EFFECT OF ERIOBOTRYA JAPONICA ON BLOOD GLUCOSE LEVELS OF NORMAL AND ALLOXAN DIABETIC RABBITS

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Abstract

The alcoholic extract of the leaves of Eriobotrya japonica was administered in doses of 100, 150 and 200 mg/kg body weight to normal and alloxan diabetic rabbits. The blood glucose levels were estimated before and 1,2,3 and 4 hours after the administration of the extract. The extract exerted a significant ($P < 0.05$) hypoglycaemic effect in normal rabbits which was however short-lived. The hypoglycaemic effect was not significant ($P > 0.1$) in alloxan-treated rabbits. The doses used did not show acute toxicity and behavioural changes. From this study, it may be concluded that the extract acts by initiating the release of insulin from pancreative beta cells of normal rabbits (JPMA 38:108, 1988).

INTRODUCFION

Eriobotrya japonica is a small tree belonging to the family rosaecae. It is commonly known as Lokat, and its leaves are used by practitioners of Unam system of Medicine for the treatment of diabetes mellitus[1]. The present work has been undertaken with the aim to study the effect of alcoholic extract of the leaves of Eriobotrya japonica on the blood glucose levels of normal and alloxan treated diabetic male rabbits. Acute toxicity and behavioural changes were also studied to check the safety of these doses.

MATERIAL AND METHODS

Experiments were performed on male, adult rabbits of local strain weighing 0.52—1.65 kg. They were fed on green vegetables and grains and allowed tap-water ad libitum.

Chemicals:

Alloxan monohydrate was supplied by B.D.H. Laboratories (Chemical Division), Poole, England. o. toluidine, Glacial acetic acid, Thiourea and Trichioroacetic acid were obtained from E. Merck, Darmstadt, West Germany. Tolbutamide was provided by Hoechst, West Germany.

Preparation of Extract:

Alcoholic extract was prepared by the method described by Noreen et al2. The fresh leaves obtained from local trees were dried under the shade and completely dried leaves were powdered with an electric grinder. The powder was extracted for 8 hours with petroleum ether to give a brown residue. Continued extraction with 95% alcohol gave a clear solution. Subsequently it was evaporated, then partitioned between water and chloroform ethanol (2:1) and then again evaporated under reduced pressure to obtain the desired material.

Preparation of Diabetic Rabbits:

The method described by Akhtar et al3 was adopted. A group of rabbits was made diabetic by injecting intravenously 150 mg/kg body weight of alloxanmonohydrate. Eight days after injection, the blood glucose levels of surviving rabbits were estimated. Rabbits with blood glucose levels above 200

mg/100 ml were considered as diabetic.

Grouping of Rabbits:-

Normal rabbits were divided into 5 groups (1-5) of six animals each. Group 1 served as control and received 15 ml of water only. Group 2 received tolbutamide 500mg/kg body weight. Group 3-5 received extract of Eriobotrya japonica dissolved in 15 ml of water in doses of 100 mg/kg, 150 mg/kg and 200 mg/kg respectively. The diabetic rabbits were also divided in 5 groups on the same pattern.

Preparation and Administration of Extract Solution:-

The amount of Eriobotrya japonica extract required for each rabbit was calculated on body weight basis and it was dissolved in 10 ml of water to make a colloidal solution and final volume made upto 15 ml. The extract was administeTed to each rabbit by using a stomach tube attached to a standard syringe containing the 15ml of extract solution. The tube was inserted into the stomach through oesophagus and the plunger was pressed slowly and steadily. Immediate sneezing and coughing indicated injection into the lungs and in such a condition the tube was at once withdrawn and another animal was taken instead. The tolbutamide solution was administered in a similar manner.

Collection of Blood:—

The procedure for collection of blood was adopted as described by Akhtar et al[4]. The rabbit was held in a wooden rabbit holder and, immediately before administration of drug, 0.2 ml of blood for glucose estimation was collected from an ear vein. Similar blood samples were also collected at 1 ,2,3 and 4 hours after the drug administration. After collection of blood, the pricked site of the ear was rubbed with cotton wool soaked with 70% alcohol to protect the rabbit against infection.

