CLINICAL RESEARCH

Human ultralente insulin

R R HOLMAN, J STEEMSON, P DARLING, W G REEVES, R C TURNER

Abstract

The greater solubility of human insulin and its possible faster action have led to doubts about whether a sufficiently long acting formulation could be produced to provide a basal supply for diabetics. In a double blind crossover study in 18 diabetics human ultralente insulin was as effective as beef ultralente insulin in controlling basal plasma glucose concentrations (median 5·7 mmol/l (103 mg/100 ml) with human and 6·3 mmol/l (114 mg/100 ml) with beef ultralente insulin respectively). There was no significant difference between human and bovine insulin in the rise in plasma glucose concentration from 0400 to 0700 after an injection the previous morning and no difference between patients receiving an adequate or insufficient dose of human ultralente insulin. Bovine insulin antibody binding was reduced with human insulin ($p < 0.002$), which suggests that human insulin is less antigenic than beef insulin.

Once daily human ultralente insulin provides a suitable formulation for the basal insulin requirement of diabetics.

Introduction

The progression of diabetic tissue damage appears to be prevented by improved diabetic control.[1-3] Although continuous subcutaneous insulin infusion has been used to induce near normoglycaemia,[4] it is not yet generally applicable to all patients, and an alternative, easily managed regimen has been to provide the basal insulin requirement with a once daily subcutaneous injection[5] of very long acting ultralente insulin[6] and then to give additional soluble insulin to cover meals if necessary.[7]

Antibodies to even highly purified beef ultralente insulin develop in most patients and may be a problem in some.[8 9] Synthetic or semisynthetic human insulins theoretically ought not to be antigenic, although polymerisation or deamidation may initiate an immune response.[10] In addition, some clinical studies with short and medium acting human insulin preparations have shown both a more rapid onset and shorter duration of action.[11 12]

We carried out a study aimed at determining whether human ultralente insulin can be successfully used in place of beef ultralente insulin.

Patients and methods

We carried out a double blind crossover study comparing conventional highly purified beef ultralente with semisynthetic human ultralente insulin. Eighteen diabetic patients in good health and receiving stable treatment gave informed consent for the study. Nine had type II diabetes and were taking once daily beef ultralente insulin (Ultratard MC, Novo) alone in a dose sufficient to decrease their stressed fasting plasma glucose concentration to below 6 mmol/l (108 mg/100 ml)[13]; other preprandial plasma glucose concentrations were 4-7 mmol/l (72-126 mg/100 ml). The remaining nine patients essentially had type I diabetes and were taking beef ultralente insulin but required additional twice daily soluble pork insulin (Actrapid MC, Novo). At entry all patients were switched to insulins of the same strength (40 U/ml), asked to take their ultralente injection first thing in the morning, and requested to do home capillary blood glucose monitoring, aiming for preprandial concentrations of 4-7 mmol/l. All were supplied with appropriate disposable plastic syringes.

After a two week run in period patients were randomised to receive either human ultralente insulin (Ultratard HM, Novo) or highly purified beef ultralente insulin for six weeks and were then switched to the alternative insulin for a further six weeks. The soluble insulin was pork throughout the study. Patients were seen fortnightly in a special clinic set aside for the purpose, and advice was available by telephone at any time. At each visit blood was drawn for estimation of the stressed fasting plasma glucose concentration and haemoglobin A_{1c} concentration and the patient was weighed and asked about hypoglycaemic episodes. After each six week period patients were admitted overnight for a plasma glucose profile, samples being taken from 1730 to 0700. They took their normal insulin, evening meal, and bedtime snack.

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Plasma glucose concentration was measured manually by a glucose oxidase method and haemoglobin A_{1c} concentration by an isoelectrofocusing method[14] in which the dialysed sample was incubated for five hours at 37°C to exclude short term glucose adducts. Plasma immunoreactive C peptide concentration was measured by radioimmunoassay,[5] cholesterol concentration with a Technicon autoanalyser by the Liebmann-Burchard reaction, and lipoprotein concentrations by precipitation with heparin manganese chloride[15] and sodium dodecyl sulphate.[16] IgG antibodies, reactive with bovine or human insulin, were measured by an immunochemical method.[8]

The nocturnal profiles of glucose concentrations were compared time point by time point in mmol/l, as the area under the glucose concentration time curve/min from 1730 to 0700 (mean glucose concentration) and from 0200 to 0700 (mean basal glucose concentration) and as the change in value from 0400 to 0700. Parametric analyses include Student's paired t test with $\log_{10}$ transformed triglyceride concentrations and insulin antibody values. Results are given as means (1 SD) except where stated.

