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Evaluation of Antihyperlipidemic Activity of Ethanolic Extract of Lathyrus Sativus Linn. on Rats.

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KEYWORDS

Lathyrus sativus Linn., triton x-100, high fat diet, atorvastatin

ABSTRACT:

Introduction Hyperlipidemia is a major contributor to atherosclerosis and its accompanying conditions, including coronary heart disease, ischemic cerebrovascular disease, and peripheral vascular disease. The majority of morbidity and mortality among middle-aged people's mortality and morbidity and older adults still results from atherosclerosis-related events, although their incidence has decreased in the United States. As a result of the obesity epidemic and the ageing of the American population, both the incidence and the absolute number of annual events will rise over the coming ten years..

Objectives Evaluation of Antihyperlipidemic Activity of Ethanolic Extract of Lathyrus sativus Linn. on rats.

Methods: Screening of antihyperlipidemic was done by using *Lathyrus sativus Linn*.. Leaves part extracts used for antihyperlipidemic activity leaves (200mg/kg). Hyperlipidemia was administered in experimental animals using triton X-100 administered intraperitoneally at a dose of (100 mg/kg). In order to collect blood for triton-induced hemolysis, retroorbital punctures were performed every 24 hours. hyperlipidemia and 28 days for high fat diet induced hyperlipidemia. Antihyperlipidemic activity of *Lathyrus sativus Linn*. plant extracts were evaluated by estimation of lipid profiles and standard drug Atorvastatin for 28 days. The biochemical parameters such as The levels of Total Cholesterol, Triglycerides, Very Low Density Lipoprotein (VLDL), High Density Lipoprotein (HDL), and Low Density Lipoprotein (LDL) are assessed.

Results: Estimation of lipid profile shows that *Lathyrus sativus Linn*. leaves extract (200mg/kg), *Lathyrus sativus Linn*. leaves extract (200mg/kg) shows the less significant antihyperlipidemic activity. It decreases TC, TG, LDL, VLDL and increases HDL levels. *Lathyrus sativus Linn*. leaves extract 200 mg/kg shows significant antihyperlipidemic activity as standard drug Atorvastatin.

Conclusions: This finding tends to reveal that the hyperlipidemic effects of *Lathyrus sativus Linn*. are similar to the effect of standard drug Atorvastatin. This plant can get in consideration for the searching new drug to treat hyperlipidemic from plant.

1. Introduction

Hyperlipidemia is a major contributor to atherosclerosis and its accompanying conditions, including coronary heart disease, ischemic cerebrovascular disease, and peripheral vascular disease. The majority of morbidity and mortality among middle-aged people's mortality and morbidity and older adults still results from atherosclerosis-related events, although their incidence has decreased in the United States. As a result of the obesity epidemic and the ageing of the American population, both the incidence and the absolute number of annual events will rise over the coming ten years. (1), Dyslipidemia, which

includes hyperlipidemia, hypercholesterolemia, and low HDL levels, is a major contributor to the increased atherogenic risk seen in developed nations all over the world. Genetic disorders and a lifestyle diet rich in calories, saturated fat, and cholesterol also play a part in this condition. (2)

Both traditional and modern systems of medicine continue to benefit from the therapeutic benefits of medicinal plants.(3) According to the World Health Organization (WHO), up to 90% of the population in developing countries uses plants and its products as traditional medicine for primary health care [4]. Medicinal plants also offer good prospects to finding new drugs

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particularly against conditions for which modern drugs are inadequate.

But still most of the medicinal plants are not scientifically validated. Scientific studies on those plants are likely to provide valuable medicines [5].

Lathyrus sativus Linn. Miq. (Moraceae) is an important traditional medicinal plant distributed throughout India, mostly near to the Indian temple for the spirituality. It has several vernacular names including peepal tree and arasa maram. The bark of the plant contains carbohydrates, flavonoids, aminoacids, steroids, saponins and tannins etc. are present [6]. Its fruits and leaves contain flavonoids and leaves also contain sterols.7, Flavonoids and sterols are known to possess antidiabetic activity in various other plant species.8 So, it was thought that fruits and leaves may also possess antidiabetic, antihyperlipidemic and anti-oxidant activities.

The antihyperlipidemic effect of the leaves of *Lathyrus sativus linn* has not been investigated despite the pharmacological properties mentioned above. As a result, the current study was started to assess the ethanol extract of *Lathyrus sativus linn*.'s antihyperlipidemic efficacy.