Blood Glucose Estimation: —

Blood glucose estimation was done by the
method of Winckers and Jacobs.[5]

To Study Acute Toxicity of the Extract:—

The possible toxic effects of the extract were studied in rabbits of local strains weighing between 0.50 —1.7 kg. The rabbits were divided into 4 groups (1—4) of six animals each. Group 1 served as control and received 15 ml of water only. Group 2—4 received extract of Eriobotrya japonica dissolved in 1 5 ml of water in doses of 100 mg/kg, 150 mg/kg and 200 mg/kg, respectively. Animals were observed for 8 hours after administration of the extract to check toxic symptoms. They were kept under observation for 7 days.

RESULTS

The effect of different doses of the extract of Eriobotrya japonica and tolbutamide on blood glucose levels of normal and alloxan diabetic rabbits are shown in

Table 1. Effect of Tolbutamide and Eriobotrya Japonica Extract on Blood Glucose Level of Normal and Alloxan – Diabetic Rabbits.

Time Interval (Hours)	Blood Glucose in mg/100 ml.									
	Normal Rabbits					Alloxan Diabetic Rabbits				
	Control	Tolbu-Tamide (500 mg/kg)	Eriobotria Japonica 100 mg/kg	150 mg/kg	200 mg/kg	Control	Tolbutamide (500 mg/kg)	Eriobotrya Japonica 100 mg/kg	150 mg/kg	200 mg/kg
0	92.6±1.41 (6)	90.7±2.05 (6)	88.7±5.6 (6)	91.9±5.48 (6)	97.5±4.6 (6)	383.5±4.60 (6)	374.4±3.13 (6)	239.8±7.33 (6)	377.8±4.9 (6)	372.5±5.71 (6)
1	92.3±1.43 (6)	82.0±1.64 (6)	65.7±3.6 (6)	63.7±3.1 (6)	60.60±2.2 (6)	377.5±4.64 (6)	369.9±3.29 (6)	237.3±6.91 (60	375.0±4.86 (6)	362.5±4.89 (6)
2	93.2±1.50 (6)	76.2±1.54 (6)	63.9±3.5 (6)	59.7±2.8 (6)	50.0±1.71 (6)	378.0±5.14 (6)	367.0±2.79 (6)	234.6±7.06 (6)	373.2±5.16 (6)	360.1±5.20 (6)
3	92.2±1.42 (6)	75.3±1.39 (6)	57.6±3.07 (6)	47.3±2.14 (6)	38.3±1.16 (6)	378.0±5.03 (6)	369.5±3.35 (6)	231.7±5.90 (6)	370.1±4.56 (6)	358.2±5.94 (6)
4	92.2±1.40 (6)	71.4±1.21 (6)	85.2±4.8 (6)	87.6±4.5 (6)	92.9±3.75 (6)	378.0±1.44 (6)	370.0±3.0 (6)	235.0±7.32 (6)	371.8±5.60 (6)	361.0±5.2 (6)

Figures in parenthesis indicate number of animals
Each value represents the mean ±standard error.

Table II. Mean Percent Decrease in Blood Glucose by Tolbutamide and Eriobotrya Japonica Extract in Normal and Alloxan Diabetic Rabbits.

Time Interval (Hours)	Normal Rabbits					Alloxan – Diabetic Rabbits				
	Control	Tolbutamide 500 mg/kg	Eriobotrya Japonica 100 mg/kg	150 mg/kg	200 mg/kg	Control	Tolbutamide 500 mg/kg	Eriobotrya Japonica 100 mg/kg	150 mg/kg	200 mg/kg
1.	0.37±0.10 (6)	9.5±0.77 (6)	25.72±0.78 (6)	30.46±0.80 (6)	37.51±0.76 (6)	1.56±0.10 (6)	1.19±0.10 (6)	1.01±0.18 (6)	0.73±0.08 (6)	2.7±0.22 (6)
2.	0.48±0.10 (6)	15.7±0.80 (6)	27.76±0.92 (6)	34.80±0.85 (6)	48.52±0.78 (6)	1.57±0.20 (6)	1.95±0.16 (6)	2.14±0.25 (6)	1.21±0.45 (6)	3.3±0.15 (6)
3.	0.41±0.09 (6)	17.0±0.79 (6)	34.87±0.77 (6)	48.25±0.78 (6)	60.52±0.74 (6)	1.39±0.25 (6)	1.28±0.18 (6)	3.29±0.61 (6)	0.39±0.16 (6)	3.85±0.14 (6)
4.	0.44±0.10 (6)	21.1±0.84 (6)	3.67±2.03 (6)	4.43±0.81 (6)	4.52±0.75 (6)	1.44±0.22 (6)	1.32±0.14 (6)	1.95±0.21 (6)	1.59±0.23 (6)	3.07±0.21 (6)

