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Short communication

Masoprocol (nordihydroguaiaretic acid): a new antihyperglycemic agent isolated from the creosote bush (*Larrea tridentata*)

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Abstract

An ethnomedically-driven approach was used to evaluate the ability of a pure compound isolated from the creosote bush (*Larrea tridentata*) to lower plasma glucose concentration in two mouse models of type 2 diabetes. The results indicated that plasma glucose concentration fell approximately 8 mmol/l in male C57BL/ks-*db/db* or C57BL/6J-*ob/ob* mice following the oral administration of masoprocol (nordihydroguaiaretic acid), a well known lipoxygenase inhibitor. The decline in plasma glucose concentration following masoprocol treatment in the mice was achieved without any change in plasma insulin concentration. In addition, oral glucose tolerance improved and the ability of insulin to lower plasma glucose concentrations was accentuated in masoprocol-treated *db/db* mice. These data raise the possibility that masoprocol, or other lipoxygenase inhibitors, represents a new approach to the pharmacological treatment of Type 2 diabetes. © 1998 Elsevier Science B.V.

Keywords: Masoprocol; Nordihydroguaiaretic acid; Diabetes, type 2; Anti-hyperglycemic compound; Insulin action

1. Introduction

Plants used to treat non-insulin-dependent diabetes mellitus (NIDDM) offer valuable leads for the development of pharmaceuticals (Luo et al., 1998; Oubré et al., 1997). Oral decoctions and extracts of the creosote bush (*Larrea tridentata*) have been used by the Pima Indians in the Southwest United States and Mexico for the treatment of diabetes (Winkelman, 1989). Masoprocol (nordihydroguaiaretic acid) a pure compound from the creosote bush, was evaluated in mouse models of NIDDM for its ability to lower blood glucose concentration. Since this compound is of a chemical class not previously known to have antihyperglycemic effects, we believe it is useful to describe its characteristics in this short communication.

2. Material and methods

Male C57BL/ks-*db/db* mice (*db/db* mice), or male C57BL/6J-*ob/ob* mice (*ob/ob* mice), 8–9 weeks old, were purchased from the Jackson Laboratory (Bar Harbor, ME, USA). Animals were housed (four mice/cage) under standard laboratory conditions at 22°C and 50% relative humidity, and were maintained on a diet of Purina rodent chow and water ad libitum unless indicated otherwise. Prior to treatment, blood was collected from the tail vein of each animal and mice that had extreme plasma glucose concentrations, (>33 or <17 mmol/l) were excluded. Each treatment group consisted of 4–8 mice, distributed so that the mean glucose levels were equivalent in each group at the start of each study. Mice were dosed orally by gavage with either vehicle or masoprocol (nordihydroguaiaretic acid) at 150 mg/kg twice a day. This dose was based upon the results of preliminary experiments indicating that it was the *lowest* dose that

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reduced plasma glucose concentrations consistently in mouse models of Type II diabetes. The duration of the treatment varied among experiments and is indicated in individual experiments. Testing materials were delivered in a liquid vehicle containing 0.25% (w/v) carboxymethylcellulose. Blood samples were taken from the tail vein 3 h after dosing on the sampling day in non-fasted conditions unless indicated otherwise. Individual body weight and mean food consumption (each cage) were measured every 1–3 days.

Oral glucose tolerance test and short insulin tolerance tests were performed in *db/db* mice, treated for 3 days, followed by an overnight (16 h) fast. For oral glucose tolerance test, the morning dose of masoprocol was given and the oral glucose tolerance test was performed 3 h later by giving a 2 g/kg glucose load orally. Blood samples were taken for plasma glucose measurement at 0, 15, 30, 60 and 120 min after the glucose load. The morning dose of masoprocol was omitted before the short insulin tolerance test. Mice were injected intravenously in the morning after the overnight fast with 0.5 U/kg of regular insulin and blood samples were taken for plasma glucose measurement at 3, 6, 9, 12, 15 and 18 min after the insulin injection. Plasma glucose concentrations were determined with the Glucose Diagnostic Kit from Sigma Chemical (St. Louis, MO), and plasma insulin levels with the Rat Insulin RIA Kit from Linco Research (St. Charles, MO).

Data are expressed as mean ± S.E.M., and one-way analysis of variance (ANOVA) or Student's *t*-test were used where appropriate to assess statistical significance of differences.

