Research Article

The Effect of *Enicostemma littorale* Blume on Adrenaline-Induced Hypertensive Rats

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ABSTRACT

Aim: The primary objective of the study was to investigate the possible effects of 70% Ethanolic extract of *Enicostemma littorale* Blume on adrenaline induced hypertensive rats. The hypertension was induced by Adrenaline at dose of 0.5 mg/kg i.p for 5 consecutive days.

Materials and Methods: The test doses of 250, 333.33 and 500mg of 70% Ethanolic extract of *Enicostemma littorale* (EEEL) and 10mg Propanolol were given orally and Intraperitonially respectively for 7 days. The various biochemical parameters like blood glucose, serum triglyceride, cholesterol, serum creatine phosphokinase, lactate dehydrogenase were measured.

Results: The extract administered groups had shown significantly lowered in LDH, CPK and pulse rate comparing to the normal groups. The biochemical parameters were lowered and significantly different (P•0.01), when compared with the disease control.

Conclusion: It can be concluded that 70% EEEL imparted a protective action against adrenaline induced hypertension in rats.

Keywords: *Enicostemma littorale* Blume, adrenaline, antihypertensive activity, Hypertensive rats.

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INTRODUCTION

Hypertension or high blood pressure, occasionally called arterial hypertension is a chronic medical condition in which the blood pressure in the arteries is elevated. It is themost common cardiovascular illness and is a major public health issue in developed as well as in developing countries[1]. Hypertension is the most significant modifiable risk factor for coronary heart disease, stroke, congestive heart failure, end stage renal disease, and peripheral vascular disease[2]. Human hypertension is most likely triggered by environmental influences such as increased salt intake, obesity and lack of exercise. Long-term hypertensive's frequently have other cardiovascular risk factors including high cholesterol levels, reduced high-density lipoproteins, diabetes, left ventricular hyper disorder trophy and obesity. An untreated hypertensive patient is acutely in stroke, coronary artery disease leading to myocardial infarction and acute renal failure[3]. Total peripheral vascular resistance is affected by the viscosity of blood, local and circulating substances and as well as autonomic nervous systems i.e. sympathetic and parasympathetic. The pathogenesis of essential hypertension is multi-factorial and highly complex which will be caused an increase in sympathetic nervous system activity, increase in production of sodium-retaining hormones and vasoconstrictors. The deficiencies of vasodilators such as prostacyclin, nitric oxide, inappropriate renin secretion and genetic predisposition. The pathogenesis of secondary hypertension will be caused by chronic kidney disease, renovascular disease, Cushing's syndrome,

pheochromocytoma, drugs such as non-steroidal anti-inflammatory drugs and oral contraceptives[4].

Enicostemma littorale Blume a glorious persistent herb belonging to the family Gentianceae. Upon literature survey, it was found that herb is used for the treatment of diabetes, elevated body temperature, stomach pain, heartburn and malaria. It is also reported to possess antitumor[5], antiarthritic[6], hypoglycemic[7] and antimalarial activities[8]. There are reports that the plant possesses flavonoids, alkaloids, catechins, saponins, sterols, triterpenoids, phenolic acids, flavonoids and xanthones and so on in the herb[9].

MATERIALS AND METHODS

Enicostemma littorale Blume herb was collected from the Kanavihalli near Harapanahalli in the month of June 2012. The plant was identified and authenticated by Prof K Prabhu, Department of Pharmacognosy, S.C.S. College of Pharmacy, Harapanahalli. A herbarium specimen No. SCSCOP.Ph.Col Herb.No.005/2011-2012 was preserved in our college museum. The dried powders of the *Enicostemma littorale* Blume herb were defatted with Pet. ether, chloroform and then extracted with 70% ethanol using Soxhlet apparatus. The extracts were concentrated under reduced pressure using rota flash evaporator and stored in airtight container in the refrigerator below 10°C. Phytochemical investigations of alkaloids, flavonoids, saponins, phenols, tannins, anthraquinone, cardiac glycosides, glycosides, triterpene, and Saponosides were carried out[10].