Figures in parenthesis indicate number of animals.
Each value represents the mean ±standard error.

Table I-II and Figures 1-4.

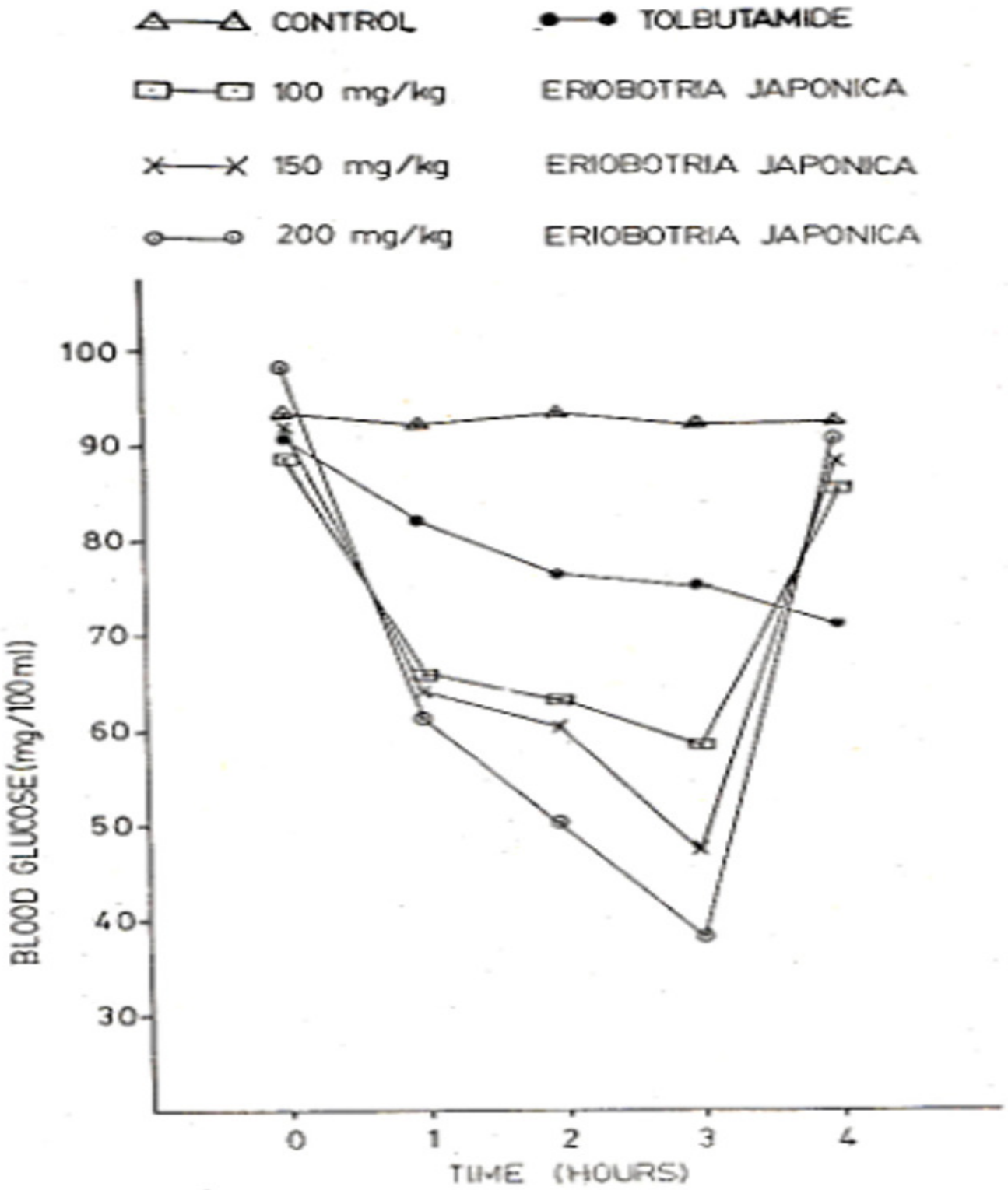

Figure 1. Effect of tolbutamide and