

EVALUATION OF HYPOLIPIDEMIC AND ANTI-OBESITY ACTIVITIES OF *BIOPHYTUM SENSITIVUM* LINN EXTRACTS ON HIGH FAT DIET INDUCED HYPERLIPIDEMIC RATS

Rajanikant T Kakade*, Nagesh Sandu and KL. Senthilkumar

Padmavathi College of Pharmacy, Periyanahalli, Dharmapuri- 635 205, Tamilnadu, India.

ABSTRACT

Biophytum Sensitivum Linn., leaves and stems were extracted with Petroleum ether, Ethyl acetate and Methanol by successive hot extraction. All the extracts were evaluated for their in vitro hypolipidemic and anti-obesity effect in high fat diet induced rats. High fat diet treated animals have developed hyperlipidemia and obesity when compared with normal animals. Whereas MES and EAS significantly ($p \leq 0.01$) reduced the elevated levels of total cholesterol (TC), serum triglyceride (TG), LDL-cholesterol and VLDL-cholesterol, AST and ALT and elevate the decreased level of HDL-cholesterol. Other observations shows that rats treated with EAS & MES underwent a time-dependent reduction in body weight. These results suggest that, EAS extracts possess good hypolipidemic and anti-obesity activity. EAS has more potential than MES because of high phenolic and flavonoid content.

Keywords: *Biophytum Sensitivum* Linn., Anti-obesity, Hypolipidemic.

INTRODUCTION

Obesity is a global health problem, resulting from an energy imbalance caused by an increased ratio of caloric intake to energy expenditure^{1,2}. The medical problems caused by obesity begin at the head and end at the toes. Several of these problems contribute to the earlier mortality associated with obesity and include coronary artery disease, severe hypertension, impaired cardiac function, adult-onset (type 2) diabetes mellitus, obesity hypoventilation, sleep apnea syndromes, cirrhosis, venous stasis and hypercoagulability with an increased risk of pulmonary embolism and necrotizing panniculitis³.

It is estimated that 12 million deaths per year occur from cardiovascular diseases, while one million of deaths in the European country occur due to obesity per year⁴. Considerable advances have been made in diet, exercise and behavior approaches to treatment for obesity and new drugs with even better profile of pharmacological activity continues to be introduced on a regular basis.

Since available drugs have high side effects, while herbal treatment are safe and is relatively

cheap and locally available, around 80% of the world population is relied upon plant for their medication⁵.

Biophytum sensitivum linn is a variable annual herb belonging to family oxalidaceae. It is commonly found as a weed in open habitats during the rainy season throughout the hotter parts of India, ascending to 18m in the Himalayas, Tropical Asia, Africa and America⁶.

The plant is bitter, thermogenic, diuretic, lithontriptic, suppurative, expectorant, stimulant and tonic. It is useful in strangury, urinary calculi, hyperdipsia in bilious fever, wounds, abscesses, gonorrhoea, asthma, phthisis, stomachalgia and snake bite⁷. The plant is reported to possess tonic, stimulant and styptic properties and is used to treat chest complaints, insomnia, convulsions, cramps and inflammatory tumours and its ash is used in stomach ache⁸.

MATERIALS AND METHODS

Plant material and extracts

The stems of *Biophytum sensitivum* Linn. were collected from Chambharleni region of Nashik, in the state of Maharashtra (India) during

period of August - September. The powdered stems were successively extracted with solvent petroleum ether, ethyl acetate and methanol. After complete extraction extracts were collected and dried by using Rotavac evaporator. The ethyl acetate (EAS) and methanolic extracts (MES) were kept in airtight containers for future use.

Animals

Adult albino rats of either sex weighing around 160 – 200 gms were used. Animals were acclimatized to the experimental condition for one week prior to the experiment under controlled conditions of temperature ($27 \pm 2^{\circ}\text{C}$) and were housed in sterile polypropylene cages containing sterile paddy husk as bedding material with maximum of six animals in each cage. The rats were fed on standard food pellets and water *ad libitum*. The studies conducted were approved by the Institutional Animal Ethical Committee, Padmavathi College of Pharmacy, Dharmapuri, Tamilnadu.

Acute toxicity study⁹

Acute toxicity study of plant extract was performed as per the Organization for Economic Co-operation and Development (OECD) guideline 423. Based on these agreements, a limit test was performed to categorize the toxicity class of the compound and then main test was performed to estimate the exact 50% of lethal dose (LD50). No sign of toxicity observed even at the dose of 2000mg/kg, hence the dose range of 200 and 400 mg/kg was selected for EAS and MES extracts.

HYPOLIPIDEMIC & ANTI-OBESITY ACTIVITY

10, 11, 12

Experimental design

Fifty four albino rats weighing 160-200 gm were randomly divided into nine groups of six each and kept in their cages for 1 week prior dosing to allow for acclimatization to the laboratory conditions. The chronic experimental hyperlipidemia and obesity was produced in rats by the following treatment as shown in table1.

