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STUDIES ON THE EFFECT OF *ACACIA ARABICA* FRUITS (KIKAR) AND *CARALLUMA EDULIS* ROOTS (CHUNG) ON BLOOD GLUCOSE LEVELS IN NORMAL AND ALLOXAN-DIABETIC RABBITS

*Muhammad Shoaib Akhtar and Qaiser Mahmood Khan

Blood glucose levels of the normal and alloxan-diabetic male albino rabbits were determined after oral administration of various doses of the powdered fruits of the *Acacia arabica*, Willd. (Fam. Leguminosae) and roots of *Caralluma edulis*, Benth. (Fam. Asclepiadaceae). Powdered *A. arabica* fruits produce significant hypoglycaemic effect only in the normal rabbits whereas *C. edulis* did not produce this effect. In the alloxan-treated rabbits both the medicinal plant drugs did not affect the blood glucose levels. Moreover, acute toxicity studies and record of behavioural patterns carried out in rabbits and rats, respectively showed no adverse effects of *A. arabica* fruits in the dosages tested. It is conceivable that *A. arabica* fruits contain some hypoglycaemic principle(s) which act probably by initiating the release of insulin from pancreatic beta-cells of normal rabbits.

INTRODUCTION

Since centuries the plants have been used as source of drugs. It is not surprising, therefore that some of them have also been used to treat diabetes. The plants of *Acacia arabica* and *Caralluma edulis* locally known as Kikar and Chung, respectively have been used in traditional medines for various purposes, e. g. for the treatment of diabetes, liver diseases, digestive troubles and some metabolic disorders (Chopra *et al.*, 1956; Lewis and Elvin-Lewis, 1977 and Ikram, 1981). The present investigations were taken to study the effect of powdered *Acacia arabica* and *Caralluma edulis* plants on blood glucose levels following oral administration to normoglycaemic and alloxan-treated hyperglycaemic male albino rabbits. Acute toxicity studies and record of behavioural patterns were also carried out in the animals to determine the safety of these folk medicines.

*Department of Physiology and Pharmacology, University of Agriculture, Faisalabad.

MATERIALS AND METHODS

Plant Materials : Fresh green fruits of *Acacia arabica* and roots of *Caralluma edulis* plants locally known as Kikar and Chung, respectively, were used in this study. The fruit of *Acacia arabica* was collected from the trees in and around the Agriculture University Campus, while *Caralluma edulis* was purchased from dlocal market of Faisalabad. These were carefully washed with tap water and dried under shade. The dried plants were milled in an electric grinder an stored in well-closed cellophane bags at 4°C in the refrigerator.

Chemicals Used : Alloxan-monohydrate and α-d-glucose (anhydrous) were obtained from B. D. H., Laboratories (Chemical Division), Poole, England; glacial acetic acid, benzoic acid (sublimed), O-toluidine, thiourea and trichloracetic acid were obtained from E. Merck; Darmstadt, West Germany. Gum tragacanth was purchased from the local market. Tolbutamide (Rastinon) was obtained from Hoechst (Pakistan) Ltd , Karachi.

Animals Used : Male, adult, healthy Albino rabbits of a local strain weighing between 1000-1200 g were used in these experiments. The animals were kept in an air-conditioned animal room of the Physiology and Pharmacology Department at the University of Agriculture, Faisalabad. The animals were offered a balanced rabbit feed prepared by the Animal Nutrition Department of this University and allowed tap water *ad libitum*. The effects of *Acacia arabica* and *Caralluma e dulis* were studied on blood glucose levels of the normal rabbits. In addition, separate experiments were performed to study their effects on blood glucose level of diabetic rabbits. For actue toxicity studies and behavioural pattern recordings, albino rabbits and Sprague-Dawley rats of either sex were used.

Preparation of Diabetic Rabbits : A group of rabbits were made diabetic by injecting intravenously 150 mg/kg body weight of alloxan-monohydrate (Akhtar *et al.*, 1981). Eight days after injection, the blood glucose levels of all surviving rabbits were determined by the O-toluidine method. Rabbits with blood glucose level of 250-500 mg/100ml were considered as diabetic and were employed for further study as already used by Akhtar *et al.* (1981 & 1984) in their experiments.