Results

Table I gives the demographic details of the patients at entry to the study. Those patients taking ultralente plus soluble insulins were

TABLE I—*Demographic details of patients according to whether they received ultralente insulin alone or ultralente plus additional soluble insulin. Figures are mean (SD) values (and ranges)*

	Ultralente alone	Ultralente + soluble
No of patients	9	9
Men:women	7:2	7:2
Age (years)	55·2 (8·9) (35-62)	43·4 (11·1) (25-61)
Body mass index (kg/m²)	28·4 (5·9) (23·8-42·6)	26·9 (5·0) (21·5-35·5)
C peptide (pmol/l):		
Basal	0·46 (0·28) (0·10-0·81)	0·15 (0·23) (0·01-0·71)
2 hours postprandially	1·16 (0·51) (0·42-2·15)	0·25 (0·36) (0·02-1·09)
Insulin dose (IU/kg):		
Ultralente	0·31 (0·15) (0·12-0·60)	0·56 (0·19) (0·28-0·85)
Soluble		0·26 (0·20) (0·09-0·74)

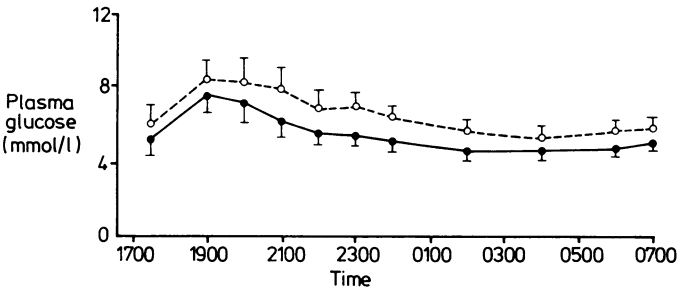

FIG 1—Overnight plasma glucose concentrations (mean (1 SEM)) in nine patients with type II diabetes while taking beef (- - - - -) and human (————) ultralente insulin.

Conversion: SI to traditional units—Glucose: 1 mmol/l ≈ 18 mg/100 ml.

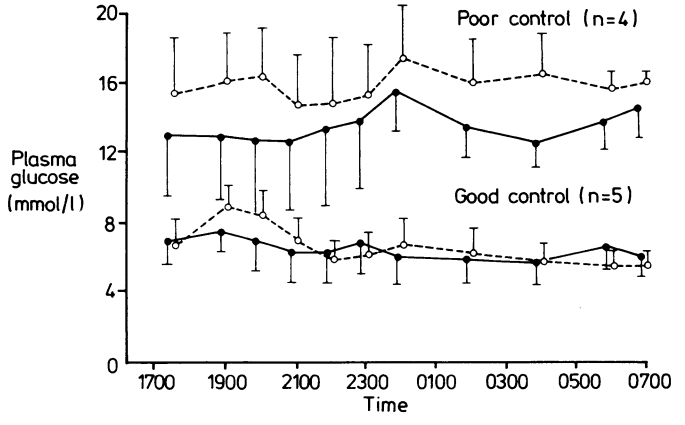

FIG 2—Overnight plasma glucose concentrations (mean (1 SEM)) in nine patients with type I diabetes while taking beef (- - - - -) and human (————) ultralente insulin according to control.

Conversion: SI to traditional units—Glucose: 1 mmol/l ≈ 18 mg/100 ml.

difference was not significant when either individual times or the mean basal glucose concentrations (fig 3) were compared. There was no significant difference in the rise in plasma glucose concentrations from 0400 to 0700 or in the haemoglobin A_{1c} concentrations (table II). The mean haemoglobin A_{1c} concentrations in those patients

TABLE II—*Mean (SD) values after six weeks' treatment with beef or human ultralente insulin. Some patients received additional soluble insulin*