Collection of blood sample

1 ml of blood was collected by puncture of retro-orbital vein. The blood was allowed to clot for 30 minutes at room temperature. The serum was separated by centrifugation at 2000 rpm for

10 minutes. The serum was then used for the biochemical parameter estimation.

Statistical Analysis

The values were expressed as Mean \pm SEM are calculated for each parameter. For determining the significant inter group difference each parameter was analysed separately and one-way analysis of variance followed by Dunnett test.

RESULTS AND DISCUSSION

The animals treated with Simvastatin, EAS, and MES were showed significant ($P < 0.01$) decrease in body weight as compared to HFD group when measured on day 30 and day 45. (Table 2, Figure 1).

The animals treated with Simvastatin, EAS and MES were showed significant ($P < 0.01$) decrease in total cholesterol, HDL- Cholesterol, LDL-cholesterol, VLDL- Cholesterol and triglyceride level as compared to HFD group when measured on day 45 (Table 3).

When compared to normal control group the serum biochemical parameters like total Cholesterol, Triglyceride, LDL-C, HDL-C and VLDL-C levels were significantly higher in HFD induced obese rats. Simvastatin treated group exhibited a significant anti obesity activity by reducing all the biochemical parameters.

CONCLUSION

From the overall result of the biochemical and behavioural results, it could be inferred that EAS extracts showed hypolipidemic and antiobesity activity and activity was dose dependent and more at highest dose tested. Present studies reveal that EAS and MES can be used as effective hypolipidemic and anti-obesity agent. Further experiments are required to prove the mechanism and advantage of EAS and MES extracts over other drugs. Also the plants could be extended for the isolation and structure determination of the hypoglycemic principles.

ACKNOWLEDGEMENTS

Authors are highly thankful to Dr. K. L.Senthilkumar, Principal, Padmavathi College of Pharmacy, Dharmapuri, Tamilnadu, for providing the required infrastructure to carry out the research activities and Dr. N. Sandu for his constant support.

Table 1: Groups of EAS and MES in High fat diet (HFD) induce obesity in rats

Groups	Treatment	Dose	Evaluation parameters
I	CMC	1 ml/kg p.o	Biochemical parameters Total Cholesterol • Triglyceride • HDL-C • LDL-C • VLDL-C Morphological parameter Body weight
II	HFD	cholesterol 4%, cholic acid 1% and 1 ml coconut oil	
III	HFD + Sim	4 mg/kg p.o.	
IV	HFD + EAS	100 mg/kg p.o.	
V	HFD + EAS	200 mg/kg p.o.	
VI	HFD + EAS	400 mg/kg p.o.	
VII	HFD + MES	100 mg/kg p.o.	
VIII	HFD + MES	200 mg/kg p.o.	
IX	HFD + MES	400 mg/kg p.o.	

Table 2: Effect of extracts on body weight in HFD induced obesity in rats

Groups	Mean body weight (g)			
	Day 1	Day 15	Day 30	Day 45
Control	179.8±5.43	184.1±6.56	191.54±5.66	205.2±5.33
HFD	173.0±5.62	210.1±7.55#	253.52±7.42##	279.3±6.23##
HFD + Sim	177.3±7.51	183.1±7.54*	199.61±7.55**	217.0±4.55**
HFD + EAS100	178.3±5.78	198.2±7.44	239.3±7.11	238.33±5.67*
HFD + EAS200	177.2±5.33	193.6±7.21	218.7±7.33*	230.21±6.66**
HFD + EAS400	174.1±6.46	190.0±7.32	201.2±7.66**	221.32±7.55**
HFD + MES100	177.3±6.87	197.2±7.44	230.3±5.35	240.2±7.66*
HFD + MES200	174.8±6.11	191.2±5.89	216.7±5.67*	237.7±6.87*
HFD + MES400	173.5±5.66	185.0±6.09	200.2±6.44**	220.0±7.88**

Results are presented as Mean ± SEM, (n=6). ANOVA followed by Dunnett test.
 ** p<0.01 compared with control. *p<0.05, ##p<0.01 when compared with HFD.

