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Hypoglycemic effect of *Rheum ribes* roots in alloxan induced diabetic and normal mice

by ***Hanefi Özbek*, Ebubekir Ceylan[1], Mehmet Kara[2], Fevzi Özgökçe[3], Mehmet Koyuncu[4].***

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Summary

A decoction extract of *Rheum ribes* roots was given orally and tested for hypoglycemic effect in healthy and in alloxan-diabetic mice. The results were compared with glibenclamide, a standard hypoglycemic agent, and control groups. It was found that the extract possesses a hypoglycemic effect in diabetic animals.

Introduction

Rheum ribes is locally known as *"ışgın, uşgun or uçgun"* and found mostly in Eastern Turkey, Lebanon and Iran. The roots of *Rheum ribes*, collected from Bingöl, contain tannins (8%) and anthracene derivatives (0.025%) (*Baytop, 1999*). According to the results of the chemical study on material collected from Erzincan, chrysophanol, physcion, rhein, aloe-emodin, physcion-8-O-glucoside, aloe-emodin-8-O-glucoside, sennoside A and rhaponticin were found in the subterranean parts of the plants. The amount of the anthracene derivatives is about 0.1%. Its fresh stems and petioles are consumed as a vegetable. The same parts of the plants are also used as a digestive and appetizer in Bitlis. The subterranean parts of the plants are used for the treatment of hemorrhoids and diabetes (*Tuzlacı et al., 1991*).

In view of the fact that there is no systematic study on the hypoglycemic effect of *Rheum ribes*, an extract of the roots of *Rheum ribes* was tried on healthy and on diabetic mice and its effects were compared with glibenclamide, a standard hypoglycemic agent.

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Materials and Methods

Plant material

The roots of *Rheum ribes* were collected freshly from Van, Turkey. The roots were air-dried at room temperature and stored in bottles until use. The material was identified in the Department of Medical Biology, Faculty of Medicine. A voucher specimen (B-05) has been kept in our laboratory for future reference. The dried roots (100 g) were powdered in an electric blender, put in boiling water (1litre) for 10 min and the mixture then left to cool to room temperature. The decoction was filtered through a Whatman No 1 filter paper, and the resulting extract used in the biological assays below.

Chemicals

Alloxan was obtained from Sigma (Steinheim, Germany) and glibenclamide was obtained from Nobel Ilaç Sanayii (İstanbul, Turkey).

Experimental animals

Male, conventional, outbred Swiss albino mice weighing 22-27 g were maintained in the Animal House of Yüzüncü Yıl University, Faculty of Medicine. The mice were bred in our institution animal house but the lineage was originally obtained from Ankara Health Protection Institute (a governmental organisation). All animals were housed in standard cages (48x35x22 cm) at room temparature

(20±2°C) with artificial light from 7.00 am to 7.00 pm, and provided with pelleted food (Van Animal Feed Factory, Van-Turkey) and water ad libitum. Ambient relative humidity was 55-60%. Prior to each study, the animals were subjected to fasting for 18 h. The protocol for the study was approved by the Ethical Committee of Animal Breeding and Research, Yüzüncü Yıl University, Faculty of Medicine (2001/03-138).

Induction of experimental diabetes

Experimental diabetes in mice, which had been subjected to fasting for 18 h, was induced by intraperitoneal (i.p.) administration of alloxan (*Rodriguez et al., 1975*). The total dose of alloxan (450 mg/kg body weight) was administered in three injections at intervals of 48 h (150 mg/kg body weight each time). Seven days after the last administration, the animals were fasted for 18 h and blood glucose levels were determined.

Biological assays

Hypoglycemic activity of Rheum ribes root extract in alloxan-diabetic mice

Alloxan-induced diabetic mice were divided into three groups of 10 animals each (I-III). Group I served as control, group II served as reference and group III served as the test animals. All groups were fasted for 18 h. Group I received only isotonic saline solution (ISS) (5 ml/kg, p.o.). Group II received fast-action glibenclamide as reference (3 mg/kg, p.o.). Group III received the decoction extract administered orally (5 ml/kg) by gavage.