The Study was conducted on wistar albino rats of either sex, weighing between 150- 250g and was procured from Sri Venkateshwara Enterprises, Bengaluru. The animals were acclimatized for one week under laboratory conditions. They were housed in polypropylene cages and maintained at 27°C ± 2°C less than 12 hours dark/light cycle. They were fed with standard rat feed (Gold Mohur Lipton India Ltd.) and water *ad labium* was provided. Ethical clearance for handling the animals was obtained from the Institutional animal ethical committee (IAEC) prior to the beginning of the project work, with the registration no. is SCSCP/583/5/2011-12 CPCSEA and date is 26.11.2011. Initially blood estimation was noted down in experimental rats. The rats were injected adrenaline by intraperitonially for five consecutive days to induce hypertension. On 5th day animals were evaluated for the increase in blood parameters. To confirm the induction of hypertension pulse rate, body weight, serum cholesterol, blood glucose, serum triglycerides, serum lactate dehydrogenase and serum creatine phosphokinase levels were measured. The results were compared with that of the control rats that received only normal saline, the standard drug as Propanolol and the test rats receive 70% *Ethanolic Extract of Enicostemma Littorale* (EEEL).

Experimental design1:

Group 1: Negative Control- received Normal saline (0.9% w/v 2ml/kg/day, p.o) for 5days.

Group 2: Positive Control- Adrenaline (0.5 mg/kg i.p) for 5 days.

Group 3: Standard - Propanolol (10 mg/kg, i.p.) for 7days.

Group 4: Low dose of EEEL (250mg/kg/day, p.o) for 7 days.

Group 5: Medium dose of EEEL (333.33mg/kg/day, p.o) for 7 days.

Group 6: High dose of EEEL (500mg/kg/day, p.o) for 7 days.

Various doses of 70% EEEL given orally and commercial drug (Propanolol) were administered through intraperitoneal (I.P) route once daily for one week. Collected blood samples were analyzed for the determination of blood biochemical parameter level by auto-analyser (Star 21).

Statistical analysis:

To check the significance of data, following statistical test were performed: ANOVA: to see the variability within all the groups.

RESULTS

The phytochemical screening of 70% EEEL exhibited the existence of phenols, glycosides, anthraquinone,

tannins, alkaloids, saponins, and flavonoid. Cardiac glycosides and triterpene were absent.

Table 1 presents the mean body weights, heart weights and pulse rate of rats treated with 70% EEEL and propanolol before and 5 days and one week after induction of treatment. There was increase in the body weights and heart weights of all the animals throughout the experimental period. The increase in body weight was statistically significant (P<0.01). This increase in weight might be due to the extract effect and the nutritive. This is an indication that the plant is relatively non-toxic as decrease in body weight could mean toxicity.

There was seen a considerable (P<0.01) increase in the pulse rate of AIHR, when compared with the Normal control. The increase in pulse rate of animals or humans could be due to heart diseases or increased metabolic activity. Adrenaline increases metabolic activity as well as pulse rate. Treatment with 70% EEEL and Propanolol for one week significantly (P<0.01) decreased the pulse rate of hypertensive rats presented in table 1. The reduction in pulse rate of the hypertensive rats confirms the hypotensive effect of 70% EEEL. Its reductive ability is comparative to the standard drug (propanolol) used.

The excess of Pressure load on the heart such as from hypertension, consequences in pathological cardiac hypertrophy produces a reduction of cardiac function and eventually heart failure. This has been reported that glycolytic energy metabolism is stimulated in hypertensive cardiac hypertrophy induced by pressure over load due to aortic banding or pulmonary hypertension.

There was significant (P<0.01) decrease in blood glucose and cholesterol level of the experimental rats when compared with the disease control group for doses of the extract. Significant increase in serum triglyceride levels were observed in animals treated with the extract and propanolol.

The results of the present investigation demonstrated that 70% EEEL and propanolol significantly (P<0.01) decreased CPK and LDH activity, compared with the controls which indicates that the extract may act to reduce blood pressure.

DISCUSSION

Hypertension is a common debilitating illness among peoples in both developed and developing countries. Community surveys in industrialized countries have shown a prevalence of 15-33% in people aged 30 years[11]. The disease continues to be a leading cause of morbidity and mortality from the coronary artery disease and stroke. Fortunately, antihypertensive drug therapy is available to reduce blood pressure to a normal level is necessary to manage cardiovascular disease, coronary heart disease and other cardiovascular related complications. In this respect, herbal drugs are helpful and render encouraging results in comparison to synthetic drugs due to their fewer side effects and easy availability[12].

Table 1: Effects of *Enicostemma littorale* Blume and Propanolol on different parameters in Adrenaline induced rats.