eriobotria-japonica on Blood glucose level of normal rabbits.

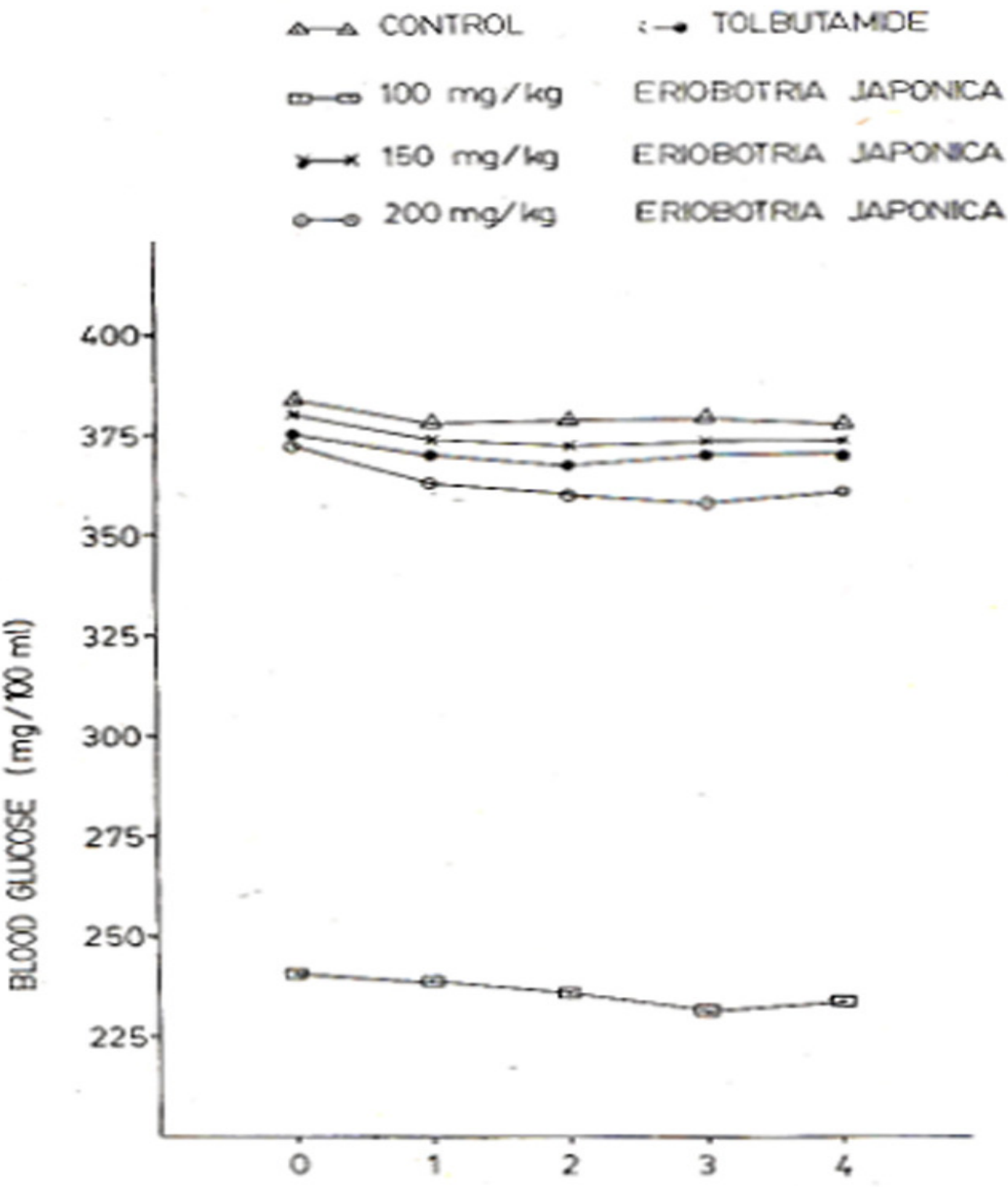

Figure 2. Effect of tolbutamide and eriobotria Japonica on blood glucose level of alloxan diabetic rabbits.