Hypoglycemic activity of Rheum ribes root extract in healthy mice

Healthy mice were divided into two groups of 10 animals each (IV-V). Group IV served as control, and group V served as the test animals. Both groups were fasted for 18 h. Group IV received only isotonic saline solution (5 ml/kg, p.o.). Group V received the decoction extract administered orally (5 ml/kg) by gavage.

Blood samples were obtained from the tail vein in fasting animals, and 0, 1, 2, 4 and 24 h after administration of the test substance. Glycemia was determined by the glucose-oxidase peroxidase method with reagent strips and their evaluation was made on the Prestige LX™ Blood Glucose System (U.S.).

Statistical analysis

Blood glucose levels in groups were expressed as mean ± Standard Error of Mean (S.E.M). The data were statistically analysed by the Student's *t*- test and one-way ANOVA followed by Tukey's test. P values less than 0.05 were considered significant (*Sümbüloğlu, 1998*).

Results

The mean blood glucose of fasted animals at various time intervals after p.o. administration of decoction extract of *Rheum ribes* roots, glibenclamide and isotonic saline solution in alloxan-induced diabetic mice are shown in Table I. The mean blood glucose of fasted animals at various time intervals after p.o. administration of decoction extract of *Rheum ribes* roots and isotonic saline solution in healthy mice are shown in Table II.

Conclusion

Rheum ribes has been used as a traditional Turkish therapeutic agent for diabetes mellitus and hemorrhoids in Eastern Anatolia. The plant is also used as a digestive and appetizer in Bitlis.

In the present study it was observed that the decoction extract of *Rheum ribes* roots possess significant blood-sugar lowering activity in alloxan-induced diabetic mice. However, this extract did not show hypoglycemic action in healthy mice. The hypoglycemic activity of the decoction extract was compared to those of glibenclamide and isotonic saline solution in alloxan induced diabetic mice.

This study has shown that *Rheum ribes* roots decoction extract is more potent than glibenclamide during the first hour following the application. The hypoglycemic effects of the extract and glibenclamide were equal after the second hour of the study and the fourth hour the hypoglycemic effect

Table 1.
Effect of *Rheum ribes* roots extract on blood glucose concentration in alloxan-induced diabetic mice *(n=10)*.

Group	Study	Fasting serum glucose levels (mg/dL)				
		0 h	1 h	2 h	4 h	24 h
I	Control (ISS)	337.2 ± 10.7	318.4 ± 11.6	308.0 ± 15.7	233.0 ± 13.2	205.4 ± 8.9
II	Glibenclamide	314.3 ± 21.4	266.6 ± 25.1	220.0 ± 17.5[a]	102.6 ± 1.9[c]	110.6 ± 3.1[c]
III	*Rheum ribes*	318.8 ± 22.5	198.8 ± 30.5[b]	222.8 ± 29.4[a]	165.6 ± 21.2[ad]	203.4 ± 17.1[e]
	F-value	*0.401*	*6.212*	*5.301*	*16.578*	*24.136*
	P value	*0.671*	*0.004*	*0.008*	*0.000*	*0.000*

The values represent the mean ± S.E.M.
[a] p<0.05 with respect to control group.
[b] p<0.01 with respect to control group.
[c] p<0.001 with respect to control group.
[d] p<0.05 with respect to glibenclamide group.
[e] p<0.001 with respect to glibenclamide group.

Table 2.
Effect of *Rheum ribes* roots extract on blood glucose concentration in healthy mice *(n=10)*.

Group	Study	Fasting serum glucose levels (mg/dL)				
		0 h	1 h	2 h	4 h	24 h
IV	Control (ISS)	91.5 ± 5.1	70.0 ± 3.7	59.2 ± 1.4	61.2 ± 1.6	51.5 ± 3.1
V	*Rheum ribes*	100.0 ± 03.5	72.2 ± 02.1	78.7 ± 3.8[a]	55.7 ± 2.9	52.5 ± 3.2

The values represent the mean ± S.E.M.
[a] p<0,001 with respect to control group.

of glibenclamide was higher than that of the *Rheum ribes* decoction extract.
The present study indicates that the extract of this plant should be studied further.

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