	Ultralente alone		Ultralente + soluble	
	Beef	Human	Beef	Human
Weight (kg)	81·4 (9·6)	81·9 (9·8)	78·2 (14·0)	80·0 (14·4)
Insulin dose (IU/kg):				
Ultralente	0·31 (0·15)	0·29 (0·14)	0·56 (0·19)	0·53 (0·19)
Soluble			0·26 (0·20)	0·24 (0·22)
Haemoglobin A_{1c} (%)	7·7 (1·1)	7·3 (1·1)	10·1 (1·3)	10·8 (1·6)
Basal glucose (mmol/l)	5·7 (1·7)	4·7 (1·2)	10·5 (6·4)	9·4 (5·0)
Change in glucose between 0400 and 0700 (mmol/l)	0·5 (0·3)	0·3 (0·5)	−0·4 (2·3)	1·2 (1·3)
Insulin antibodies ($\log_{10}$ μg/l):				
Bovine	0·94 (0·31)	0·71 (0·24)	0·87 (0·47)	0·76 (0·46)
Human	0·73 (0·29)	0·51 (0·23)	0·63 (0·44)	0·45 (0·40)
Triglycerides (mmol/l)*	1·95 (2·84, −1·15)	1·94 (3·50, −1·25)	1·94 (3·67, −1·27)	2·43 (3·37, −1·41)
LDL cholesterol (mmol/l)	3·29 (0·83)	3·24 (0·86)	2·82 (0·60)	3·02 (1·09)
HDL cholesterol (mmol/l)	1·15 (0·42)	1·18 (0·30)	1·24 (0·33)	1·70 (1·63)

LDL = Low density lipoprotein. HDL = High density lipoprotein.
*Expressed as $\log_{10}$ mean ± SD.
Conversion: SI to traditional units—Glucose: 1 mmol/l ≈ 18 mg/100 ml. Cholesterol: 1 mmol/l ≈ 38·7 mg/100 ml.

on average younger ($p<0·05$), had lower mean basal C peptide concentrations ($p<0·05$) and lower C peptide concentrations two hours postprandially ($p<0·001$), and required larger mean doses of ultralente insulin ($p<0·01$) than those patients taking ultralente insulin alone. There was no significant difference in the mean body mass index between the two groups, and the ratio of men to women was identical.

No significant weight changes were observed during the study. Mean plasma glucose concentrations tended to be lower (figs 1 and 2) when patients were taking human ultralente insulin, although the taking ultralente insulin alone was within the normal range (5·3-8·5%) with both beef and human ultralente insulin. Mean plasma triglyceride, low density lipoprotein cholesterol, and high density lipoprotein cholesterol concentrations were not significantly altered.

While taking human ultralente insulin three patients (two taking ultralente alone, one taking ultralente plus Actrapid) had to have their insulin doses reduced (36 IU to 26, 18 IU to 16, and 32 IU to 26) because of hypoglycaemic episodes. Despite this reduction these patients still had lower basal plasma glucose concentrations when taking human insulin (5·1 *v* 3·2 mmol/l (77 *v* 58 mg/100 ml); 4·3 *v*

3·5 mmol/l (77 *v* 63 mg/100 ml); and 19·9 *v* 17·0 mmol/l (359 *v* 306 mg/100 ml) respectively). The geometric means for bovine insulin antibody binding and human insulin antibody binding were reduced after six weeks' treatment with human ultralente compared with beef ultralente insulin ($p<0·002$ and $p<0·001$ respectively) (fig 4). There was no significant relation between individual changes in plasma glucose concentrations and insulin antibody concentrations.

Discussion

Human ultralente insulin was as effective as beef ultralente insulin in maintaining basal plasma glucose concentrations in patients with type II diabetes and in those with type I diabetes requiring additional short acting insulin. There was no clinical evidence in this study, of patients with insulin antibodies, to suggest that human ultralente insulin has an appreciably shorter half life of absorption from the skin than beef ultralente insulin in that there was no demonstrable tendency for the plasma glucose concentrations to rise overnight. Indeed, basal plasma glucose concentrations tended to be lower during treatment with human insulin, and three patients had lower basal plasma glucose concentrations even though their doses of human ultralente insulin were reduced because of hypoglycaemia. The significantly reduced insulin antibody binding suggests that semisynthetic human ultralente insulin is less antigenic than the beef equivalent. As formation of antibodies to even highly purified beef ultralente insulin sometimes leads to increasing insulin requirements, painful skin reactions, and occasionally lipoatrophy, human ultralente insulin may be advantageous, and its long duration of action means that once daily injections can provide the basal insulin requirement.