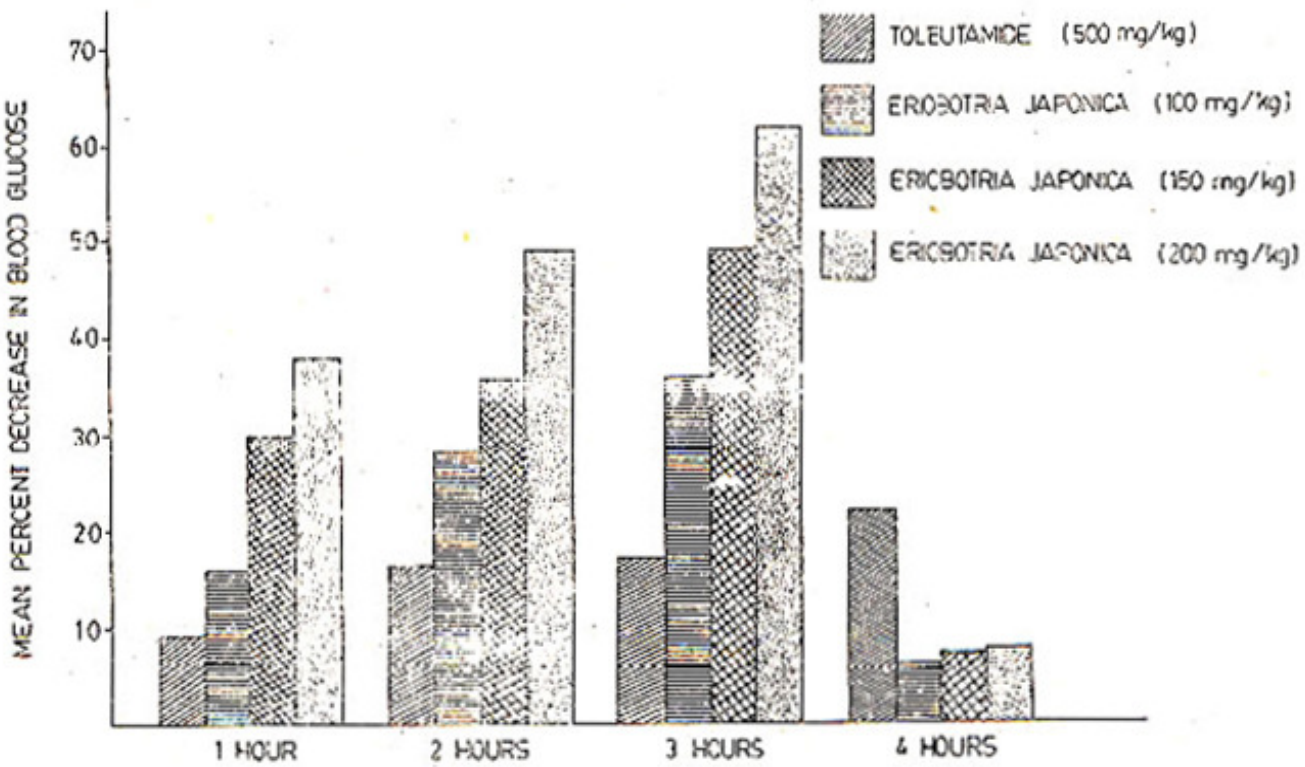

Figure 3. Effect of tolbutamide and eriobotria japonica on blood glucose level of normal rabbits.