Grouping of Rabbits : Rabbits were randomly divided in 10 groups (I-X) of six animals each. Group I served as control and received orally 15 ml of 20% gum

tragacanth solution in water only. The animals of groups II, III and IV were treated orally with 2, 3 and 4 g/kg body weight of *A. arabica* powder suspended in 2% gum solution, respectively. Group V was treated with tolbutamide, 500mg/kg body weight. Animals of groups VI to X were alloxan-diabetic. Out of theseVII, VIII and IX received orally 2, 3 and 4 g/kg body weight of *A. arabica* powder, respectively. The diabetic rabbits of X were treated with tolbutamide, 500 mg/kg body weight. Similar grouping was followed to study the effects of *C. edulis* (2, 3 and 4 g/kg body weight) on blood glucose of normal and diabetic rabbits.

Preparation and Administration of Drug Suspensions : *A. arabica* and *C. edulis* powers were administered on body weight basis after suspending in 10 ml of 2% gum tragacanth solution. Final volume was made up to 15 ml. They were administered to each animal by using a stainless steel feeding needle attached to a syring. The needle was inserted into the stomach through the oesophagus and the plunger was pressed slowly and steadily. Immediate sneezing and coughing indicated injection into the lungs and in such a condition the needle was at once withdrawn and another animal was taken instead.

Collection of Blood : After administration, the animal was held in a wooden rabbit holder and immediately 0.2 ml of blood was collected from an ear vein. Similar samples of 0.2 ml were collected at 4, 8, 10, 12 and 24 hour time intervals. After collecting blood, absolute alcohol was applied on the pricked site to protect the ear against infection.

Determination of Blood Glucose : Blood glucose was dertermined by the method of Fings *et al.* (1970) using O-toluidine reagent. This method is very sensitive and can detect even small amounts of blood glucose and the results obtained are close to those obtained by the glucose oxidase method. The detailed methodology has already been reported by Akhtar *et al.* (1981).

Acute Toxicity and Behavioural Pattern Study : In order to study any possible toxic effect or change in normal behavioural pattern of animals, different doses of *A. arabica* fruit were orally administered to groups of 6 male albino rabbits or Sprague - Dawley rats. The symptoms including awareness, mood, motor activity, CNS excitation, posture, motor incoordination, muscle tone, reflexes, autonomic responses, etc. as described in detail by Laurence and Bacharach (1964) were checked. Animals were closely observed for 6 to 8 hours after dosage for any of the toxic symptoms and for possible mortality. They were kept under observation for 7 days.

Statistical Analysis : Mean blood glucose levels were expressed in mg/100ml ± SEM in all experiments and Student's 't' test was used to check their significance.

RESULTS

Standard Curve for Glucose Determination : Standard curve for Glucose estimation was linear up to 300 mg/100 ml and blood glucose levels of the samples having more than 300 mg/100ml were re-determined after diluiton.

Effect of Alloxan Administration to Rabbits : These effects have already been described in detail by Akhtar *et al.* (1981 & 1984). The rabbits with blood glucose levels above 200 mg percent were considered diabetic and were used in these studies. Such hyperglycaemic rabbits have already been employed by Marquis *et al.* (1977) and Akhtar *et al.* (1981 & 84).

EFFECTS OF *ACACIA ARABICA*

Effect of A. arabica on Blood Glucose in Normal Rabbits : The mean blood glucose concentration of different doses of *A. arabica* powder at various time intervals are shown in Figure 1. Gum tragacanth did not change the blood glucose levels of rabbits. Administration of 2 g/kg of *A. arabica* powder produced a significant ($P < 0.05$) decrease in blood glucose at 10 and 24 hour intervals. The animals treated with 3 g/kg of *A. arabica* showed a significant decrease at 4, 10 and 24 hours. The blood glucose of rabbits treated with 4 g/kg body weight of drug was found to be significantly reduced ($P < 0.05$) at 4 and 10 hours. The decrease in glucose level at 24 hours was significant ($P < 0.001$). Maximum decreasein blood glucose levels at all doses of *A. arabica* powder in normal animal was produced at 24 hours.

Effect of A. arabica Powder on Blood Glucose in Diabetic Rabbits : Administration of 2% gum solution to the diabetic rabbits did not alter their blood glucose levels at 0, 4, 10 and 24 hours. Similary, blood glucose levels of animals treated with 2, 3 and 4 g/kg body weight of *A. arabic* powder did not produce any significant change in glucose levels at any interval after drug administration.

