The effect of a low-calorie diet with and without fenfluramine, and fenfluramine alone on the glucose tolerance and insulin secretion of overweight non-diabetics

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Summary

Glucose tolerance and insulin secretion have been measured in six overweight non-diabetic subjects on first presentation and after 5 weeks of fenfluramine treatment without dietary restriction. There was a significant improvement in glucose tolerance and a marginally significant decrease in insulin secretion.

In another six overweight non-diabetic subjects, insulin secretion and glucose tolerance were measured on first presentation, after 10 weeks of low-calorie diet, and then after a further 10 weeks of the same low-calorie diet with the addition of fenfluramine. Diet alone did not produce any significant effect on glucose tolerance, but did bring about a significant decrease in insulin secretion. The addition of fenfluramine to the dietary therapy was associated with a marginally significant improvement in the glucose tolerance and a highly significant decrease in insulin secretion. This further decrease in insulin secretion was significantly greater than the decrease produced by diet alone in these six subjects.

Introduction

Obesity in non-diabetics is known to be associated with high insulin secretion in response to glucose (Yalow *et al.*, 1965; Bagdade, Bierman & Porte, 1967; Nikkilä & Taskinen, 1971). Weight reduction through low-calorie diet decreases the hyperinsulinism (Salans, Knittle & Hirsch, 1968; Farrant, Neville & Stewart, 1969; Kalkhoff *et al.*, 1971) and improves glucose tolerance (Farrant *et al.*, 1969; Kalkhoff *et al.*, 1971).

Fenfluramine, a non-addictive therapeutic agent chemically related to amphetamine, has appetite-depressing activity (Munro, Seaton & Duncan, 1966) and is used together with dietary control in the treatment of obesity since it produces a greater loss in weight than other anorexigenic amphetamines (Craddock, 1969). An indication that fenfluramine might have a further action in addition to its appetite-depressing property arose when von Herold, Kemper & Opitz (1965) recorded that the drug brought about a reduction of the blood glucose concentration in rats. Subsequently, using the forearm preparation of Butterfield & Holling (1959), several workers have shown that intra-arterial infusion of the drug increases the uptake of glucose and oxygen

by muscle independently of alteration in blood flow (Whichelow *et al.*, 1971; Turtle, Burgess & Bauckham, 1971).

Carbohydrate metabolism has been further investigated in obese non-diabetic subjects by studying the effect of fenfluramine alone and as an aid to dietary treatment, on glucose tolerance and insulin secretion.

Patients and methods

Six obese non-diabetic subjects had a baseline oral glucose-tolerance and insulin-secretion test before and then 5 weeks after treatment with fenfluramine. 40 mg fenfluramine was given twice daily to four of the patients, the remaining two receiving 20 mg twice daily and 60 mg twice daily respectively.

Baseline oral glucose-tolerance and insulin-secretion tests were carried out on another six obese non-diabetic subjects who were then treated for a period of 10 weeks with low-carbohydrate, low-calorie diet. A second oral glucose-tolerance and insulin-secretion test was performed at the end of this time. Fenfluramine 40 mg twice daily was then added to the treatment and the subjects continued on the diet plus fenfluramine for a further period of 10 weeks; a third oral glucose-tolerance and insulin-secretion test was then carried out.

All the subjects were more than 10% overweight as calculated from the height and ideal weight tables issued by the American Society of Actuaries, 1959.

Normal controls

A normal range of insulin secretion was obtained from twenty-one normal individuals (eight males and thirteen females) age range 36-63 years. Each had a normal oral glucose-tolerance test, was within 5% of his or her ideal body weight and had none of the associations of the prediabetic state (large babies, family history, etc.). In order to establish the reproducibility of measurement of the parameters being studied, oral glucose-tolerance and insulin-secretion tests were repeated on eleven of these normal subjects (six males and five females).

Laboratory estimations

For at least 4 days before the initial oral glucose-tolerance and insulin-secretion test, each subject had eaten a diet containing at least 200 g carbohydrate/

day. Each test was carried out as follows: after the fasting venous blood specimen was taken, each subject ingested 50 g glucose in 400 ml water within 3 min. Venous blood specimens were taken at 15, 30, 60, 90, 120 and 150 min for glucose and insulin assay. Urine specimens were collected immediately before, and at 1 and 2 hr after the glucose ingestion, and were tested semi-quantitatively for sugar content by Clinitest tablets. All subjects fasted for 13 hr prior to each test. Fenfluramine was withdrawn for 13–15 hr before each test.