Groups	Animal body Wt.(gm)	Wt. of the heart (gm)	Pulse rate (Bpm)	Blood glucose (mg/dl)	T.C.L (mg/dl)	T.G.L (mg/dl)	CPK (U/L)	LDH (U/L)
Control	159.0±5.859	0.603±0.034	251.2±10.65	88.77±9.712	113.3± 14.91	1.456±0.1769	14.58±2.846	7.592±3.231
Disease Control	244.2±4.902	0.926±0.021	379.3±16.47	156.3±19.27	187.5±9.539	0.071±0.007	107.5±3.720	60.78±5.405
Standard	155.0±2.236***	0.616±0.018***	257.0±5.26***	66.15±3.357***	108.2±6.803***	1.129±0.243**	40.68±0.754***	12.72±0.707***
70% EEEL(250mg/kg)	205.0±6.191NS	0.798±0.029NS	329.7±17.71NS	96.10±12.12NS	144.7±12.09NS	0.739±0.115NS	59.51±5.712NS	24.56 ±2.623NS
70% EEEL(333.33mg/kg)	180.0±2.236*	0.736±0.032*	285.2±12.45*	85.69±4.271*	136.6±5.418*	1.060±0.307*	54.87±5.068*	21.03±2.734*
70% EEEL (500mg/kg)	167.5±5.284**	0.678±0.047**	272.2±9.02**	68.56±12.18**	116.5±8.984**	1.103±0.233**	46.70±10.76**	18.14±1.387**

*Each value is expressed as mean ± SEM (n = 6), where, ns represents non significant; ***p<0.001 – highly significant; **p<0.01- very significant; *p<0.05- significant, when compared to adrenaline alone treated rats. One-way ANOVA followed by Dunnett's comparison test. AIHR= Adrenaline Induced Hypertensive Rats, ethanolic extract of leaves of Enicostemma littorale, group 1 was not induced nor treated and group 2 was induced but not treated.*

Standard values of different biochemical parameters were investigated. Except serum triglyceride level, the standard values of body weight, heart weight, blood glucose level and serum cholesterol level were high in adrenaline induced hypertensive rats when compared to control rats. Because of metabolic effects of adrenaline, serum triglyceride level was low in hypertensive rats[13]. This study was performed to analyze the differential effects of 70% EEEL and propanolol on heart weight, blood glucose level, serum triglyceride level, serum cholesterol level, pulse rate and body weight of hypertensive rats and compared with those of control rats. There was increase in the body weights, heart weights and pulse rate of all the animals throughout the experimental period. The increase in body weight, heart weight and pulse rate was statistically significant ($P<0.01$). The increase in pulse rate of animals or humans could be due to heart diseases or increased metabolic activity[14]. Adrenaline increases metabolic activity as well as pulse rate. Treatment with 70% EEEL and propanolol significantly ($P<0.01$) decreased the pulse rate of hypertensive rat. This decrease was dose dependant.

The effect of 70% EEEL and propanolol on blood glucose, serum triglyceride and cholesterol level are as presented in table 1. There was significant ($P<0.01$) decrease in blood glucose, total cholesterol levels when compared with the disease group. This significant decrease might be due to the effect of the extract, increased metabolic activity of adrenaline used to induce hypertension.

LDH and CPK release has been associated with cardiac tissue damage. A higher concentration of LDH and CPK could be a symptom of heart damage. The plant extract has protective effect on the biological utilization of the lipids than the propanolol[15].The results of the present investigation demonstrated that alcoholic extract and propanolol significantly ($P<0.01$) and ($P<0.01$) respectively decreased CPK and LDH activity, compared with the disease, which indicates that the extract may act to reduce blood pressure as presented in table 1. The present study revealed that 70% EEEL has got profound hypotensive activity and this study has similarity with previous investigation[16-17].

CONCLUSION

In conclusion, our investigation suggests that *Enicostemma littorale* Blume herb extract has got profound hypotensive activity and this study has correlation with previously reported investigations using other plants. The mechanism by which *Enicostemma littorale* Blume lowers blood pressure is not yet fully established. However, the anti hypertensive effect may be due to the stimulation of muscarinic receptors of the parasympathetic nerve by the compounds or their actions as an antagonist of α2-adrenergic receptors. The plant possesses significant quantity of alkaloid, flavonoid and tannin. It is attributed to show antihypertensive properties. Conclusively, the 70% ethanolic extract of Enicostemma littorale Blume herb has Antihypertension against adrenaline induced hypertension in rats.

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