In this study the ultralente insulin was given as a morning injection whereas in routine use it is probably more appropriately given in the evening to help counteract any tendency for the early morning fasting plasma glucose concentration to rise. In view of the simplicity of providing a basal insulin supplement by a single daily injection of human ultralente insulin[5] it is doubtful whether the expense and additional risks of the alternative implanted constant rate infusion pump[17] are warranted.

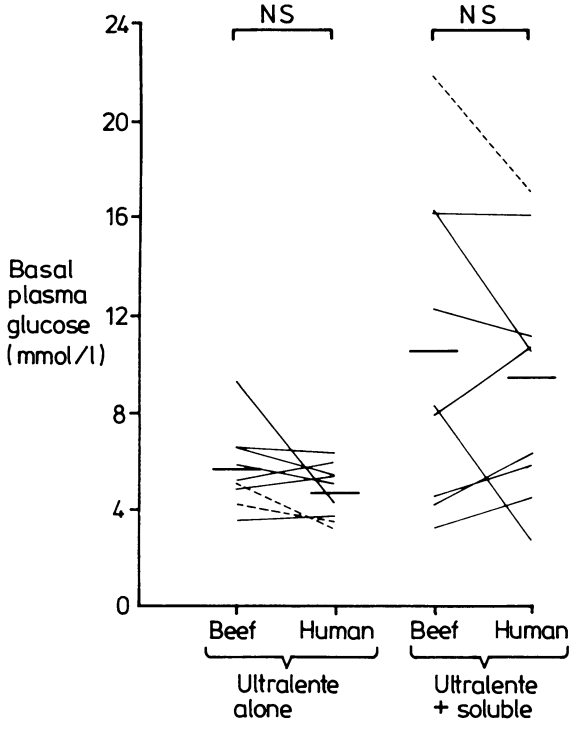

FIG 3—Basal plasma glucose concentrations (mean area under glucose concentration time curve/min from 0200 to 0700) in each patient while taking beef and human ultralente insulin. Horizontal bars denote mean values. Dotted lines indicate the three patients in whom the dose of human ultralente insulin was reduced because of hypoglycaemia.

Conversion: SI to traditional units—Glucose: 1 mmol/l ≈ 18 mg/100 ml.

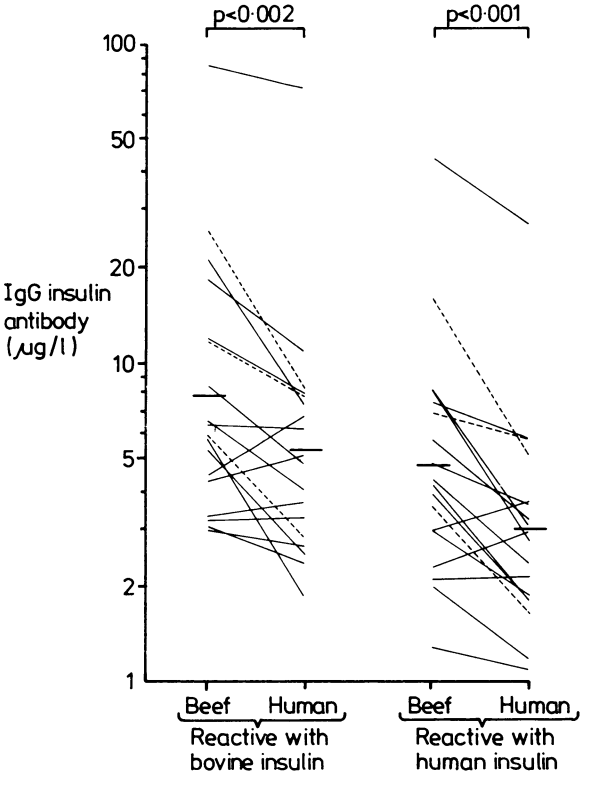

FIG 4—Bovine and human insulin antibody binding concentrations after six weeks' treatment with beef and human ultralente insulin in random order. Horizontal bars denote geometric mean values. Dotted lines indicate the three patients in whom the dose of human ultralente insulin was reduced because of hypoglycaemia.

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The sicca syndrome in thalassaemia major

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Abstract

A 20 year old man with β thalassaemia developed symptoms of the sicca syndrome. His serum contained rheumatoid factor and antinuclear antibodies. A biopsy specimen of labial salivary gland showed large accumulations of haemosiderin within the parenchymal cells of the acini.

