August 2008 Volume 4 Issue 2

Trade Science Inc.

Natural Products

An Indian Journal

Full Paper

NPAIJ, 4(2), 2008 [159-161]

Antihyperglycemic effects of *clitoria ternatea* linn. Leaves in albino rats

N.Gopal[1]*, V.S.Saravanan[2], S.Sengottuvelu[3]

[1]Department of Pharmaceutical Chemistry, Balaji Institute of Pharmaceutical Sciences, Laknepally, Narsampet, Warangal 506 331, Andhra Pradesh, (INDIA)

[2]Department of Pharmaceutical Chemistry, Nandha College of Pharmacy, Erode-638 052, Tamilnadu, (INDIA)

[3]Department of Pharmacology, Nandha College of Pharmacy, Erode-638 052, Tamilnadu, (INDIA)

E-mail : ngl72@yahoo.com

Received: 28th June, 2008 ; Accepted: 3rd July, 2008

ABSTRACT

The aqueous extract of *Clitoria ternatea* Linn leaves were screened for its antihyperglycemic activity in streptozotocin induced and glucose induced diabetic rats in doses of 100, 250 and 500 mg/kg body weight. At a dose of 100 mg/kg (bw), aqueous leaf extract did not produce significant antihyperglycemic activity, however a doses of 250 mg/kg of *Clitoria Ternatea* leaf extracts significantly reduced the glucose levels in the blood as compared to the standard drug, chlorpropamide at the end of 2 hrs. Increase of the dose does not show any significant increase in the activity. Aqueous leaf extract of *Clitoria Ternatea* significantly reduced ($p<0.001$) the elevation of glucose in the glucose-induced diabetic rats at 1 hr. From the results, it is concluded that the acute dose of aqueous extract of *Clitoria ternatea* Linn leaves exhibits good antihyperglycemic activity.

KEYWORDS

Clitoria ternatea;
Anti-hyperglycemic activity;
Streptozotocin;
Anti diabetic.

INTRODUCTION

Diabetes mellitus is a metabolic disease and is one of the most prevalent chronic diseases in the world affecting nearly 25 % of the population. Synthetic hypoglycemic agents in use can produce serious adverse effects and in addition, they are not suitable for use during pregnancy. Therefore, the search for more effective and safer hypoglycemic agents has been continued to be an important area of active research. In traditional methods, the indigenous hypoglycemic medicinal plants have been used as fresh petals, juice or dry powder. Recently, more works are being published on the antidiabetic activities of various plants in response to WHO recommendation of diabetes mellitus.

Clitoria Ternatea Linn belonging to the family of Fabaceae, cultivated throughout India, in hedges and thickets and also cultivated in garden. Roots, leaves and seeds are used traditionally for medicinal purposes. It has high calcium concentration, which is a significant source of calcium brewed as an herbal drink. Leaves are widely appreciated for its therapeutic effects in more than 30 ailments/diseases in different countries of the world. Most commonly it is used as anti-inflammatory, antimicrobial, analgesic, antithrombotic, antiemetic, antitumor, antitussive, immune supportive agents[1-6]. Epidemiological studies have shown that there are more diabetic patients in black tropical belt of the world to which both India and Nigeria belong. No systematic studies have been reported for its hypoglycemic effects. The present study focused on the antihyperglycemic effects of *Clitoria ternatea* leaf extract and justification of its use in the traditional system of medicine.

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EXPERIMENTALS

Preparation of plant samples

In the present study fresh *Clitoria ternatea* were collected from a local area in Erode district, Tamilnadu. The plant leaves were authenticated at the Department of Botany, Bharathiar University, Coimbatore, Tamilnadu. 100 gms of approximately weighed shade dried and powered leaves were extracted with water for 72 hrs. The extract was filtered and concentrated under reduced pressure and residue was obtained and dried (yield-12.3 %).

Animals

Swiss albino mice of either sex (weighing 20-25 gms) and Wistar albino rats of either sex (weighing 150-180 gms) supplied by Institute of Road and Transport Technology Perundurai Medical College, Erode, Tamilnadu, were used in the activity. The animals were housed under standard laboratory conditions with natural light and dark cycle. They were fed on standard pellet diet and water *ad libitum*. Animals were acclimatized to their environment for one week prior to experimentation.

Acute toxicity studies[7]

Healthy adult male albino Swiss mice, weighing between 20 and 25g were used in the present investigation. The leaf extract were tested up to 2g/kg body weight (as suspension in 0.5% CMC) in having groups of 6 animals by oral administration. The control groups of animals received only the vehicle (0.5% CMC). The animals were observed for 48 hrs from the time of administration of extract to record the mortality.

Antihyperglycemic study

Streptozotocin induced diabetes mellitus

Diabetes was induced by simple intravenous administration of Streptozotocin (STZ) 45-mg/kg body weight through the tail vein of rats. These rats were left for a period of 7 days to allow diabetic condition to develop. Group of six animals were selected for each dose of the extract and the normal and diabetic control group. On seventh day, single dose of the extract at a dose of 100, 250 and 500 mg/kg body weight was orally fed and water was fed to the normal control group intraperitoneally.

