Hypoglycemic effect of *Malmea depressa* root on streptozotocin diabetic rats

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Abstract

The hypoglycemic effects of water, ethanolic and butanolic extracts prepared from the root of *Malmea depressa* (Baill) R.E. Fries. (Annonaceae) were studied in diabetic rats (streptozotocin induced). Oral application of water extracts at doses of 40 and 80 mg/kg, ethanolic (112 mg/kg) and butanolic (80 mg/kg) extracts significantly lowered the plasma glucose levels in diabetic rats within three hours. Glibenclamide and metformin were used as references and showed similar hypoglycemic effects like the extracts. The three extracts have a similar chemical composition (HPLC analysis).

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1. Introduction

*Malmea depressa* (Baill) R.E. Fries. (Annonaceae) were studied in diabetic rats (streptozotocin induced). Oral application of water extracts at doses of 40 and 80 mg/kg, ethanolic (112 mg/kg) and butanolic (80 mg/kg) extracts significantly lowered the plasma glucose levels in diabetic rats within three hours. Glibenclamide and metformin were used as references and showed similar hypoglycemic effects like the extracts. The three extracts have a similar chemical composition (HPLC analysis).

Although there exist ethnobotanical reports as a hypoglycemic plant there is no scientific report about its efficacy. The aim of our study was to investigate the hypoglycemic effect of water, ethanolic and butanolic extracts in streptozotocin (STZ)-induced diabetic rats and to identify the main chemical constituents occurring in the applied extracts.

2. Materials and methods

According to our previous studies (Andrade-Cetto et al., 2000; Andrade-Cetto and Wiedenfeld, 2001; Andrade-Cetto and Wiedenfeld, 2004) and further definitions (Holmsted, 1991), we perform an ethnopharmacological study with re-
2.5. Induction of experimental diabetes

Diabetes was induced by a single intraperitoneal injection of a freshly prepared streptozotocin (STZ) solution (Sigma, no. 242-646-8) (65 mg/kg in acetate buffer 0.1 M, pH 4.5) to overnight-fasted rats. Control rats received only the buffer. Diabetes was identified by polydipsia, polyuria and by measuring non-fasting plasma glucose levels 48 h after injection of STZ. Animals which did not develop more than 250 mg/dl glucose levels were rejected.

2.6. Experimental groups

The diabetic animals were classified into eight groups (1–8) each of them with eleven rats. Group 1 as non diabetic control received 1.5 ml of physiological NaCl-solution (vehicle), group 2 as the diabetic control received also 1.5 ml of physiological NaCl-solution (vehicle), the rats of group 3 were given the standard oral hypoglycemic agent glibenclamide (3 mg/kg bodyweight (bw)) in the same vehicle, group 4 received the hypoglycemic agent metformin (14 mg/kg bw), groups 5 and 6 received WE (40 mg/kg bw) and WE (80 mg/kg bw), respectively, group 7 received ET (112 mg/kg bw) and group 8 received BE (80 mg/kg bw). The extracts were redissolved in 1.5 ml of physiological NaCl-solution and administered orally by a canule.

2.7. Collection of blood and determination of blood glucose

Blood samples were taken from the tail vein (according to the Guideline 9/3(10/099) IACUC) before oral administration of the extracts or the vehicle and at times 0, 60, 120 and 180 min thereafter. Thirty-two microliters of blood were collected from each rat and placed on a Heparinized cap (Roche). The plasma glucose levels were measured in plasma serum with Reflotron equipment and confirmed by Accutrend GC and Accu-check compact equipments (Roche).

2.8. Statistical analysis

The data were statistically analyzed by ANOVA tukey test. The plasma glucose levels were expressed as the mean (S.E.M.).

3. Results

3.1. Ethnobotany

The results of our field study in Chikinzonot Yucatan confirmed that Malmea depressa is used as a hypoglycemic agent against diabetes type 2, it is also used against kidney problems. The tree is locally named by its Maya name “Elemuy”. In general, the people drink the infusion of the root after boiling 30–36 g in 1 l water; also the same amount
of plant is overnight macerated on cold water. The tea or the macerated is drunk during the day as “agua de uso”. The dry root is sold at the main market place in Merida, Yucatan, with the same preparation way and a similar recommended dose, furthermore an ethanolic preparation was found, this preparation is recommended for the treatment of type 2 diabetes, some drops (about 1.5 ml) are added to 250 ml water, and drunk several times per day. Those results confirm the previously reported use of *Malmea depressa* for the treatment of diabetes type 2.

