e-ISSN:2322-0139
p-ISSN:2322-0120

RESEARCH AND REVIEWS: JOURNAL OF PHARMACOLOGY AND TOXICOLOGICAL STUDIES

Comparative Hypoglycemic Effects of Different Extract of *Clitoria Ternatea* Leaves.

Abhishek Kumar Saxena*, Saxena Vikas, Mishra Pankaj, and Kumar Amit.

Department of Pharmcology, Rakshpal Bahadur College of Pharmacy, Bareilly- 243001, Uttar Pradesh, India.

Research Article

Received: 03/03/2014
Revised: 27/04/2014
Accepted: 04/05/2014

***For Correspondence**

Department of Pharmcology, Rakshpal Bahadur College of Pharmacy, Bareilly- 243001, Uttar Pradesh, India.

Keywords: Hypoglycemic, *Clitoria ternatea,* antidiabetic activity.

ABSTRACT

We evaluated the hypoglycemic effects of methanol, water, petroleum ether, chloroform extract of *Clitoria ternatea* leaves. The hypoglycemic effect was evaluated in Streptozotocin induced diabetic rats for acute and sub acute effects. The extract of *Clitoria ternatea* also significantly ($P<0.001$) reduced blood glucose level in Streptozotocin induced diabetic rats twelve hours after administration.

INTRODUCTION

Diabetes mellitus (DM) is a widespread disorder, which has long been recognized in the history of medicine, before the advent of insulin and oral hypoglycemic drugs, the major form of treatment involved the use of paints. More than 400 plants are known to have been recommended ad recent investigations have affirmed the potential value of some of these treatments [1].

Diabetes mellitus is a group of endocrine syndromes characterized by hyperglycemia; altered metabolism of lipids, carbohydrates, and proteins, and an increased risk of complications from vascular disease. Most patients can be classified clinically as having either type I diabetes mellitus (type I DM formerly known as insulin dependent diabetes of IDDM) or type II diabetes mellitus (type II DM formerly known as non-insulin dependent diabetes of NIDDM) [2].

A good-looking perennial twining herb with terete stems and branches, leaves compound, imparipinnate, leaflets 5-7, sub- coriaceous, elliptic- oblong, obtuse; flowers blue or white , solitary axillary or in fascicles, corolla papilionacaous; fruits nearly straight, Flattened pods, Sharply beaked; seeds 6-10, smooth, yellowish brown. *Clitoria ternatea* is used as aphrodisiac tonic and are useful in ophthalmopathy. The leaves are useful in otalgia, hepatopathy and eruptions. The root also has anti-inflammatory, analgesic and antipyretic properties. *Clitoria ternatea* is used in leucoderma, burning sensation and pains. The roots are used as bitter, refrigerant, ophthalmic and laxative [3,4,5].

MATERIALS AND METHODS

Plant material

The fresh leaves of *Clitoria ternatea* was collected during the month of September 2011, from the pratap Nursury, karamchari nagar Bareilly. The plant materials was taxonomically identified and authenticated by Dr. Umesh Chand Pandey, HOD and in charge Botany Department, Bareilly college, Bareilly (BCB/BOT/376/24-01-2012).

e-ISSN:2322-0139
p-ISSN:2322-0120

Preparation of Extract

The leaves of *Clitoria ternatea* were shaded dried until cracking sound was observed during breakage, and then these are made into coarsely powdered from using dry grinder. The powder leaves of the plant (500 gm) was macerated with each different solvents methanol, water, chloroform, petroleum ether (1500 ml) at room temperature for 72 hours with occasionally stirring. The extracts were separated from the residues by filtering 1st through several layers of muslin cloth for coarse filtration and then through what man No. 1 filter paper. The residue was further extracted using the same procedure. The filtrates obtained were combined and then evaporated to dryness at temperature not exceeding 40^{0}C and then give moderate heating on water bath at temp 40±5^{0}C. The extracts were kept indifferent Petri dish and it was stored in refrigerator (-4c) at cool place till use. During experiment the crude extracts were diluted (100 mg of the extract was dissolved in 0.5 ml water) with distilled water just before administration to the animal [6,7,8].

