Clofibrate enhances the affinity of insulin receptors in non-insulin dependent diabetes mellitus

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Glucose-lowering mechanism by clofibrate was studied in non-insulin dependent diabetics managed with dietary therapy alone. Clofibrate 1500 mg was administered for 1 month to 15 patients, and 75 g oral glucose tolerance test and insulin tolerance test were carried out before and after 1 month of the treatment. Fasting plasma glucose values were decreased from 9.34 ± 0.53 mmol/l to 7.58 ± 0.33 mmol/l ($P < 0.01$), and fasting insulin levels were decreased from 13.8 ± 1.7 μ u/ml to 10.1 ± 1.5 μ u/ml ($P < 0.05$). However, insulinogenic index and Σ IRI/ Σ glucose ratio during 75 g oral glucose tolerance test were not changed. Enhanced glucose fall in insulin tolerance test was also observed. As these results suggested the enhanced tissue sensitivity to insulin, we examined the insulin binding to erythrocytes before and at 3 months' treatment of clofibrate in 10 non-insulin dependent diabetics. Insulin bindings were increased from $3.41 \pm 0.17\%$ to $4.11 \pm 0.20\%$ in the presence of 1 ng/ml of native insulin ($P < 0.01$). This increased binding was due to an increased affinity without a change in the number of insulin receptors. These results suggest that improved glucose tolerance by clofibrate might be derived from the enhanced tissue sensitivity to insulin, probably through an enhanced affinity of insulin receptors.

Keywords clofibrate glucose tolerance insulin receptor

Introduction

The glucose-lowering effect of clofibrate in normal (Ferrari *et al.*, 1978), non-insulin dependent diabetic (Barnett *et al.*, 1977) and in insulin dependent diabetic patients (Miller, 1963, Schade *et al.*, 1978) has been recognized, but its mechanism remains unknown. The study reported here was designed to clarify the glucose-lowering mechanism by clofibrate in non-insulin dependent diabetics receiving diet therapy alone.

Methods

Eight female and seven male Japanese non-insulin dependent diabetics were studied. The

mean age, percentage of ideal body weight, serum total cholesterol and triglyceride were 59.7 ± 2.7 years, $114.4 \pm 3.6\%$, 5.71 ± 0.27 mmol/l and 1.97 ± 0.23 mmol/l, respectively. All patients were treated with diet therapy alone. Stability of body weight and plasma glucose levels for 1 month prior to the study were necessary criteria, and clofibrate, 1500 mg/day in three divided doses, was given for 1 month. They received no medication except clofibrate. There was no significant change in body weight during study, 61.2 ± 2.5 kg and 61.8 ± 2.4 kg before and after 1 month of the treatment, respectively. Oral glucose tolerance test (75 g GTT) and intravenous insulin tolerance test (ITT) (0.1 unit/kg of mono component Actrapid insulin) were carried out before and after 1 month of the treatment.

In another study, the effect of clofibrate on

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insulin binding to erythrocytes was examined in five female and five male Japanese non-insulin dependent diabetic patients before and at 3 months of the treatment with the same dosage. Their mean age, percentage of ideal body weight and fasting plasma glucose values were 56.3 ± 1.7 years, $114.3 \pm 6.7\%$ and 8.97 ± 0.88 mmol/l, respectively. Their diabetes was treated with diet therapy alone, and the stability of body weight before and during the study was kept constant statistically. Methods of preparation of erythrocytes and insulin binding assay have been described elsewhere (Kobayashi *et al.*, 1980).

In the results, the sum of all blood samples in 75 g GTT was expressed as Σ glucose or Σ IRI, and similarly Δ IRI or Δ glucose mean the change in levels. Plasma glucose was measured by a glucose oxidase method and immunoreactive insulin (IRI) by radioimmunoassay using single antibody technique (Wide & Porath, 1966). The results were expressed as mean $\pm$ s.e. mean, and unpaired and paired Student's *t*-test were used in statistical analysis.