EFFECTS OF *CARALLUMA EDULIS*

Effect of C. edulis on Blood Glucose Levels in Normal Rabbits : The 2% gum solution did not significantly effect the glucose levels of rabbits. Administration of 2, 3 and 4 g/kg body weight of *C. edulis* powder also did not significantly change the blood glucose levels at any interval.

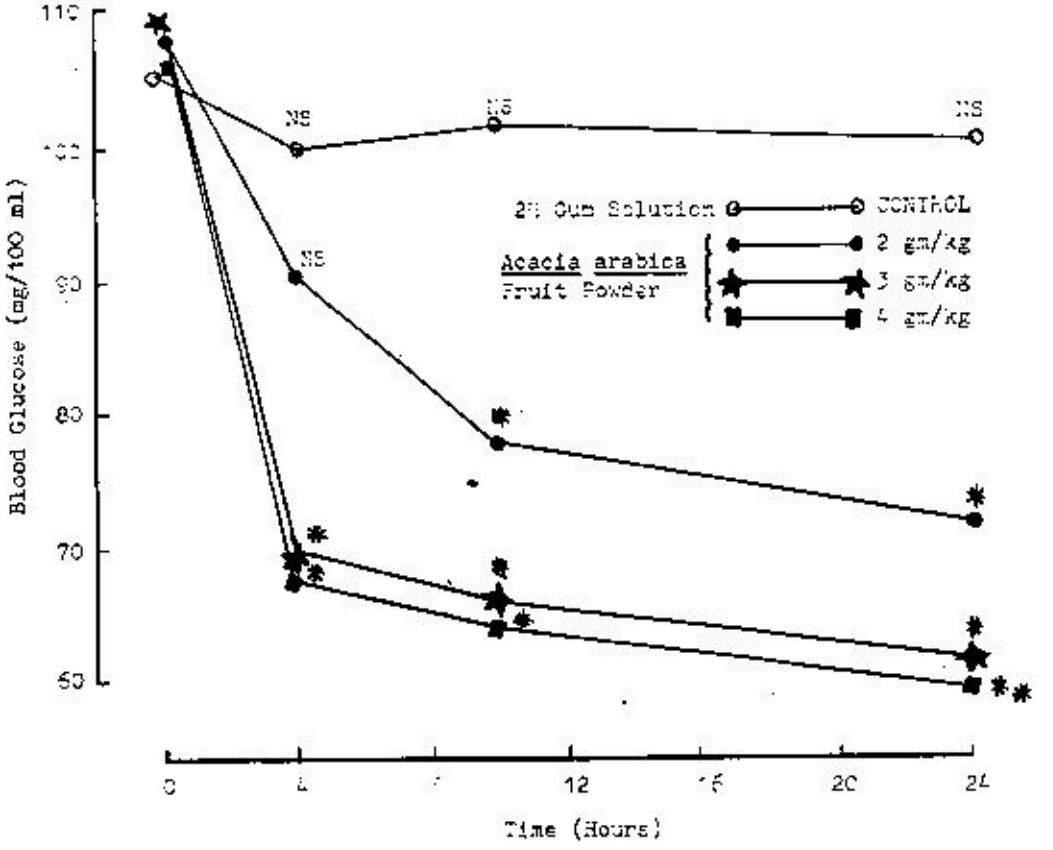

Fig. 1. Blood glucose levels of normal rabbits (mg/100 ml) at various time intervals after oral administration of 2% gum tragacanth solution and *Acacia arabica* powder (2, 3 and 4 g/kg) body weight orally suspended in 2% gum solution.

Effect of C. edulis Powder on Blood Glucose Levels in Diabetic Rabbits : Administration of 2% gum tragacanth solution did not alter the blood glucose levels of the diabetic rabbits. The blood glucose levels of animals treated with 2, 3 and 4 g/kg of *C. edulis* powder did not produce any significant ($P < 0.05$) change in blood glucose levels at any of the intervals.

Effects of Tolbutamide on Blood Glucose Levels in Normal and Diabetic Rabbits : The mean glucose concentration ± SEM of animals treated with tolbutamide (500 mg/kg orally) at zero hour interval was 94.5 ± 2.5 mg/100 ml and at 4, 8, 12 and 24 hours, the values were 68.5 ± 2.4, 72.3 ± 2.4 and 84.5 ± 2.4 and 91.0 ± 2.6 mg/100 ml, respectively. The values at 4, 8 and 12 hour intervals were found to be significantly lower than at zero hour. The decrease was maximum at 4 hour interval.

