88 mmol/l; 4·2-30·0 mg/100 ml) in patients with beta-cell function but 5·47 mmol/l (56·9 mg/100 ml) (3·70-6·81 mmol/l; 38·5-70·9 mg/100 ml) in patients without residual secretion ($P < 0·01$).

Glycerol concentrations were significantly increased above basal

values after two hours in both groups (P = 0·02). Thereafter patients without beta-cell function showed a higher mean concentration, which was significant during the last four hours (P < 0·05 at eight hours; P < 0·01 at 10 and 12 hours). Plasma non-esterified fatty-acid concentrations rose significantly during the last six hours in both groups, again a much greater increase occurring in patients without residual secretion.

C-peptide concentrations in the insulin-secretory group showed a mean increase closely correlated with the mean blood glucose concentration (r = 0·83; P = 0·02). In no patient did the C-peptide concentration reach the lower limit of normal fasting values. The individual mean C-peptide concentrations showed a significant inverse correlation with the final blood glucose concentration (r = 0·91; P < 0·02). Correlations between C-peptide values and glycerol (r = −0·71), 3-hydroxybutyrate (r = −0·51), and non-esterified fatty-acid concentrations (r = −0·36) failed to reach significance.

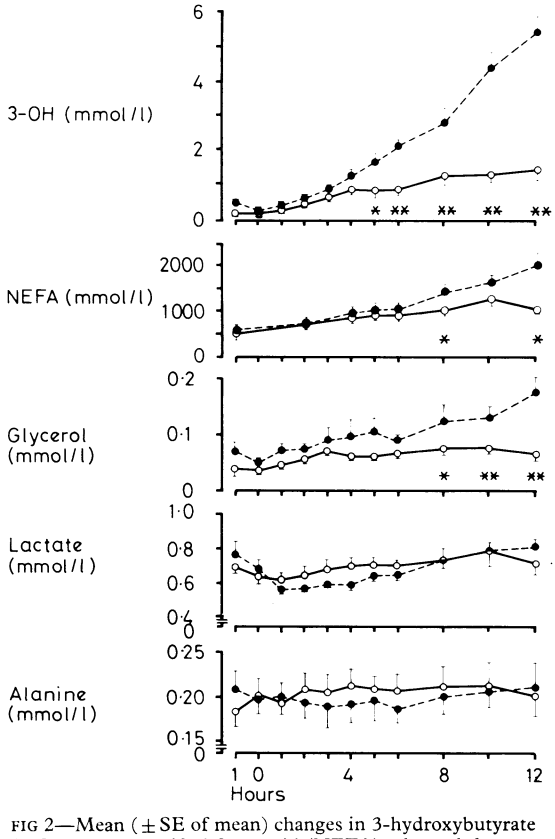

FIG 2—Mean (±SE of mean) changes in 3-hydroxybutyrate (3-OH), non-esterified fatty-acid (NEFA), glycerol, lactate, and alanine concentrations after insulin deprivation. ○ = Patients with beta-cell function. ● = Patients without beta-cell function. *P < 0·05. **P < 0·01.

Conversion: SI to traditional units—3-Hydroxybutyrate: 1 mmol/l ≈ 10·4 mg/100 ml. Non-esterified fatty acid: 1 mmol/l = 1 mEq/l. Glycerol: 1 mmol/l ≈ 9·2 mg/100 ml. Lactate: 1 mmol/l ≈ 9·0 mg/100 ml. Alanine: 1 mmol/l ≈ 8·9 mg/100 ml.

All patients without beta-cell function showed small but significant amounts of insulin binding to IgG, with a mean of 0·50 U/l (range 0·015-1·531 U/l). The concentration of this IgG was inversely and significantly (P < 0·05) correlated with the final blood glucose concentration (r = −0·82) and final concentration of 3-hydroxybutyrate (r = −0·89) but not with any other metabolite.

Discussion

We find that about one-third of insulin-dependent diabetic patients retain some capacity to secrete insulin[5] and that their beta cells can modulate insulin secretion in response to variations in blood glucose concentration.[6 13] Compared with patients without residual beta-cell function such patients generally achieve satisfactory metabolic control despite small doses of insulin.[13–16] We have now provided further evidence of the metabolic importance of residual beta-cell function in insulin-dependent diabetes. Thus while fasting and being deprived of exogenous insulin patients who had retained on average 20% of normal insulin secretion developed a much attenuated degree of hyperglycaemia and hyperketonaemia. Indeed their increase in 3-hydroxybutyrate concentration was similar to that observed in normal subjects during an equivalent period of fasting.[17]

These results, obtained in otherwise healthy diabetic patients during fasting and under metabolic ward conditions, do not imply that patients with some beta-cell function may not develop hyperglycaemia and ketoacidosis. They strongly suggest, however, that the residual insulin secretion may protect against the development of severe ketoacidosis. This accords with the clinical impression that few insulin-dependent diabetic patients develop severe ketoacidosis during the first years of insulin treatment—that is, when they have demonstrable residual endogenous insulin production.[5 6]

Other studies of the metabolic response to insulin deprivation have shown great variability between patients.[1–4] This may partly be explained by the inclusion of patients both with and without residual insulin secretion. Varying insulin delivery into the blood stream from subcutaneous depots of intermediate-acting insulin may also contribute to the varying metabolic responses.[9] This was not the case in our study because the patients had been treated with crystalline neutral insulin (Actrapid) subcutaneously for two days and then intravenously for some 10 hours before insulin deprivation.

A circulating depot of insulin bound to insulin antibodies may also contribute to the heterogeneity of metabolic responses. Our discovery of a close correlation between insulin binding to IgG and hyperglycaemia in the patients without residual beta-cell function was highly suggestive of such an effect.

Studies on the development of ketoacidosis have emphasised the importance of the anti-insulin hormones, particularly glucagon.[1–4] Our findings strongly suggest that residual insulin secretion is also of major importance. The results also show the importance of evaluating endogenous insulin secretion in studies of the pathophysiology of ketoacidosis.

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