3. Results

The anti-hyperglycemic effect of masoprocol in *db/db* mice is shown in Fig. 1. Plasma glucose concentrations fell progressively with masoprocol treatment, and by day 12 there was a 8.5 mmol/l difference as compared to vehicle (20.7 ± 2.6 vs. 29.2 ± 1.1 mmol/l, $P < 0.01$, $n = 8$). Plasma insulin concentrations (726 ± 78 vs. 540 ± 96 pmol/l), food intake (5.8 vs. 5.2 g/mouse per day) and body weight (40.3 ± 1.3 vs. 40.7 ± 0.8 g/mouse) were similar in these two groups when measured on day 12. In addition, the total integrated glucose response area over baseline was significantly ($P < 0.01$) lower (1954 ± 105 vs. 2570 ± 118 mmol/l min, $n = 8$) following an oral glucose load in *db/db* mice treated with masoprocol. To further evaluate the antihyperglycemic activity of masoprocol, studies were also performed in *ob/ob* mice. The protocol used was similar to that described for *db/db* mice, with the exception that the effect was measured after 3 days of masoprocol (150 mg b.i.d.) treatment. In this instance, plasma glucose concentration was also significantly ($P < 0.01$) lower in masoprocol-treated mice (15.4 ± 1.4 vs. 22.4 ± 1.1 mmol/l, $n = 8$ in each group). As in the case of the *db/db* mice, plasma insulin concentrations did not change with masoprocol treatment (3972 ± 828 vs. 4014 ± 468 pmol/l), and both body weight (52.1 ± 0.8 vs. 52.0 ± 0.7 g/mouse) and food consumption (4.3 vs. 4.0 g/mouse per day) were similar in the two groups.

Since masoprocol lowered plasma glucose in *db/db* mice without any increase in insulin concentration, we tested the hypothesis that it was acting to enhance insulin

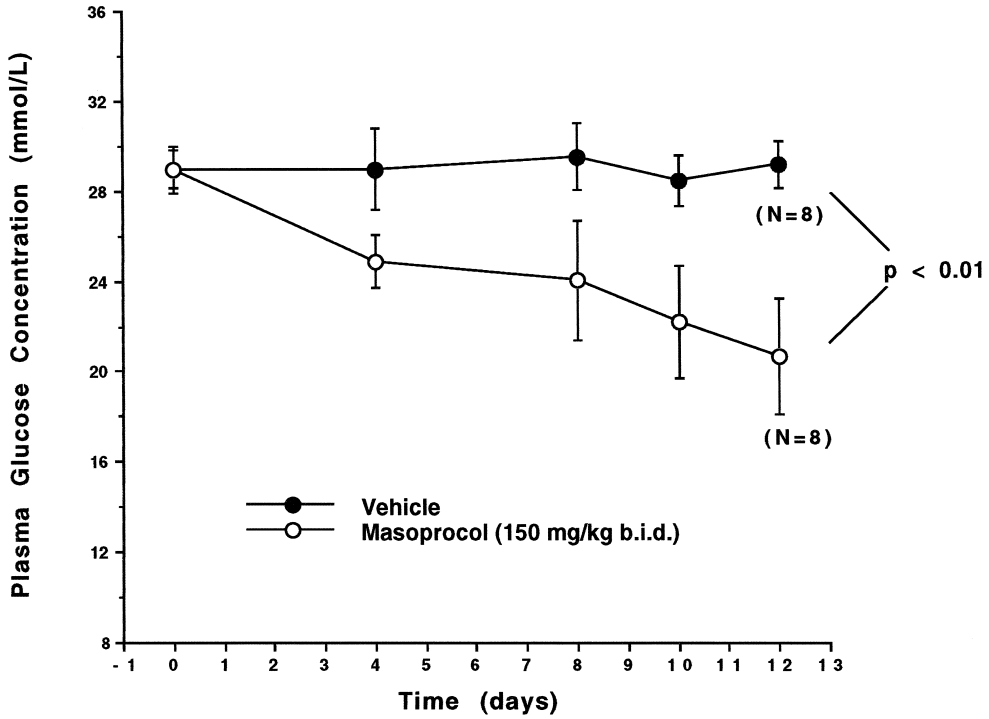

Fig. 1. Plasma glucose concentrations in *db/db* mice receiving either vehicle or masoprocol over a 12-day treatment period ($n = 8$).