Animals

Male swiss albino mice of body 150-200 gm weight were taken before and after experiment with the help of single pan balance were used for the study. The animals were housed in clean metabolic cages and maintained in controlled temperature (27± 2^{0}C) and light cycle (12 hrs. light and 12 hrs. dark). They were fed with standard pellet diet (Gold mohar brand, Lipton India Ltd.) and water. The protocol was approved by Institutional animal ethics committee 1452/po/a/11/cpcsea.

Streptozotocin

Streptozotocin (STZ) is a naturally occurring nitrosourea product of *Streptomyces achromogenes.* Usually, the intraperitoneal injection of a single dose (25 mg/kg body weight) of it exerts direct toxicity on β cells resulting in necrosis within 48-72 h and causes a permanent hyperglycemia. STZ breaks nuclear DNA strand of the islet cells [9].

Preparation of Dose

The Dose of 200 mg/kg and 400 mg/kg of methanol extract was selected for the test. All the doses was given orally after making emulsion in vehicle i.e. 1% acacia gum and the standard drug i.e. glibenclamide was given orally (10 mg/70kg) in the vehicle.

EXPERIMENTAL WORK

Effect of different extract on streptozotocin induced diabeticrats

Induction of diabetes

Streptozotocin manufactured by Sisco Research laboratories Pvt. Ltd. Mumbai, India and was freshly dissolved in 0.1 M citrate buffer (pH = 4.5) at the dose of 25 mg/kg body weight and injected intraperitoneally within 15 min of dissolution in a vehicle volume of 0.4 ml with 1 ml of tuberculin syringe fitted with 24 gauge needle, where as normal control group was given citrate buffer only (0.4 ml). Diabetes was confirmed by the determination of fasting glucose concentration on the third day post administration of streptozotocin [10].

Sample collection

Blood sample were collected from tail nipping and glucose level was determined by an automatic electronic glucometer (Accuchek comfort).

Procedure

After checking the fasting blood glucose in overnight fasted diabetic rats. They were divided into five groups of five rats each and one group of non-diabetic rats.

All the doses were given in the following manner [3]

- 1st Group- normal control group received vehicle.

e-ISSN:2322-0139
p-ISSN:2322-0120

- 2nd Group-diabetic control received vehicle.
- 3rd Group-Received methanol extract at dose of 200 mg/Kg orally.
- 4th Group- Received methanol extract at dose of 400 mg/Kg. orally.
- 5thGroup-Received water extract at dose of200 mg/kg orally.
- 6th Group- Received water extract at the dose of 400 mg/kg orally.
- 7th Group-Received chloroform extract at the dose of 200mg/kg orally.
- 8th Group Received chloroform extract at the dose of400 mg/kg orally.
- 9th Group Received petroleum extract at the dose 200 mg/kg orally
- 10th Group Received petroleum extract at the dose 400 mg/kg orally
- 11th Group- Received standard drug i.e. Glibenclamide (10 mg /Kg. in Vehicle) orally.

The treatment was continued for 4 hours. During the period water was supplied ad *libitum*. All the doses were administered orally by the oral feeding needle. The effect of extract on Blood glucose levels was estimated on overnight fasted rats on 0 hour, 30 min, 60min, 120min and 240min by the method described before. The general behaviors of the animals were recorded. The blood glucose level in (Mean ± SEM) is shown in the Table.