2. Objectives

Lathyrus sativus linn. are rich source of polyphenolic compounds, flavanoids which are responsible for strong antioxidant properties that help in prevention and therapy of various oxidative stress related diseases such as neurodegenerative and hepatic diseases.

Antioxidant plant have a property to treat many disease like malaria, anamia, diabeties, AIDS, inflamation, analgesic etc. *Lathyrus sativus Linn*. are important ingredients in many Ayurvedic and traditional formulations. The barks, leaves, fruits and latex are considered to be very effective in various treatments, such as diabetes, skin diseases, ulcers, dysentry, diarrohoea, stomachache, piles and as carminative, astringent, anti-inflammatory, antioxidant and anticancer Agent.

Lathyrus sativus Linn. have a property of antioxidant and other valuable oil shows in literature survey we found that it can have antihyperlipidemic activity.

3. Methods

Materials And Methods

Collection & Authentification of plant

The fresh leaves of *Lathyrus sativus Linn*. were collected from local area of Bhopal. Plants were confirmed by Department of Botany, Dr. Hari Singh Gour University Sagar.

Preparation of leaves extract

Extract preparation was carried out according to the method of Oktay, et al, 2003 (9). After collection, *Lathyrus sativus Linn*. leaves were shade-dried for five days and then ground. 0.95 kg of powdered medicinal material was extracted in an airtight container at a ratio of 1:2 (w/v) ratio of 99 percent pure ethanol. In a steam bath, the produced extract was dried, and the dried mass was weighed and recorded. The yield percentage was determined. The weight of the dried crude extract that was recovered was roughly 0.16 g, reflecting a yield of 17.16 percent.

The process consist of keeping the crude drug in intimate contact with whole of the menstrum in a closed vessel with occasionally shaking for 7 days, straining, pressing the marc. Mixing the liquid, & finally clarifying by subsidence or filtration. The drug should be properly communicated. The cellular structure get penetrated & the soluble portion are softening & dissolved. Occasionally shaking bring about a rapid equilibrium between the intra and extracellular fluid. A closed vessel is recommended so as to prevent loss of menstrum. As the degree of pressing the marc may vary the final product in not adjusted to any complete extraction. The drug menstum ratio is 1:10. a sediment may form on standing for a few day, before use. Maceration process is very simple & does not require a skilled operator.

Doses

200 mg/kg body wt/day of powder of *Lathyrus sativus Linn*. dissolved in 5ml of distilled water and administered to test rats using a metal canula on a syringe.

Selection of animals

The experiments conducted in this study used adult Wistar rats weighing 130-165g. All rats were housed in an animal room at the Truba College of Pharmacy in Bhopal, Madhya Pradesh, India, which was approved by the Institutional Animal Ethical Committee. During the study period, commercial pellet food and water were given to the rats at their discretion. 8 groups of 6 rats each were formed from a total of 48 rats.

Screening Models

1:High Fat Diet-induced hyperlipidemic model

The animals were chosen, weighed, and then given unique identification tags. Rats have developed hyperlipidemia after receiving an atherogenic weight-loss diet orally for 20 days. The rats were then administered plant extracts suspended in 2% acacia at a dosage of 200 mg/kg b.w. once daily in the morning after being gastrically intubated for 14 days.

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During these days, the dose of atherogenic food was maintained in all groups. Control animals were given a high fat diet and vehicle. At the last phase of therapeutic, the animals have been used to measure a number of biochemical markers. A rat's heart was punctured while it was under ether anaesthesia to collect blood, which was afterwards centrifuged for 30 minutes at 2000 rpm to obtain serum (10).

Experimental Design

Table 1. Doses & Group schedule for the High Fat Dietinduced hyperlipidemic model

SL.NO	GROUP	TREATMENT	ANIMAL USED
1	I	Positive control received a 2% acacia diet for 20 days (p.o.).	6
2	II	2% acacia + High fat diet for 20 days	6
3	Ш	Positive control received standard drug atorvastatin (10mg/kg/ day p.o.) for 14 days	6
4	IV	Aqueous Extract (200mg/kg/day) fine suspension of 2% acasia + High fat diet for 14 days	6
5	V	Ethanol Extract (200mg/kg/day) fine suspension of 2% acasia + High fat diet for 14 days	6

2: Triton-induced hyperlipidemic model

Animals fasted for 18 hours will be administered intraperitoneally with 100 mg/kg body weight of saline Triton (Triton x-100). Plant extracts in a dose of 200 mg/kg body weight, was ingested after gastric intubation. After the injection of triton, the first dose is given immediately, after 20 hours the second dose follows and the extraction process continues for 7 days. Animals had been employed for many biochemical measures after 7 days of administration. Under ether anaesthesia, blood was drawn from the rat's heart and centrifuged for 30 minutes at 2000 rpm to get serum. (11).