The specimens were assayed for glucose by the AutoAnalyzer glucose oxidase method of Morley, Dawson & Marks (1968), the coefficient of variation being 4–6% at a blood glucose concentration of 105 mg/100 ml. Plasma immuno-reactive insulin was measured in duplicate by the method of Hales & Randle (1963) using radioactive iodinated insulin and Oxoid membrane filters supplied by the Radiochemical Centre, Amersham. The pre-precipitated antibody and standard human insulin were supplied by Burroughs Wellcome. The standard deviation of the assay method was (a) $\pm 2.92 \mu\text{U/ml}$ for 115 duplicate determinations of 0–49 $\mu\text{U/ml}$, (b) $\pm 6.51 \mu\text{U/ml}$ for 64 duplicate determinations of 50–99 $\mu\text{U/ml}$ and (c) $\pm 7.25 \mu\text{U/ml}$ for 55 duplicate determinations of 100–200 $\mu\text{U/ml}$. If a plasma insulin concentration $> 200 \mu\text{U/ml}$ was found, it was diluted to $< 200 \mu\text{U/ml}$, and then re-assayed. Otherwise all specimens from one subject were determined in the same batch.

Statistical methods of comparison

The area under the glucose-tolerance test curve from 0 to $2\frac{1}{2}$ hr was measured as an index of glucose tolerance (Burns *et al.*, 1965; Bagdade *et al.*, 1967); a decrease in the area represents an improvement in glucose tolerance. Insulin secretion was estimated by the area under the plasma insulin curve from 0 to $2\frac{1}{2}$ hr (Perley & Kipnis, 1965). The highest figure obtained for insulin secretion from these normal controls was 5003 $\mu\text{U-min/ml}$. All the obese subjects studied secreted quantities of insulin greater than this on first clinical presentation.

In order to compare the glucose-tolerance index and insulin secretion from each test on each patient with the results from the test immediately preceding, it was necessary to take account of the amount of change due to variation in estimation of the parameters. To obtain an assessment, the difference between the two glucose-tolerance indices from each of the eleven normal subjects on whom the oral glucose-tolerance test was repeated, was expressed as a percentage of the mean of the two indices. In the obese patients, the difference between the glucose-tolerance indices of two consecutive tests was expressed as a percentage of the mean of the two

indices. Similar percentages were calculated for the differences in the respective insulin secretions. Knowing the reproducibility of the test as shown by the eleven normal controls, the probability that the changes in glucose tolerance and insulin secretion were due to random variation in the estimation of these parameters was calculated. This was done by statistically comparing the patients' percentages with those from the controls by the non-parametric Mann-Whitney U-test described by Sokal & Rohlf (1969).

The results on the obese patients on fenfluramine alone were compared with the reproducibility results on the eleven subjects.

The results on the obese patients on diet plus fenfluramine were compared with: (1) the reproducibility results on the eleven normal subjects, (2) the changes in the results due to diet alone for the first 10 weeks in the same obese subjects.

Results

Figures 1 and 2 show on a semi-logarithmic scale the actual glucose tolerance and insulin secretion of: (a) twenty-one normal subjects, (b) the eleven normal subjects on whom a repeat test was performed and (c) twelve obese non-diabetic subjects, baseline and after therapy. Table 1 demonstrates the statistical significance of dietary therapy alone and diet-plus-fenfluramine on these parameters.

Discussion

Treatment of six obese non-diabetic subjects with fenfluramine alone was associated with a significant improvement in glucose tolerance and a marginally significant decrease in insulin secretion ($0.1 > P > 0.05$).