Glucose induced diabetic mellitus

Diabetes was induced on 16 hours fasted albino rats using 1 gm glucose/kg body weight, fed orally (dissolved in water) through a canula fitted needle attached to a syringe. Just after glucose fed single dose, leaf extract of the 250 mg/kg body weight was fed to study the antihyperglycemic effect of the extract.

Experimental procedure for glucose estimation

In all the groups, blood was collected from the animals by tail snipping at 0 hr, 1st hr, 2nd hr, 3rd hr and 4th hr. 0.1 ml of blood was collected into a centrifuge tube containing 1.5 ml of distilled water and 0.5 ml of disodium edetate solution. The blood mixture was shaken well and 0.2 ml of 0.3 M barium hydroxide was added with continuous shaking. 0. 2ml of 4.5 % zinc sulphate was then added, then mixture was centrifuged at 3000 rpm for 10 min at room temperature. 1 ml of the supernatant liquid was collected and was mixed with 2 ml of 5 % phenol followed by the addition of 5 ml conc. sulfuric acid. The tubes were shaken and allowed to cool. The absorbance was taken using spectrophotometer at 490 nm. Blood glucose values reported as mean ± SEM, in mg/dl, for six animals in each group. (Individual value was compared with the corresponding value as in the normal control group as well as in the diabetic control group)

Statistical analysis[8]

The results were analyzed statistically using Student's t-test and the level of significance for all determinations was evaluated at $p<0.05$, 0.005, and 0.001 in diabetic control animals.

RESULTS AND DISCUSSION

In the acute toxicity study, the aqueous extract of *Clitoria ternatea* leaves did not show any mortality up to a dose of 2g/kg body weight. Even at this high dose, there were no gross behavioral changes.

The mean blood glucose concentrations of all the groups were tabulated in TABLE 1. The data in TABLE 1 clearly indicates that the diabetic control group animals under study showed increase in blood glucose levels with maximum blood glucose level at 217 mg/dl.

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TABLE1: Antihyperglycemic effect of crude aqueous extract of *Clitoria ternatea* linn

S. no	Doses	Mean blood glucose concentration in mg/dl					
		0 hr (1st Day)	0 hr (7th Day)	1h	2 hr	3hr	4 hr
1	Control (Normal)	108±2.9	119±9.5	102±22.5	117±12.5	93±9.0	105±2.6
2	Control (Diabetic)	107±13.4	138±3.8	163±12.8	125±3.6	217±28.5	205±21.5
3	Chlorpropamide	109±7.3	153±2.9	125±3.5*	110±10.2*	105±9.3*	81±13.2**
	Streptozotocin induced diabetic model						
4	100 mg/kg (bw)	109±7.3	130±5.2	132±3.9	134±3.9	108±3.1*	84±3.7**
5	250 mg/kg (bw)	102±1.5	135±3.8	124±4.9	111±1.7*	84±2.9 **	84±2.2***
6	500 mg/kg (bw)	104±7.0	132±3.7	106±10.2*	82±2.9***	86±2.4**	81±4.2**
	Glucose induced diabetic model						
7	250 mg/kg (bw)	94±1.9	110±2.9	98±3.4***	72±2.4***	80±1.8***	90±3.0***

Values are mean ± SEM (n=6), * p< 0.05, ** p<0.005, *** p < 0.001

The Reference drug, chlorpropamide at a dose of 45 mg/kg body weight lowered the glucose values at the end of 1 hr, also giving lower values, almost always at the end of 3 hrs. The desired final glucose values were brought down close to the normal fasting blood glucose level within 3 hrs shows that definite hypoglycemic effects of the aqueous extract. At a dose of 100 mg/kg body weight of the leaf extract did not showed good activity against the standard drug Chlorpropamide. The effect of leaf extract at 250 mg/kg body weight is significantly comparable to normal control at 2 hrs itself. The increase of the dose to 500 mg/kg bodyweight does not show any significant increase in the activity. In glucose induced hyperglycemic group, the aqueous extract markedly reduced progressive elevation of blood glucose and the effect was statistically significant ($p<0.001$) after 1hr. It suggests that the extract could be reducing the absorption of glucose from the intestine.

The result shows that the acute administration of the aqueous extract of *Clitoria ternatea* has antihyper glycemic effects and results were compared to the standard drug, chlorpropamide. However, these two agents (*Clitoria ternatea* extract and chlorpropamide) could have different mechanism of action. Chlorpropamide produces hypoglycemia essentially by stimulation and reactivation of the residual pancreatic ß-cells, thereby, increasing the production of insulin, therefore, the action of chlorpropamide shown in this study confirms it's slow onset and long action by its observed effect on the blood glucose level of rats with STZ induced hyperglycemia. The *Clitoria ternatea* extract could be acting through an extra pancreatic mechanism such as increase in glucose utilization in the liver or other tissues, or by reducing glucose absorption form the intestine[9,10]. This assertion is derived from the fact that the antihypergly cemic effect of *Clitoria ternatea* extract was significantly greater in the glucose-induced hyperglycemia than STZ induced hyperglycemia.

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