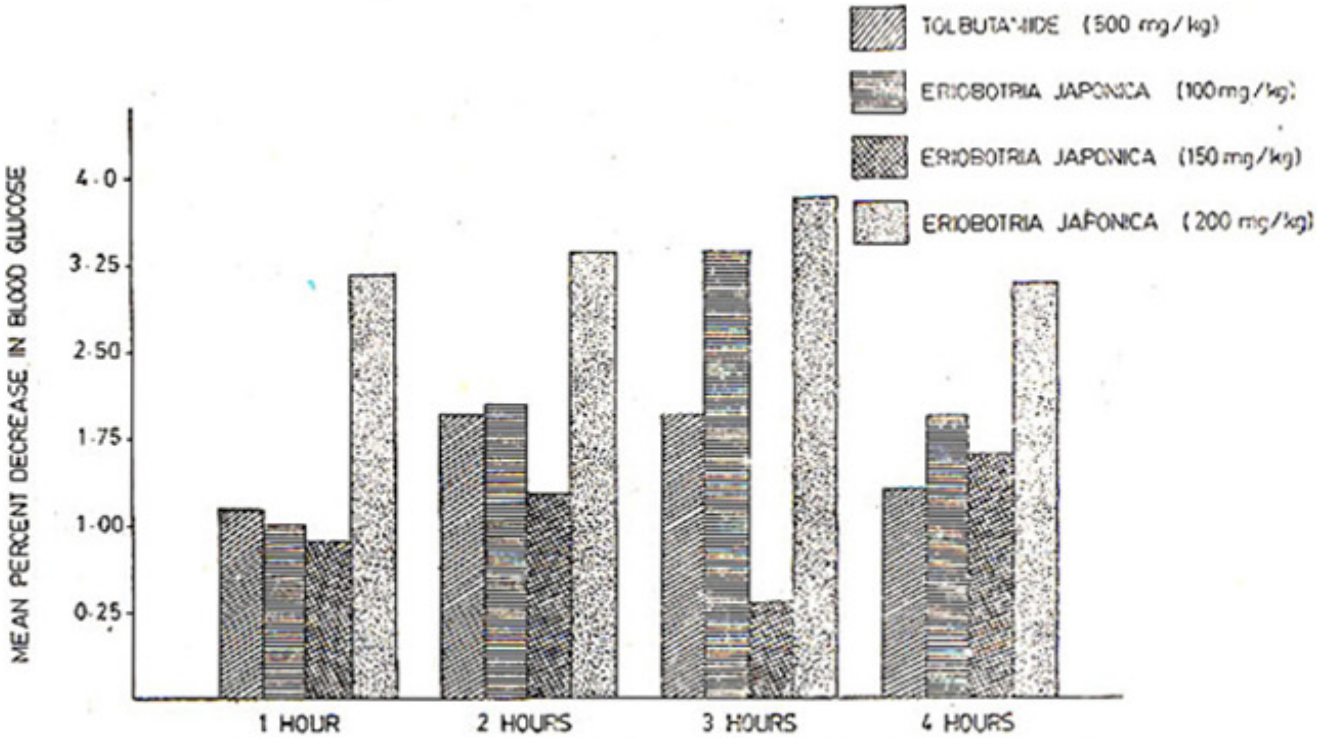

Figure 4. Effect of tolbutamide and eriobotria japonica on blood glucose level of alloxan diabetic rabbits.

The observations show that water used as control in these experiments did not produce any significant