### 3.2. Compounds

A similar composition of ingredients in all three extracts was confirmed by HPLC analysis (Fig. 1). Because both water preparations (hot decoction and cold maceration) show the same compounds only the first preparation was tested in the animal model.

The structures of the isolated compounds (Fig. 2) were established by spectroscopical methods as phenylbutane-derivatives (2-hydroxy-3,4,5-trimethoxy-1-(2′,4′-dihydroxy-3′-dihydroxy)butyl-benzene 1; 2-hydroxy-3,4,5-trimethoxy-1-(2′,3′,4′-hydroxy)butyl-benzene 2).

### 3.3. Activity in diabetic rats

STZ administration at a dosage of 65 mg/kg bw to normal rats significantly elevated the blood glucose levels compared with rats injected citrate buffer alone (Table 1) as in previous reports (Andrade-Cetto et al., 2000; Andrade-Cetto and Wiedenfeld, 2001; Andrade-Cetto and Wiedenfeld, 2004).

In our diabetic rats, the extracts showed significant hypoglycemic effects (Table 1). The water extract at doses of 40 and 80 mg/kg bw showed activity at 120 and 180 min, with a significant reduction (*p* < 0.01) of plasma glucose levels. The water extract at a dosage of 40 mg/kg bw did not show significant activity at 60 min while at dose of 80 mg/kg the hypoglycemic effect show statistical significance (*P* < 0.01).

The ethanolic (112 mg/kg) and the butanolic (80 mg/kg) extracts led to a significant decrease in plasma glucose level compared with the control. The effect was significant from 60 min ongoing until 180 min, the ET presented a higher effect at 60 min. The maximum activity of both extracts was observed after 180 min, too.

Glibenclamide (3 mg/kg) and the metformin (14 mg/kg) produced a significant decrease in plasma glucose from 60 min until 180.

These results indicate that there is no significant difference between the tested plant preparations in compar-
Effect of oral administration of extracts of *Malmea depressa* root on plasma glucose concentration in diabetic rats

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Time (min)</th>
<th>Blood glucose levels (mg/dl) ± standard error</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (−) no diabetic (vehicle)</td>
<td>T0</td>
<td>131 ± 6a</td>
</tr>
<tr>
<td>Control (+) no diabetic (vehicle)</td>
<td>T0</td>
<td>143 ± 2a</td>
</tr>
<tr>
<td>Control (−) (vehicle)</td>
<td>T60</td>
<td>137 ± 3a</td>
</tr>
<tr>
<td>Control (+) no diabetic (vehicle)</td>
<td>T120</td>
<td>132 ± 5a</td>
</tr>
<tr>
<td>Control (+) no diabetic (vehicle)</td>
<td>T180</td>
<td>141 ± 3b</td>
</tr>
<tr>
<td>Glibenclamide (15 mg/Kg bw)</td>
<td>T0</td>
<td>408 ± 26a</td>
</tr>
<tr>
<td>Glibenclamide (15 mg/Kg bw)</td>
<td>T60</td>
<td>413 ± 19b</td>
</tr>
<tr>
<td>Metformin (14.16 mg/Kg bw)</td>
<td>T0</td>
<td>340 ± 26a</td>
</tr>
<tr>
<td>Metformin (14.16 mg/Kg bw)</td>
<td>T60</td>
<td>317 ± 20b</td>
</tr>
<tr>
<td>Metformin (14.16 mg/Kg bw)</td>
<td>T120</td>
<td>318 ± 19b</td>
</tr>
<tr>
<td><em>Malmea depressa</em> aqueous extract (40 mg/Kg bw)</td>
<td>T0</td>
<td>390 ± 20a</td>
</tr>
<tr>
<td><em>Malmea depressa</em> aqueous extract (40 mg/Kg bw)</td>
<td>T60</td>
<td>396 ± 21b</td>
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<tr>
<td><em>Malmea depressa</em> aqueous extract (40 mg/Kg bw)</td>
<td>T120</td>
<td>396 ± 21b</td>
</tr>
<tr>
<td><em>Malmea depressa</em> aqueous extract (40 mg/Kg bw)</td>
<td>T180</td>
<td>396 ± 21b</td>
</tr>
</tbody>
</table>

The values represent the mean ± S.E.M. as compared with control time intervals.

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**References**