Experimental Design

Table 2. Doses & Group schedule for Triton-induced hyperlipidemic model

SL.NO	GROUP	TREATMENT	ANIMAL USED		
1	I	Positive control received 2% acacia for 7 days (p.o.).	6		
2	II	Negative control received 2% acacia + triton (100 mg/kg) for a days (p.o.).	6		
3	Ш	18 hours. after triton positive control received standard drug atorvastatin (10 mg/kg/ day p.o.) for 7 days.	6		
4	IV	18 hours. after triton Aqueous Extract (200mg/kg/day) of fine suspension of 2% acasia was administered for 7 days.	6		
5	V	18 hrs. after triton Ethanol Extract (200mg/kg/day) a fine suspension of 2% acasia was administered for 7 days.	6		

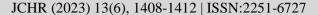
Blood Samples Collection

Blood was drawn from the patient's heart using a heart puncture on the eighth day of Triton and the twenty-first day of their hyperlipidemic therapy. Serum collected by immediately centrifuging blood samples in a Remi ultra cooling centrifuge for 30 minutes at room temperature while operating at 2000 rpm is used to instantly determine serum lipid profiles (TG, LDL, TC, HDL, and VLDL).

Statistical analysis

Mean SEM was used to express the findings. One-way ANOVA and the Tukey test were used in the statistical analysis, which was done with the use of the Graph Pad Instant

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 $52.15 \pm$

170.84

0.03

22.03±0

43.08±0

.01

49.46

 ± 2.7

39.33

programme. P value of 0.05 or above were regarded as statistically significant.

4. Results

Table 3. *Lathyrus sativus Linn*.'s impact on blood biochemical variables in rats with hyperlipidemia brought on by triton.

variables in rats with hyperinpidenna brought on by thron.						up	ml/	2±1.5	7±2.2	±2.6	±0.02	.02		
GRO UPS	DOS	TC	TG	HDL	LDL	VLDL (mg/dl)	(diet)	kg	a***	a***		a***	a***	
	Е	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)		3.Gro up	10 mg/	136.4 5±2.6	131.4 2±2.4	76.11 ±1.7	35.27± 0.09	27.09±0 .04	
1.Gro up (vehicl	1 ml/k g	111.35± 1.2	91.16±2 .8	46.35±2 .3	47.98±0 .04	19.04±0 .03	(std.+ diet)	kg	a**,b ***	a***, b***	a***, b***	a***,b ***	a***,b* **	
e) 2.Gro up (triton)	100 mg/k g	153.55 +4.4 a***	154.25 +4.2 a***	28.66+ 1.8 a**	95.23±0 .08 a***	31.64±0 .03 a***	4.Gro up (aqs.+ diet)	200 mg/ kg	216.3 5±2.6 a***, b***,	203.2 7±1.6 a***, c***	66.35 ±3.3 a***, b***	110.57 ±0.09 a***,b ***.	41.46±0 .02 a***,b* **,	
3.Gro	10 mg/k	121.17 ±2.4 b***	91.45 + 7.6 b***	56.85 +1.2 b***	47.29±0 .04 a***.b*	19.07±0 .04 b***			c***			c***	c***	
(std.+ triton)	g	Davas	Davas	D	**	Darrie	5.Gro up	200 mg/	179.3 7±3.6	166.3 5±1.7	69.46 ±2.5	77.85± 0.02	34.07±0 .02	
4.Gro up (Aqs.+ triton)	200 mg/k g	136.28 +2.3 a***,b* **, c**	133.39 +5.5 a***,b* , c***	36.57 + 3.3 c***	74.24±0 .01 a***,b* **, c***	27.48±0 .02 a***,b* **, c***	(etoh. + diet)	(etoh.	kg	a***, b***, c***,	a***, b***, c***	a***, b***	a***,b ***, c***,d	a***,b* **, c***,d*
5.Gro up (Etoh.	200 mg/k g	123.10 + 1.3 a*,b***	105.57 +3.3 a***,b* **,	51.23 +5.3 b***,d*	51.97±0 .02 a***,b* **,	21.93±0 .01 a***,b* **, D	Discussion	<u> </u>	d***			***	**	