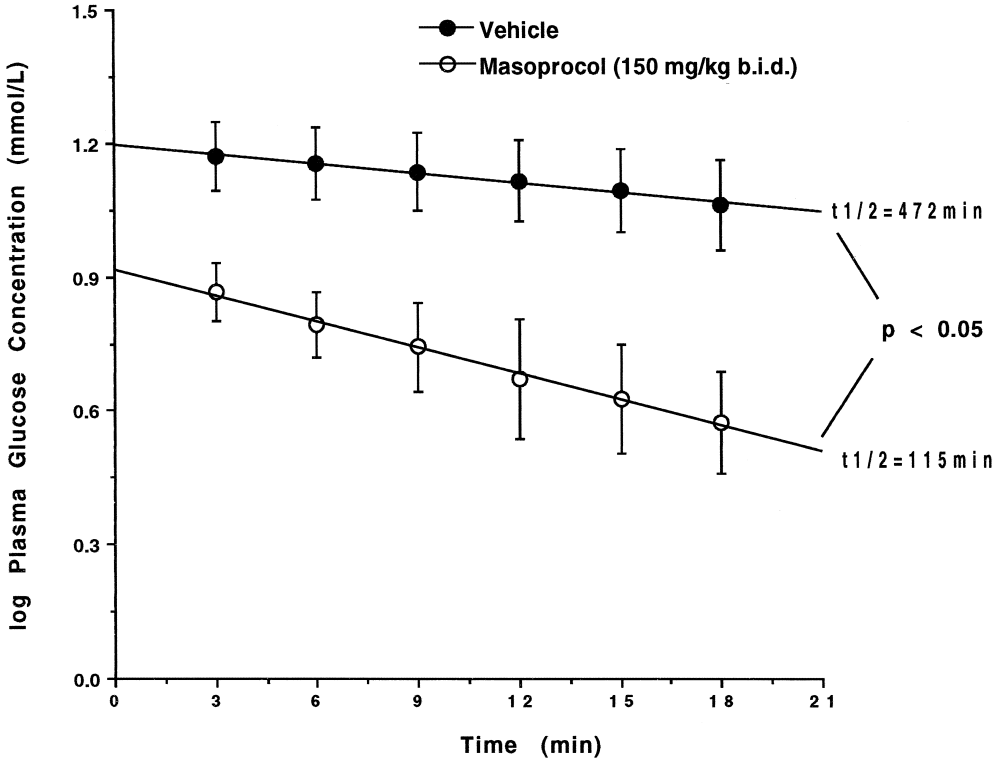

Fig. 2. Plasma glucose clearance following insulin injection in vehicle and masoprocol treated mice ($n = 5$).

action. This suggestion was supported by the results of the short insulin tolerance test shown in Fig. 2, indicating that the ability of insulin to decrease plasma glucose concentration was accentuated in *db/db* mice treated with masoprocol (half-time of removal decreased from 472 ± 39 to 115 ± 33 min, $P < 0.05$, $n = 5$).

4. Discussion

Masoprocol is a pure compound isolated from the creosote bush, *L. tridentata*, and our discovery that this compound, a lipoxygenase inhibitor (Schewe et al., 1987), was the anti-hyperglycemic fraction isolated from the creosote bush, was surprising for two reasons. In the first place, we are unaware of any previous publication demonstrating that lipoxygenase inhibitors lower plasma glucose concentrations in rodent models of diabetes. Secondly, and perhaps even more confounding, was the fact that masoprocol has in the past been identified as an in vitro inhibitor of glucose-stimulated β-cell insulin secretion (Yamamoto et al., 1982). Given this information, it might have been predicted that masoprocol would be diabetogenic, rather than a compound that can lower plasma glucose concentration in mouse models of NIDDM.

In conclusion, masoprocol, a known lipoxygenase inhibitor, has been identified as a component of the creosote bush capable of lowering plasma glucose concentration when given orally to two mouse models of NIDDM. The anti-hyperglycemic effects of masoprocol appear to be at least partially due to an enhancement of insulin action. Whether or not masoprocol will eventually develop into a useful drug for the treatment of NIDDM remains to be seen. At the least, we believe that the identification of lipoxygenase inhibitors as a possible new class of anti-diabetic drugs is worth noting.

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