Although in this case the sicca syndrome could not be definitely distinguished from Sjögren's syndrome, the patient's HLA type was not the one usually associated with Sjögren's syndrome. Histological appearances suggested that the causative factor of the sicca syndrome was iron overload owing to an intensive blood transfusion regimen.

Introduction

Iron overload has been reported to have produced the sicca syndrome, xerostomia, and xerophthalmia in a patient with idiopathic haemochromatosis.[1] We report here the first case of the sicca syndrome in a young adult with β thalassaemia major and severe iron overload owing to repeated blood transfusions.

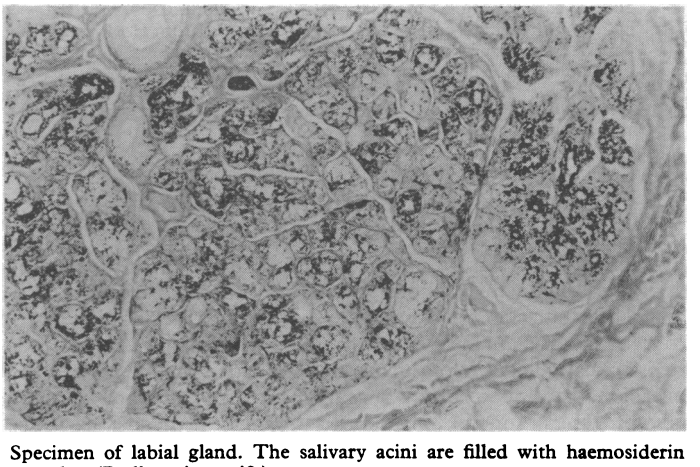
Case report

The patient was a 20 year old man in whom β thalassaemia major had been diagnosed at the age of 1 year. Transfusions were started a month later and continued up to the time of writing at intervals of one to two months. The mean haemoglobin concentration before transfusion had not been below 9·3 g/dl throughout the past three years. We calculated from clinical records that he had received a total of over 100 l blood, containing about 50 g iron. Splenectomy was performed at age 3. There was no history of viral hepatitis. Chelation treatment was started in 1975 with desferrioxamine mesylate intramuscularly, 500 mg/day five days a week; from the beginning of 1980 he self administered an average dose of 2 g desferrioxamine mesylate subcutaneously over eight hours on alternate days. In the past year, in an attempt to reduce the iron load, he had undergone monthly wash outs with intravenous desferrioxamine mesylate, 300 mg/kg/24 h (16 g/24 h). In 1981, at age 19, he developed diabetes requiring insulin treatment. In July 1982 he had an episode of cardiac failure, which responded readily to diuretics and digitalis. Puberty did not develop spontaneously, and he was therefore given human chorionic gonadotrophin with some improvement in sexual maturation.

In April 1983 he complained of dry mouth and itchy eyes of a few weeks' duration. Pertinent clinical findings, apart from a dry scaly tongue, included moderately hyperpigmented skin and a large, hard liver palpable 7 cm below the right costal margin. No swelling of the parotid or lachrymal gland was noticeable. Liver function tests showed an alanine transferase activity three times the upper limit of normal, prothrombin activity 50% of normal, albumin concentration 35 g/l, and gammaglobulin concentration 3·2 g/100 ml. Serum was negative for hepatitis B surface antigen and anti-e antibodies and positive for anti-s and anti-c antibodies. Fibrinogen concentration was 2·8 g/l. Rheumatoid factor was present in the serum, and antinuclear antibodies were present at a titre of 1/500, with a speckled pattern. No organ specific antibodies were found. HLA typing showed B13, Bw35, DR5, and DR7. The serum ferritin concentration was 3·3 μg/l, and the average urinary excretion of iron after subcutaneous desferrioxamine mesylate was 645 μmol (36 mg)/24 h; excretion doubled when the high intravenous dose was administered. Faecal excretion of chymotrypsin was below normal. Schirmer's test gave a result of 3 mm.

Ophthalmological examination with fluorescein showed areas of abnormal epithelial cells on the cornea. A biopsy specimen of the labial salivary gland showed large accumulations of haemosiderin within the parenchymal cells of the acini. There was no evidence of lymphocytic infiltrate, cellular destruction, or interstitial fibrosis (figure).

Treatment with bromhexine and artificial tears was prescribed with little subjective improvement.



Specimen of labial gland. The salivary acini are filled with haemosiderin granules. (Perl's stain. ×43.)

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