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Assessment of hypolipidemic potentials of *Rivina humilis* against high fat diet induced hyperlipidemia in wistar rats

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Abstract

Objectives: The current investigation was carriedout to determine hypolipidemic potentials of ethanol extract of *Rivina humilis* against high fat diet induced hyperlipidemia in wistar rats.

Methods: Defatted powdered drug of ariel parts of *Rivina humilis* was subjected to ethanol extraction using soxhlet extractor. The high fat diet (HFD) induced hyperlipidemia in rat model was used for the present investigation in which all experimental rats were fed with HFD and treated with ethanol extract of *Rivina humilis* (EERH) for 21 days. Blood samples were collected from all the animals on day 21st after one hour of administration of the drugs and serum lipid profile (total cholesterol, triglycerides, HDL, LDL and IDL) was estimated. All animals were sacrificed after the blood sampling and liver samples were collected.

Results: The therapeutic rats ingested with extract of *Rivina humilis* and standard drug atorvastatin have shown significant reduction in serum cholesterol, serum triglycerides, LDL and increase in HDL indicating its ability to attenuate effect of HFD.

Conclusion: The ethanol extract of *Rivina humilis* shown significant hypolipidemic effects against HFD induced hyperlipidemia wistar rats by inhibiting cholesterol synthesis in liver.

Keywords: Hyperlipidemia, *Rivina humilis*, lipid profile, high fat diet

Introduction

A significant risk factor for the onset and development of cardiovascular diseases is hyperlipidemia, which is marked by increased levels of cholesterol, triglycerides, cholesterol esters (VLDL and LDL) as well as decreased HDL cholesterol concentrations in blood [1, 2]. Numerous factors, including dietary components, uncontrolled diabetes mellitus, excessive alcohol use, and stress [3, 4], might contribute to hyperlipidemia. Hyperlipidemia can exacerbate conditions such as atherosclerosis, myocardial infarction (MI), coronary artery disease (CAD), angina pectoris, and cerebral ischemic stroke [4]. Important classes of drugs presently used for the management of hyperlipidemia include are cholesterol synthesis inhibitors (statins), inhibitors of lipolysis (nicotinic acid and fibrates) bile acid sequestrants (cholestipol) possess significant serious adverse reactions such as liver damage, rhabdomyolysis and renal failure and hence pharmacological management of hyperlipidemia remains still unsatisfactory [5]. Hence studies on medicinal plants are increasing worldwide and screened for various complications such diabetes, CVS disorders, cancers and other life style diseases.

Ayurveda uses several medicinal plants that have been demonstrated to have hypolipidemic properties. Numerous investigations on herbals by scientists have revealed that natural remedies may be superior to synthetic medications in treating hyperlipidemia. Utilizing herbal treatments that have historically been employed in the pharmaceutical business is essential for the efficient management of diabetes and the secondary issues that are associated to it [9].

The *R. humilis* is a species of flowering plant in the family *Petiveriaceae*. It is commonly known as blood berry and used folklore and other traditional medicine for the treatment of various diseases. *R. humilis* is one among those plants used traditionally for the treatment of diabetes mellitus and hyperlipidemia but there is no scientific data available for the same [10-11]. Though the plant was extensively used in traditional medicine to reduce serum lipids, there is a lack of scientific evidence for the same. Hence the objective of the present study is to evaluate and provide scientific data for the hypolipidemic potentials of *Rivina humilis* against high fat induced hyperlipidemia in experimental rats.

Materials and methods

Collection and authentication of plant material

The ariel parts of *Rivina humilis* were collected and authenticated by Dr. Madhavachetty, HOD, Department of Botany, Sri Venkateswara University, Tirupati.

Preparation of ethanol extract

The ariel parts of *Rivina humilis* were dried under room temperature immediately after collection and subjected to milling to collect the coarse powder. About 250gm of coarse powder of *Rivina humilis* was first defatted with petroleum ether (40°-70°C) and defatted coarse powder was again subjected for extraction with ethanol for 72 hours using soxhlet apparatus [12].