change (P > 0.10) on blood glucose levels of normal as well as on that of alloxan diabetic rabbits.
Effect of Eriobotrya Japonica Extract on Blood Glucose Levels in Normal Rabbits.
The mean percent decreases in blood glucose levels produced by 100 mg/kg of Eriobotrya japonica at 1 ,2 ,3 and 4 hours were 25.72 ± 018, 27.76 ± 0.92, 34.87 ± 0.77 and 3.67 ± 2.03, respectively. The mean percent decreases are significant (P <0.05) at 1,2 and 3 hours, while not significant (P>0.lO) at 4 hours. The mean percent decreases in blood glucose levels produced by 150 mg/kg of Eriobotrya japonica at 1,2,3 and 4 hours were 30.46 ± 0.80, 34.80 ± 0.85, 48.25 ± 018 and 4.43 ± 0.81 respectively. The mean percent decreases are significant (P < 0.05) at 1,2 and 3 hours, while non-significant (P > 0.10) at 4 hours. The mean percent decreases in blood glucose levels produced by 200 mg/kg of Eriobotrya japonica at 1 ,2,3 and 4 hours were 37.51 ± 0.76, 48.52 ± 0.78, 60.52 ± 0.74 and 4.52 ± 0.75, respectively. The mean percent decreases are significant (P <0.05) at 1,2, and 3 hours, while not significant (P> 0.10) at 4 hours.
Effect of Tolbutamide on Blood Glucose Levels In Normal Rabbits
The mean percent decreases in blood glucose levels produced by 500 mg/kg of tolbutamide at 1,2,3 and 4 hours were 9.5 ±017, 15.7 ± 0.80, 17.0 ± 019 and 21.1 ± 0.84, respectively, which are significant (P < 0.05) at 2, 3 and 4 hours.
Effect of Eriobotrya Japonica Extract on Blood Glucose Levels in Diabetic Rabbits.
The mean percent decreases in blood glucose levels produced by 100 mg/kg of Eriobotrya japonica at 1,2,3 and 4 hours were 1.01 ± 0.18, 2.14 ± 0.25, 3.29 ± 0.61, and 1.95 ± 0.21, respectively, which are non-significant (P >0.10). The mean percent decreases in blood glucose levels produced by 150 mg/kg of Eriobotrya japonica at 1,2,3 and 4 hours were 0.73± 0.08, 1.21 ± 0.45, 039 ± 0.16 and 1.59 ± 0.23,respectively, which are non-significant (P >0.10). The mean percent decreases in blood glucose levels produced by 200 mg/kg of Erióbotrya japonica at 1,2,3 and 4 hours were 2.7 ± 0.22, 3.3 ± 0.15, 3.85 ± 0.14 and 3.07 ± 0.21, respectively, which are non-signifidant (P> 0.10). Effect of Tolbutamide on Blood Glucose Levels ‘in Diabetic Rabbits.
The mean percent decreases in blood glucose levels produced by 500 mg/kg of tolbutamide at 1,2,3 and 4 hours were 1.19 ± 0.10, 1.95 ± 0.16, 1.28 ± 0.18, and 132 ±0.14, respeclively, which are not significant (P > 0.10). Acute Toxicity Study:-
The rabbits receiving 100 mg, 150 mg and 200 mg/kg body of the extract of Eriobotrya japonica did not show any visible signs of toxicity i.e. excitement, restlessness, respiratory distress, convulsions or coma. Moreover, they remained alive upto 7 days.

DISCUSSION

The present study revealed that alcoholic extract of Eriobotrya japonica produced a significant hypoglycaemic effect when administered orally to normal rabbits. This effect is short lived and lasts for only 3 hours after which the blood glucose level reverts to normal. The extract, however, did not show any significant effect on blood glucose levels of alloxan treated rabbits. For comparison, the effect of standard hypoglycadmic drug tolbutamide (500 mg/kg) was observed on the blood glucose levels of normal and alloxan treated diabetic rabbits. Tolbutamide produced significant hypoglycaemic effects in normal rabbits but not in alloxan treated diabetic rabbits. This finding is in accordance with the observation of Augusti and Benaim[6] and Akhtar et al.[7] Sulphonylureas including tolbutamide have been reported to produce hypoglycaemia by stimulating pancreatic beta cells to release more insulin into the blood stream, increasing glycogen deposition in the liver, causing reduction of glucagon levels and having an extrapancreatic effect to possibly increase the number of insulin receptors.[8] In view of the similarity between the effects of tolbutamide and Eriobotrya japonica, it may be likely that the hypoglycaemic effect of Eriobotrya japonica may also be mediated through the release of insulin from the pancreatic beta cells. Similar mechanism has also been proposed to explain the hypoglycaemic

effect in normal rabbits of other indigenous plants such as tecoma stans[9], Momordica foetida[10], Momordica Charantia[3], Euphorbia prostrata and Fumaria Parviflora[7], Bergenia lingulata[11] and Taraxacum officinale[12]. However, it remains to be established the short duration of action of the extract.

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