Effect of different extract on oral glucose tolerance test

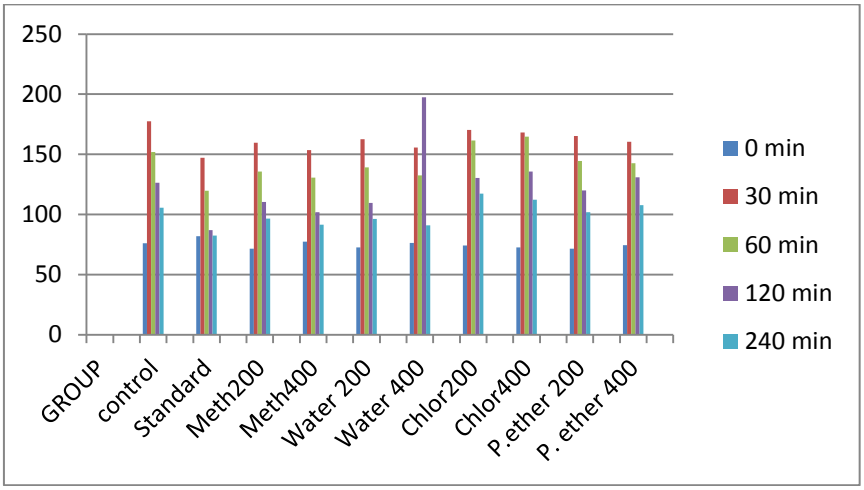
The hypoglycemic effect of extracts of *Clitoria ternatea* leaves was study on glucose loaded rats..

RESULTS AND DISCUSSION

GROUP	0 min	30 min	60 min	120 min	240 min
Control	75.92±2.21	177.50±4.38	151.89±3.54	126.32±3.61	105.67±2.76
Standard	81.85±2.52	147.01±2.00	119.81±2.86	86.97±3.03	82.34±2.13
Methanol 200	71.52±1.37	159.50±3.73	135.68 ±2.10	110.37±1.64	96.49±4.23
Methanol 400	77.30±3.07	153.40±2.52	130.73±2.38	101.74±1.60	91.54±3.29
Water 200	72.46±2.32	162.52±3.72	139.23±2.31	109.64±2.57	98.26± 3.13
Water 400	76.42±2.34	155.76±3.12	132.54±3.76	197.35±2.19	94.87± 2.31
Chloroform200	74.24±4.34	170.22±2.78	161.39±3.40	130.34±5.15	117.40± 3.73
Chloroform400	72.46±3.34	168.12±2.32	164.65±4.52	135.78±3.37	112.20± 3.82
Pet ether 200	71.44±3.37	165.34±2.49	144.53±3.32	119.98±3.45	101.87±2.56
Pet ether 400	74.47±5.90	160.41±2.89	142.65±4.52	130.78±3.27	107.63±4.40

Table 1: The Antihyperglycemic effect of different extracts on Glucose Loaded rats

The effect of extracts on the glucose loaded animal the results shows that methanol extract is most potent than the any other extract. While chloroform extract shows minimum potency the blood glucose level. Petroleum ether extract is better than the chloroform extract but it shows less activity than water extract and methanol extract.



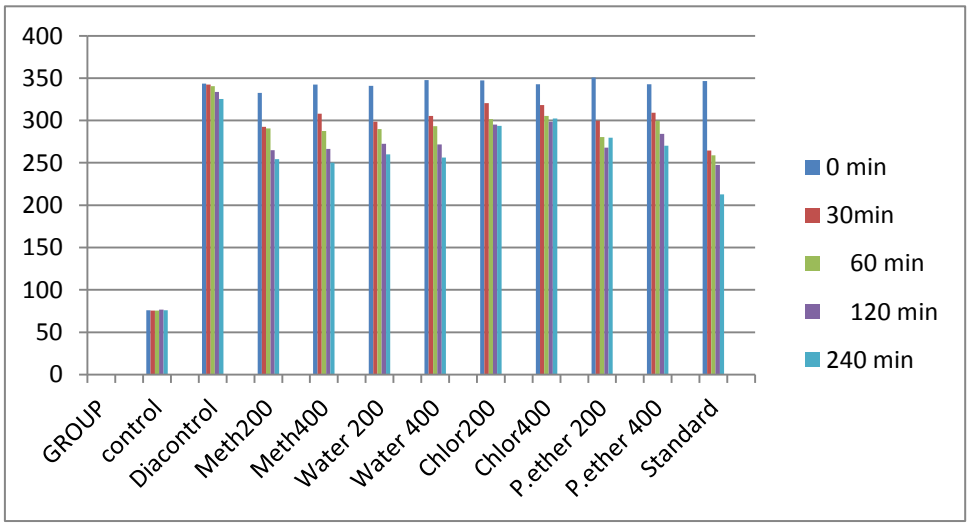
Graph 1: comparative effect of different extracts on glucose loaded rat