Table 3: Effect of extracts on lipid profiles in HFD induced obesity in rats

Group	Total Cholesterol	HDL-Cholesterol	LDL-Cholesterol	VLDL-Cholesterol	Triglyceride
Control	100.75 ± 3.18	52.13 ± 4.89	38.54 ± 5.66	49.50 ± 5.68	95.92 ± 4.33
HFD	191.63 ± 9.82##	31.63 ± 4.73##	115.38 ± 4.46##	82.75 ± 4.97##	198.13 ± 8.87##
HFD + Sim	106.63 ± 7.54**	38.65 ± 4.42**	65.17 ± 6.65**	55.25 ± 4.16	129.63 ± 7.29**
HFD + EAS100	182.87 ± 8.61	32.25 ± 2.93	101.18 ± 4.67	69.25 ± 5.82	181.12 ± 6.57
HFD + EAS200	160.25 ± 7.88*	37.75 ± 3.76*	88.16 ± 6.66*	63.00 ± 5.38*	170.63 ± 5.46
HFD + EAS400	136.63 ± 7.54**	40.38 ± 2.45**	70.46 ± 7.85**	58.63 ± 4.87**	154.88 ± 7.60**
HFD + MES100	188.88 ± 7.09	30.28 ± 4.29	103.32 ± 6.85	77.62 ± 6.66	177.25 ± 8.34
HFD + MES200	169.75 ± 6.45*	37.03 ± 3.45*	90.18 ± 6.04	64.12 ± 5.38	159.37 ± 7.83*
HFD + MES400	135.13 ± 7.21**	43.77 ± 4.43**	82.88 ± 4.55**	56.63 ± 3.16**	143.87 ± 6.07**

Results are presented as Mean ± SEM, (n=6). ANOVA followed by Dunnett test.
 ** p<0.01 compared with control. *p<0.05, ##p<0.01 when compared with HFD.

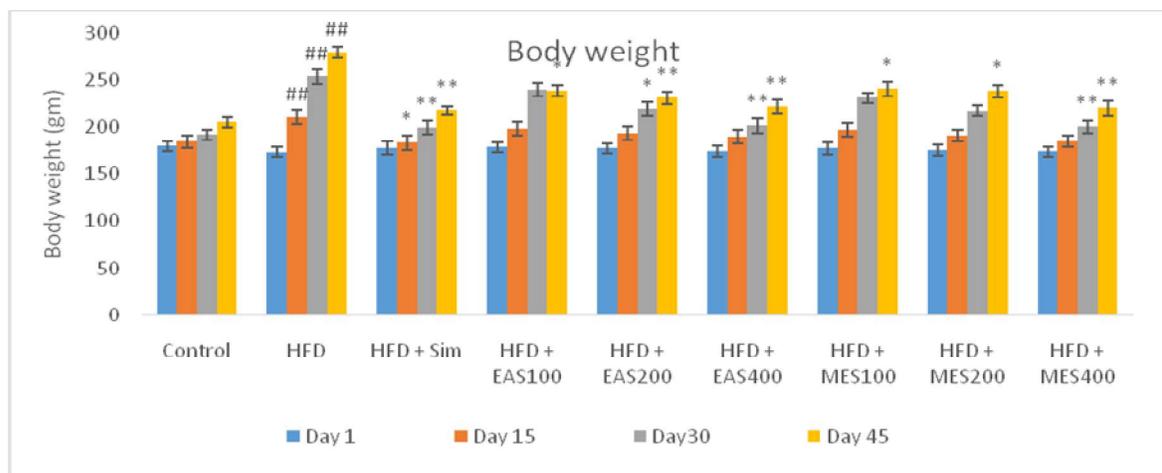


Fig. 1: Effect of extracts on body weight in HFD induced obesity in rats

REFERENCES

1. Larsson B, Bjorntorp P and Tibblin G. The health consequences of moderate obesity. *Int J Obes*. 1981;5:97-116.
2. Hartz AJ, Rupley DC, Kalkhoff RD and Rimm AA. Relationship of obesity to diabetes: influence of obesity level and body fat distribution. *Prev Med*. 1983;12:351-357.
3. Sugeran HJ. The pathophysiology of severe obesity and the effects of surgically induced weight loss. *Surg Obes Relat Dis*. 2005;1(2):109-19.
4. Coughlan BJ and Sorrentino MJ. Does hypertriglyceridemia increases risk for CAD. *Journal for Primary Care Physicians*. 2000;108(7):876-82.
5. Rakh MS and Chaudhari SR. Evaluation of analgesic activity of *Momordica dioica* Roxb. Willd fruit Pulp. *International Journal of Pharmacy Research*. 2010;1(9):53-56.
6. Parrotta JA. *Healing Plants of Peninsular India*, CABI publishing USA, 2001;557.
7. Prajapati ND, Purohit SS, Sharma AK and Kumar T. *A Handbook of Medicinal Plants*, Agrobios (India), 2003;91.
8. *The Wealth of India*, Publications and Information Directorate, CSIR, New Delhi. 1988;2B:151-152.
9. OECD, guidelines for testing of chemicals. 425: Acute oral toxicity-up-and-down procedure, 2001;1-26.
10. Umbare RP, Patil SM, Mate GS and Dongare SS. Hypolipidemic Activity of *Orthosiphon stamineus* Benth Bark Extract. *Journal of Pharmacy Research*. 2009;2(11):1735-8.
11. Kaur G and Kulkarni SK. Antiobesity effect of a polyherbal formulation, OB-200G in female rats fed on cafeteria and Atherogenic diet. *Indian Journal of Pharmacology*. 2000;32:294-299.
12. Yoriko D and Kouji M. Anti-hyperglycemic and Anti-hyperlipidemic Effects of Guava Leaf Extract. *Nutr Metab*. 2010;7:9.