1.Gro

up (vehic

le)

2.Gro

ml/

kg

1.5

121.6

 2 ± 1.3

251.2

106.1

4 + 3.0

211.3

Data obtained were analyzed by one-way ANOVA followed by Tukey Multiple Comparisons Tests. Each value represents mean \pm SEM; n=6. **p<0.01*p<0.05, ns p> 0.05. p< 0.001*** A. Significant difference compared to negative control group

- b. Significant difference compared to the control group
- C. Significant difference compared to the standard group
- d. Significant difference compared to the Aqueous group

Table 4. Effect of *Lathyrus sativus Linn*. on serum biochemical parameter in diet-induced hyperlipidemic rat model.

GRO	DO	TC	TG	HDL	LDL	VLDL
UPS	SE					
		(mg/	(mg/	(mg/	(mg/dl	(mg/dl)
		dl)	dl)	dl))	

According to Table 3-4 findings, *Lathyrus sativus Linn*. at a dosage level of 200 mg/kg was effective as a hypolipidemic drug.

Rats treated with Triton X-100 injection (100 mg/kg) effectively developed hyperlipidemia by having higher blood LDL-C levels TC and TG. The effect of *Lathyrus sativus Linn*. ethanol extracts on blood lipid profile levels is shown in Table 3. Treatment with *Lathyrus sativus Linn*. ethanol extracts at doses of 200 mg/kg significantly reduced blood TC, TG and LDLC levels compared to the hyperlipidemic control group (p 0.001) and increased serum HDL-C levels (p 0.001). The change in lipid levels in groups II, III, and IV was the same as that of the atrovastatin-treated group of rats. The one that significantly reduced elevated cholesterol levels from three servings.

The *Lathyrus sativus Linn*. demonstrated protective effects at a dose of 200 mg/kg and significantly decreased the raised blood TC, LDL-C, and triglyceride levels brought on by diet. At 200

triton)

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mg/kg, the effects were identical to those of the standard dosage of atorvastatin (Table 4).

200 mg/kg aqueous and ethanolic extract of *Lathyrus sativus Linn*. leaves were administered orally for 14 days. In contrast to the significant increase in HDL-C levels (p0.01), extracts from *Lathyrus sativus Linn*. leaves also significantly reduced the rise in blood total cholesterol, triglycerides, LDL-C and VLDL-C levels.

This study demonstrated the efficacy of Lathyrus sativus Linn. as a hypolipidemic drug when administered at a dosage level of 200 mg/kg. Further investigation would be required to determine the active constituents responsible for the activity and mechanisms of these effects in order to recover the problems in lipid metabolism shown in hyperlipidemic condition from the active ingredient found in plants. The presence of flavonoids lignans, and alkaloids, as the principal chemical ingredients of the Lathyrus sativus Linn. has been documented in chemical investigations and has been validated by phytochemical screening. It is likely that one or more of these pharmacologically active substances had a part in the Lathyrus sativus Linn.'s demonstrated hypolipidemic action. These phytochemicals are said to provide nutritional advantages. The injection of these substances to hypercholesterolemic and hypertriglyceridemic rats has reportedly been shown to significantly drop lipid levels and ameliorate dyslipidemia.

Conclusion

For a wide range of illnesses, natural therapies have been studied for decades. Even though *Lathyrus sativus Linn*. has drawn attention for its positive benefits, its medicinal and pharmacological qualities have received little scientific backing up to this point. In the current investigation, Triton X-100 (100 mg/kg) caused hyperlipidemic rats and diet-induced hyperlipidemic rats were used to test *Lathyrus sativus Linn*. for its antihyperlipidemic effect.

According to the current investigation, *Lathyrus sativus Linn*. was effective as a hypolipidemic drug when administered at a dosage level of 200 mg/kg, and its aqueous and ethanolic extract had effects comparable to those of atorvastatin. It was proven that the active ingredients in *Lathyrus sativus Linn*.'s aqueous extract may be the source of the substance's considerable anti-oxidant and hypolipidemic effects. However, more pharmacological testing is required to identify the potential mechanism of action.

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