The mean blood glucose level at zero hour interval after administration of tolbutamide 500 mg/kg to diabetic rabbits was recorded to be 305.3 ± 10.1 mg/kg. The blood glucose levels at 4, 8, 12 and 24 hour intervals were 298.8 ± 11.4, 300.8 ± 11.6, 304.3 ± 10.0 and 304.7 + 11 2 mg/100 ml, respectively. None of these values were significantly different from zero hour level.

Acute Toxicity and Behavioural Pattern Studies : Rabbits treated with 4, 5, 6 and 7 g/kg body weight of the whole dried and powdered *A. arabica* plant remained alive upto 7 days and did not show any visible signs of acute toxicity, e. g. restlessness, respiratory distress, convulsions, coma, death, etc. Moreover the behavioural pattern studies in rats also revealed no prominent change in the awareness, mood, motor activity, CNS excitation, posture and motor incoordination, autonomic responses, food consumption and body weight.

DISCUSSION

The data obtained showed that 2% gum tragacanth solution did net produce any significant change in blood glucose of rabbits. However, as is obvious from fingure I, the administration of various doses of the *A. arabica* (Kikar) have caused a decrease in the blood glucose of normal rabbits. This crude drug produced a significant and consistent hypoglycaemic response in these rabbits. For comparison, tolbutamide (500 mg/kg) was administered orally to the normal rabbits. Similar to Augusti and Benaim (1975) and Akhtar *et al.* (1984), these doses of tolbutamide produced a significant decrease of blood glucose levels at 4, 8 and 12 hours. However, as is clear from Figure I the administration of *A. arabica* (4 g/kg) produces a significant fall in blood sugar even at 24 hours interval, showing that this plant drug possesses longer duration of action than tolbutamide which produces hypoglycaemia for 6 - 12 hours (Goth, 1981). Furthermore, calculations show that the maximum decrease in blood glucose produced by *A arabica* (4g/kg) is 90% of the maximum decrease produced by the 500 mg/kg of tolbutamide.

Sulphonylureas, e.g. tolbutamide produce hypoglycaemia by stimulating pancreatic beta-cells to release more insulin into the blood stream and by increasing glycogen deposition in the liver (Guyton, 1976). Thus it may be supposed that hypoglycaemic principle(s) in *A. arabica* plant also exert an hypoglycaemic effect in rabbits by initiating the release of insulin from pancreatic beta-cells. Similar mechanism has been proposed to explain the hypoglycaemic

effects of *Momordica charantia*, *Euphorbia prostrata* and *Fumaria parviflora* (Akhtar *et al.*, 1981 & 1984). Sulphonylureas, however, do not decrease blood glucose level in alloxan-diabetic animals. Therefore, to further explore the possible mechanisms of hypoglycaemic action of *A. arabica* plant, it was also administered to the alloxan-diabetic rabbits. Data obtained show that these drugs could not significantly lower blood glucose levels in these diabetic animals. Similarly, tolbutamide did not produce any significant change in the glucose levels of the alloxan-diabetic rabbits. It is, therefore, conceivable that hypoglycaemic principle(s) in *A. arabica* plant exert a hypoglycaemic effect in rabbits with intact pancreatic beta-cells by triggering insulin release. However, it remains to be established whether or not this plant drug actually acts by stimulating the secretion of insulin. Nevertheless, the results do suggest their active principle(s) do not seem to possess insulin-like activity. However, comprehensive chemical and pharmacological investigations are intended to be carried out to further elucidate the mechanisms of hypoglycaemic effect of *A. arabica* (Kikar) plant. Moreover, as the results have showed, the plant *Oxalis corniculata edulis* (Chung) did not produce any decrease in blood glucose level of both normal and diabetic rabbits. Thus the folk belief of referring this plant as antidiabetic was not confirmed by this study. The acute toxicity studies and behavioural pattern records have indicated that *A. arabica* fruits do not produce acute toxicity symptoms at the dosages employed. Virtually, no adverse effects were observed in the treated animals and no change was observed in the normal behavioural pattern of the tested animals.

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