e-ISSN:2322-0139
p-ISSN:2322-0120

Table2: The comparative Antihyperglycemic effect of different extracts on STZ induced Diabetic rats

GROUP	Blood Glucose Level (mg/dl) at hr				
	0 min	30min	60 min	120 min	240 min
Control	75.75±3.93	75.67±2.75	75.56±2.20	76.63±1.59	76.06±1.48
Diabetic control	343.37±8.04	342.19±6.37	340.52±5.48	333.69±4.57	325.54±4.39
Methanol 200	332.67 ±3.51	292.56±3.72	290.48±3.56	264.92±2.23	254.19±3.40
Methanol 400	342.32±3.12	308.12±2.30	287.41±3.32	266.38±2.43	250.19±2.43
Water 200	340.82 ±4.51	298.45± 3.27	289.95±3.01	272.48±3.72	260.01±4.98
Water 400	347.52±4.92	305.34±1.78	293.11±2.76	271.52 ±2.48	256.19±2.50
Chloroform200	347.46 ± 3.567	320.43±4.19	301.63 ±4.76	295.01±3.70	293.72±2.8
Chloroform400	342.82 ±3.62	318.20 ±4.45	305.30 ±3.34	298.38±2.60	302.41±3,62
Pet ether 200	350.72 ± 2.51	299.65 ±3.21	280.50 ±2.52	268.01±3.98	279.45±5.4
Pet ether 400	342.82 ±3.62	309.11±3.12	299.52±2.98	284.19±3.160	270.32±2.8
Standard	346.35±4.28	264.47± 3.16	258.90±2.51	247.46 ±2.77	212.67±2.36

Diabetic control shows highest blood glucose level .on the other hand standard drug shows the best result among the all extract. But methanol dose 400mg/kg shows prominent result. Chloroform shows minimum effect on the blood glucose level. In the 200mg/kg methanol also shows good effect but these was not good than the 400mg/kg of the methanol extract.



Graph 2: The comparative Antihyperglycemic effect of different extracts on STZ induced Diabetic rats

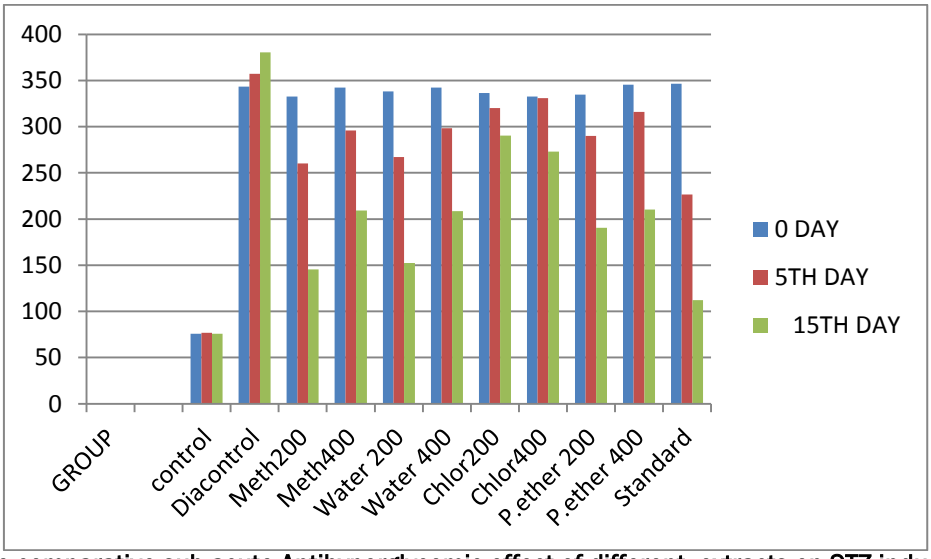
Table 3: The comparative sub-acute Antihyperglycemic effect of different extracts on STZ induced Diabetic rats