Preliminary phytochemical investigation

The ethanol extract of *Rivina humilis* (TVME) was investigated for the preliminary phytochemical compounds according to standard protocol described by Khandelwal [13].

Animals

Healthy albino Wistar rats 180-200 of weight range were procured from Sri-Venkateswara Enterprises, Bengaluru. All animals were housed in animal house facility of East West College of Pharmacy provided with well ventilation and standard temperature condition between 28±2^{0}C. The animals were provided to access feed (standard laboratory plellets) and drinking freely. The research protocol was approved by IAEC, IJAHSM (Ref. no. IJAHSM/IAEC/2014/03) with the permission from Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India.

Assessment of methanol extract of *Rivina humilis* hypolipidemic activity

The hypolipidemic activity of ethanol extract of *Rivina humilis* was evaluated against induced and high fat diet induced hyperlipidemia in rat's models [14, 15, 16].

Hypolipidemic activity of TVME against high fat diet (HFD) induced hyperlipidemia preparation of (HFD)

The High Fat Diet was prepared according to procedures described in previous studies. The composition of HFD consisting of Powdered Normal protein diet (365 g), Lard (310 g), Casein (210 g), Cholesterol (10 g), Vitamin and Minerals (60 g), D1-Methionine (3 g), Yeast powder (1 g) and Sodium chloride (1 g). All the ingredients of High Fat Diet were mixed properly using mixer and made in the form pellets using required amount of distilled water and allowed for drying under shade.

Group design: This study was also consisting of six group of albino rats containing 6 animals in each group and details of treatment are as follows.

Evaluation parameters

Biochemical parameters: On the 21st day of study, blood samples from all the animals were collected estimated for Total cholesterol, Triglycerides, LDL, HDL, VLDL, Creatinine, urea and BUN. The weight gain of animals during study period was also calculated [14, 15, 16].

Results

Preparation of extract

The percentage yield of methanol extract of *Rivina humilis* was 8.19% w/w.

Preliminary phytochemical study

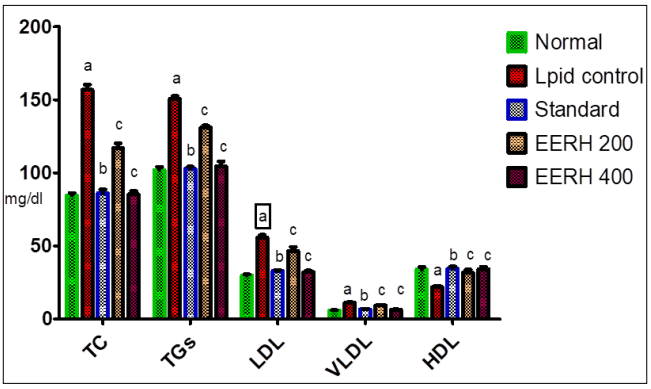
The preliminary phyto-chemical investigation for the methanol extract of *Rivina humilis* reveals the presence of poly phenols, flavonoids, tannins, steroids, alkaloids and carbohydrates.

Evaluation of anti-hyperlipidemic activity of methanol extracts

In the present study conducted to determine antihyperlipidemic activity of ethanol extract of *Rivina humilis* ingestion of high fat diet caused significant weight gain and also significant increase in the Total cholserol, Triglycerides, LDL and VLDL concentrations in the vehicle control group compare to normal animals. But co-administration of Atorvastatin and TVME significantly reduced above mentioned parameters in therapeutic animals compare to vehicle control rats. The concentration of HDL was significantly declined vehicle control group compare to normal while its range was significantly increased in therapeutic group of animals treated with medium and high doses of TVME and standard drug compare to lipid control animals. But there was no significant change in the biochemical parameters of rats treated with low dose of methanol extracts [Table 1].