Treatment of a further six subjects for 10 weeks on low-calorie diet alone produced weight-loss in all cases, a significant decrease in insulin secretion ($0.05 > P > 0.02$) but no significant change in glucose tolerance. Addition of fenfluramine to the dietary treatment for a further period of 10 weeks in these subjects produced further weight-loss in all cases, a marginally significant improvement in glucose tolerance ($0.1 > P > 0.05$) and a further and highly significant decrease in insulin secretion ($P < 0.02$) when the results were compared with the reproducibility of estimation of the parameters in normal subjects (see Table 1). When the results on the six subjects on diet-plus-fenfluramine during the second period of 10 weeks were compared with those on the same six subjects on diet alone for the first period of 10 weeks, there was no significant effect of the addition of fenfluramine on the glucose tolerance, but the drug did produce a further significant decrease in insulin secretion over the decrease produced by diet alone ($P = 0.05$) (see Table 1). In fact the insulin

FIG. 1. Glucose tolerance of: (a) twenty-one normal subjects, (b) the eleven normal subjects on whom a repeat test was performed, (c) twelve obese non-diabetic subjects baseline and after therapy. ●—●, diet alone; ● - - - ●, diet plus fenfluramine; x—x, fenfluramine alone.

FIG. 2. Insulin secretion of: (a) twenty-one normal subjects, (b) the eleven normal subjects on whom a repeat test was performed, (c) twelve obese non-diabetic subjects, baseline and after therapy. ●—●, diet alone; ● - - - ●, diet plus fenfluramine; x—x, fenfluramine alone.

secretion in three of the subjects was reduced to within the normal range, i.e. <5003 $\mu\text{U}\cdot\text{min}/\text{ml}$ (Fig. 2).

Farrant *et al.* (1969) studied nine obese non-diabetic subjects and found that they lost from between 9 and 61 kg in body weight in a period of

dietary restriction varying between 5 and 39 weeks. They showed a reduction in output of immuno-reactive insulin during the oral glucose-tolerance test (OGTT) and a less marked reduction in the mean concentration of plasma glucose. Kalkhoff *et al.* (1971) studied six obese subjects given OGTTs before

TABLE 1. Statistical significance of dietary therapy alone and of dietary therapy plus fenfluramine on the glucose tolerance and insulin secretion of obese non-diabetic subjects when the results are compared with controls

	Obese non-diabetic subjects on diet alone for first 10 weeks		Obese non-diabetic subjects on diet plus fenfluramine for second 10 weeks	
	Change in: Glucose tolerance	Change in: Insulin secretion	Change in: Glucose tolerance	Change in: Insulin secretion
Reproducibility of glucose tolerance in normal subjects	NS*	—	Marginally significant 0.1 > P > 0.05	—
Reproducibility of insulin secretion in normal subjects	—	Significant decrease 0.05 > P > 0.02	—	Significant decrease P < 0.02
Glucose tolerance of same obese patients on diet alone for first 10 weeks	—	—	NS*	—
Insulin secretion of same obese patients on diet alone for first 10 weeks	—	—	—	Significant decrease P = 0.05

* NS, not significant

and after a period of dietary restriction ranging from 10 to 18 months and involving an average weight loss of 85 lb and a decrease from 83 to 27% above ideal body weight; associated with this, these workers showed a significant improvement in glucose tolerance and decrease in insulin secretion. In the subjects described here the diets were of similar calorific value and carbohydrate content to those in the two other investigations mentioned. During the 10-week period on diet alone, the weight losses ranged from 2 to 10 kg, there was a trend towards improvement in glucose tolerance, and a significant fall in insulin secretion ($0.05 > P > 0.02$). Hence, it would seem as if the reduction in insulin secretion occurs before the improvement in glucose tolerance as one of the metabolic effects of weight loss due to dietary restriction in obesity. The addition of fenfluramine to the dietary therapy would seem to accelerate the return of normal carbohydrate metabolism, since (1) 5 weeks on fenfluramine alone without dietary restriction brought about a significant improvement in glucose tolerance and a marginally significant decrease in insulin secretion, and (2) addition of fenfluramine to the dietary therapy for a further 10 weeks of treatment after treatment with dietary therapy alone, brought about a marginally significant improvement in glucose tolerance and a highly significant decrease in insulin secretion, the latter being significantly greater than that produced by diet alone in the same subjects. Fenfluramine has a similar effect to dietary restriction in reversing some of the abnormalities in carbohydrate metabolism in obesity and it appears to act more rapidly than does diet alone and to act very effectively when given together with dietary control.

It would seem that in situations of high insulin secretion, e.g. obesity and 'high-insulin-secreting' obese maturity-onset diabetes (Dykes, 1973), treatment with a low carbohydrate diet and fenfluramine is highly effective in lowering the level of insulin. It remains to be seen, however, if this effect is important therapeutically in view of the suggested association between high insulin concentrations and the development of atheroma (Stout & Vallance-Owen, 1969).

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