GROUP	Blood Glucose Level (mg/dl) at hr		
	0 DAY	5TH DAY	15TH DAY
Control	75.75 ± 3.93	76.76 ± 3.5	75.80 ± 4.87
Diabetic control	343.37 ± 8.04	357.27 ± 7.54	380.56 ± 2.76
Methanol 200	332.67 ± 3.51	260 ± 4.8	145.46 ± 4.63
Methanol 400	342.32 ± 3.12	295.90 ± 7.28	209.32 ± 7.36
Water 200	338.30 ± 6.48	267 ± 6.3	152.27 ± 7.68
Water 400	342.32 ± 3.12	298.12 ± 5.19	208.52 ± 4.48
Chloroform200	336.36 ± 3.67	320 ± 3.54	290.48 ± 4.65
Chloroform400	332.75 ± 3.67	331 ± 3.34	272.90 ± 3.52
Pet ether 200	334.56 ± 4.90	290 ± 3.54	190.38 ± 3.28
Pet ether 400	345.53 ± 4.35	316 ± 4.78	210.32 ± 5.76
Standard	346.35 ± 4.28	226.53 ± 7.9	112.32 ± 46

The result of the sub acute activity is much differs than the acute study this result shows that on the long term use of extract the dose 200mg/kg is much better to control the blood glucose level tan the 400mg/kg dose. After the 15 days the methanol extract of 200mg/kg dose shows minimum blood glucose level than other extract. In the all extract, on long term use 200mg/kg has a better result than the

e-ISSN:2322-0139
p-ISSN:2322-0120

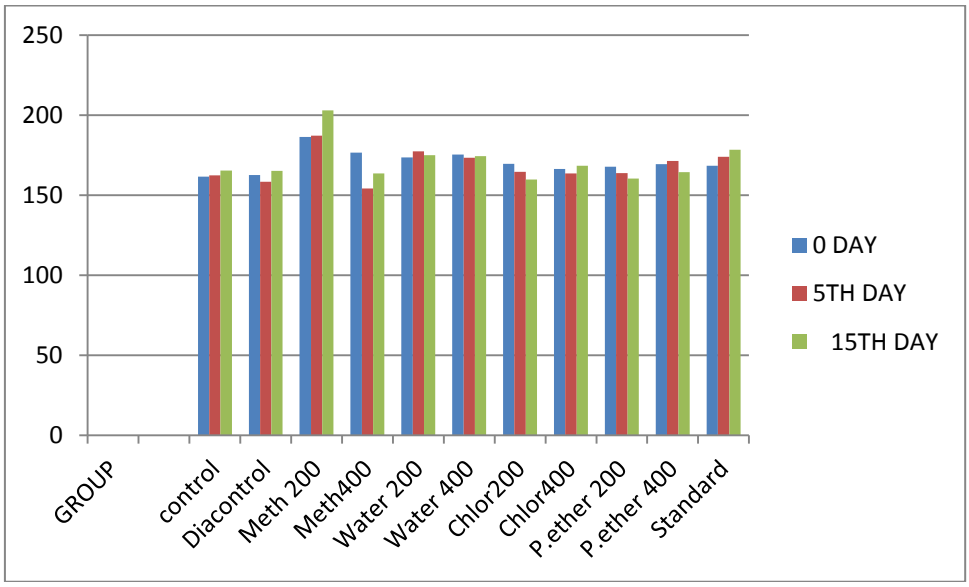
400mg/kg. but the effect were same chloroform extract has minimum effect .petroleum ether extract is better effect than chloroform extract but poor effect than the methanol extract and water extract.