Table 1

Sl. No.	Group	Treatment
I.	Normal	Wistar rats administered with once a day for 21 normal Saline 2ml/kg
II.	Lipid control	Animals were given with 30g of HFD every day and 2% tween 80 for 21 days.
III.	Standard	Animals were given with 30g of HFD every day and standard drug atorvastatin (mg/kg.,p.o) orally for 21 days.
IV.	EERH-200mg	Animals were given with 30g of HFD every day and medium dose of ethanol extract of *Rivina humilis* orally for 21 days.
V.	EERH-400mg	Animals were given with 30g of HFD every day and high dose of ethanol extract of *Rivina humilis* orally for 21 days.



Values are mean ± S.E.M, n=6 symbols represent statistical significance.,
[a]p<0.001 Normal control vs Lipid control, [b]p<0.001 Lipid control vs Standard and [c]p<0.001 Lipid control vs Standard,

Fig 1: Effect of ethanol extract of *Rivina humilis* on lipid profile against HFD induced hyperlipidemia

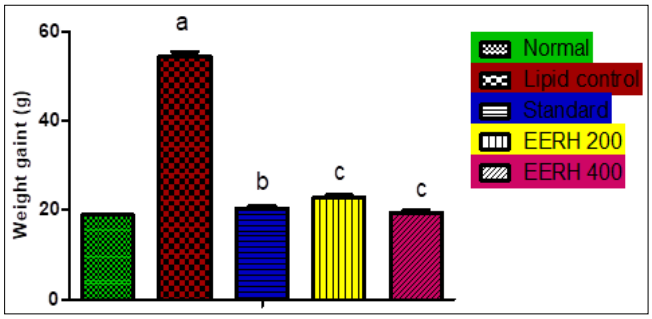


Fig 2: Effect of ethanol extract of *Rivina humilis* on body weight against HFD induced hyperlipidemia

Discussion

Atherosclerosis and its accompanying diseases, such as peripheral vascular disease, ischemic cerebrovascular disease, and coronary heart disease (CHD), are largely brought on by hyperlipidemia. Among these, ischemic heart disease is intimately associated with hypercholesterolemia and hypertriglyceridemia. CHD risk is decreased by lower serum cholesterol levels. Reducing the risk of developing

ischemic heart disease or the occurrence of further cardiovascular or cerebrovascular disease is the major goal of treatment for people with hyperlipidemia [17]. Several negative effects of the hypolipidemic medications now on the market have been reported. Hyperuricemia, diarrhoea, nausea, myositis, stomach irritation, flushing, dry skin, and altered liver function are all caused by the usage of synthetic medicines. In this regard, herbal medicines are proven to be effective drugs to reduce hyperlipidemia with minimum side effects and hence there is a scope to develop herbal remedy for the hyperlipidemia. High fat diet induced hyperlipidemia, triton induced hyperlipidemia in rats model had earlier been reported as ideal *in vivo* models for testing antihyperlipidemic drugs. Several studies reported that enriched fatty diets cause elevation of plasma TC and LDL cholesterol. High levels of TC and most importantly LDL cholesterol are predictors of atherosclerosis. Another research showed that triglycerides are directly or indirectly related to coronary heart diseases [18, 19]. In the present study, high fat diet induced hyperlipidemia in rats model was used for the evaluation of hypolipidemic activity of methanol extract of *Rivina humilis*. The administration of standard drug atorvastatin, medium and high doses of TVME, TVME and TVME could significantly reduce total cholesterol, triglycerides, LDL, VLDL and weight gain while in control group there was increase in these parameters were observed due to hyperlipidemia. There was also significant increase in the concentration of HDL in therapeutic animals was found compare to control animals ultimately suggesting the possible anti-hyperlipidemic activity of methanol extracts.

Conclusion

The results obtained from the present investigation suggesting that, methanol extract of *Rivina humilis* possess significant antihyperlipidemic potentials against high fat induced hyperlipidemia in wistar rats. Further studies are require to determine its mechanism of action and also to isolate and test specific constituent present in the methanol extract responsible for the benefits.

Conflict of interest

All authors are hereby declaring that there is no conflict of interest with respect to manuscript.

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