Results

After the 1 month clofibrate treatment, the mean fasting plasma glucose values decreased from 9.34 ± 0.53 mmol/l to 7.58 ± 0.33 mmol/l ($P < 0.01$). Plasma glucose values during 75 g GTT showed a significant decrease at each point (0, 30, 60, 90, 120 and 180 min) ($P < 0.01$), and Σ glucose decreased by $20.6 \pm 2.4\%$ ($P < 0.01$).

Figure 1 Correlation between 75 g GTT-induced Σ IRI/ Σ glucose ratio before and after 1 month of clofibrate treatment in non-insulin dependent diabetics. $y = 1.08x + 0.00$, $r = 0.87$, $P < 0.01$.

Fasting IRI values decreased from 13.8 ± 1.7 μ u/ml to 10.1 ± 1.5 μ u/ml ($P < 0.05$) in spite of the improved glucose tolerance. However, insulinogenic index (Δ IRI/ Δ plasma glucose at 30 min) and Σ IRI/ Σ glucose ratio during 75 g GTT did not change in each patient statistically (Figure 1). Nadir plasma glucose values in ITT were decreased from $41.9 \pm 3.9\%$ to $27.0 \pm 3.5\%$ of the zero time values ($P < 0.01$) after the 1 month treatment with clofibrate. In spite of the fall of fasting plasma free fatty acid (FFA) value from 515.6 ± 50.5 mmol/l to 378.1 ± 33.8 mmol/l, no statistically significant correlation was observed between the decrease of FFA levels and the improved glucose tolerance.

Following treatment with clofibrate insulin binding to erythrocytes increased significantly from $4.25 \pm 0.21\%$ to $5.73 \pm 0.31\%$ ($P < 0.001$) and from $3.41 \pm 0.17\%$ to $4.11 \pm 0.20\%$ ($P < 0.01$) in the presence of 0 and 1 ng/ml, respectively, of native insulin (Figure 2). This enhancement was observed in every patient. The Scatchard analysis showed that this increased binding was due to an increased affinity with no change in the number of insulin receptors. The calculated mean number of binding sites/erythrocyte obtained before and at 3 months of the treatment were 39 and 38 sites/cell, which is less than an average number for normal controls of 52 sites/cell.

Discussion

It has been recognized that clofibrate improves glucose tolerance in diabetics, however, its glucose-lowering mechanism is not well known. Since it has been suggested that hypoglycaemic action of clofibrate, in part, might be derived from the displacement of protein bound oral hypoglycaemic drugs (Daubresse *et al.*, 1976) in patients receiving oral hypoglycaemic drugs, we examined the mechanism in non-insulin dependent diabetic patients managed with dietary therapy alone to observe the primary hypoglycaemic action. The reduction of plasma FFA levels has been considered as one of several possible causes of this effect (Balasse & Neef, 1974), but this reduction is not specific to clofibrate which affects lipid metabolism. In fact, no significant correlation was observed between the reduction of plasma FFA levels and the improved glucose tolerance in our study, which suggests that the reduction of plasma FFA levels by clofibrate does not necessarily explain its glucose-lowering mechanism. On the other hand, decreased fasting IRI levels, no statistical change in insulinogenic index and Σ IRI/ Σ glucose ratio

Figure 2 Changes in insulin binding by erythrocytes from non-insulin dependent diabetics ($n = 10$) before (O) and after (●) 3 months of clofibrate treatment. * $P < 0.01$, ** $P < 0.001$.

during 75 g GTT together with the results of ITT suggested an enhanced tissue sensitivity to insulin. This result is compatible with the reports that doses of insulin were reduced by clofibrate treatment in insulin dependent diabetics (Miller, 1963; Schade *et al.*, 1978), which also means an enhanced tissue sensitivity to exogenous insulin because they have no insulin releasing capacity. Therefore, we examined the effect of clofibrate on insulin binding to erythrocytes. The insulin

binding was enhanced by clofibrate treatment, and its increased binding was due to an increased affinity with no change in the number of insulin receptors. Although the reason for the enhanced affinity of insulin receptors following clofibrate treatment remains to be clarified, it does appear from these results that it is this enhanced binding of insulin which is responsible for the improvement of glucose tolerance in diabetics following clofibrate treatment.

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