Graph 3: The comparative sub-acute Antihyperglycemic effect of different extracts on STZ induced Diabetic rats.

Table 4: The comparative sub-acute effect of different extracts on body weight of rats

GROUP	Body weight in gram		
	0 DAY	5TH DAY	15TH DAY
Control	161.46± 3.21	162.37± 4.21	165.44± 4.55
Diabetic Control	162.53± 3.54	158.40± 3.4	165.17± 5.4
Methanol 200	186.3± 4.78	192.2± 5.62	203.0± 3.97
Methanol 400	176.54± 5.5	154.2± 8.18	163.50 ± 3.8
Water 200	173.43 ± 3.94	177.35± 4.52	175.0±2.50
Water 400	175.25 ±4.25	173.42±6.21	174.35±3.4
Chloroform200	169.43+±3.16	165.45±3.53	162.78±4.64
Chloroform400	166.32±2.12	163.63±6.72	168.34±3.42
Pet ether 200	167.7± 5.13	163.65± 4.90	160.32± 6.32
Pet ether 400	169.34± 3.54	171.24± 4.89	164.42± 4.51
Standard	168.35±4.83	173.92± 3.79	178.40± 5.5



Graph 4

e-ISSN:2322-0139
p-ISSN:2322-0120

The effect of the standard drug and different extract the methanol extract of 200mg/kg dose showed 15 gm in the fifteen days and standard drug showed 10 gm weight gain in the animal. But in the case of chloroform extract there is something different because it showed 7gm weight loss in the 200mg/kg dose. Water 400mg/kg, 200mg/kg showed within 3gm ± weight difference in the animal. Petroleum ether 400mg/kg also showed 7gm weight loss in the animal during 15 days study. Diabetic control group showed 3gm weight gain in the animals.

REFERENCES

1. Baily J, Day C. Traditional Plant Medicines as Treatments for Diabetes. Diabetic Care. 1989;12:533.
2. Goodman and Gillman, 2001 "The Pharmacological basis of therapeutics", Mcgrow hills publication,11, 1679-1714.
3. Nagappa AN, Thakurdesai PA, Rao NV, Singh J. Antidiabetic activity of *Terminalia catappa Linn* fruits. J Ethanopharmacol. 2003, 88, 45-50.
4. The Wealth of India, 2003. A Dictionary of Indian Raw Material and Industrial product, 6, 207-216.
5. Database on Medicinal Plants used in Ayurveda Vol-1, 22.
6. Sarojini Nayak et al. Anthelmintic activity study of saraca indica leaves. International Journal of Applied Biology and Pharmaceutical Technology. 2011;2(2):377-79
7. Barkat Ali Khan et al. Investigation of the effects of extraction solvent/technique on the antioxidant activity of Cassia fistula L. J Med Plants Res. 2012;6(3):500-503.
8. Romila Y,et al, A Review on Antidiabetic Plants used by the People of Manipur Charactirized by Hypoglycemic Activity. Assam University Journal of Science & Technology: Biological and Environmental Sciences. 2010;6:167-175.
9. Sharmistha Chakravarty et al, Antihyperglycaemic effect of flower of Phlogacanthus Thyrsiflorus Nees on streptozotocin induced diabetic mice. Asian Pacific J Trop Biomed. 2012;S1357-S1361
10. Liliane Sena Pinheiro et al, Protocol of Insulin Therapy For Streptozotocin-Diabetic Rats Based on a Study of Food Ingestion and Glycemic Variation. Scand J Lab Anim Sci. 2011;38(2).
11. Prasanta Kumar Mitra et al. Comparative Evaluation of Anti-Ulcer Activity of Root Stem and Leave of Murrya koenigii (Linn.) Spreng in Rats. J Med Plants Studies. 2013;1(3):158-165.