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Research Article

INVESTIGATION OF SCHIZOPHYLLUM COMMUNE
EXTRACT FOR ITS HYPOLIPIDEMIC ACTIVITY INVITRO
AND INVIVO MODELSHarsha Vardhana M G¹, Suresha B S², T Balasubramanian³¹Department of Pharmacology, Bharathi College of Pharmacy, Bharathinagara, Karnataka, India.²Professor and Head, Department of Pharmacology, Bharathi College of Pharmacy, Bharathinagara, Karnataka, India.³Principal, Department of Pharmacology, Bharathi College of Pharmacy, Bharathinagara, Karnataka, India.**Abstract:**

Cardiovascular diseases are major causes of morbidity and mortality worldwide and are strongly associated with abnormalities in lipid metabolism. The present study was undertaken to evaluate the hypolipidemic activity of the ethyl acetate extract of *Schizophyllum commune* (EAESC) in high-fat diet-induced hyperlipidemic Wistar albino rats. Thirty male Wistar rats were randomly divided into five groups of six animals each. Hyperlipidemia was induced by oral administration of vanaspati ghee and coconut oil in a 3:1 ratio at a dose of 3 mL/kg for 21 days. The treatment groups subsequently received EAESC at doses of 250 and 500 mg/kg, while atorvastatin (10 mg/kg) was used as the standard drug. At the end of the experimental period, serum lipid parameters including total cholesterol (TC), triglycerides (TG), high-density lipoprotein cholesterol (HDL-C), very-low-density lipoprotein (VLDL), low-density lipoprotein (LDL), and atherogenic index (AI) were evaluated. The hyperlipidemic control group showed marked increases in TC, TG, VLDL, LDL, and AI, along with a reduction in HDL-C. Treatment with EAESC produced improvements in the lipid profile, with the high dose (500 mg/kg) showing greater effects than the low dose (250 mg/kg). At the high dose, TC, TG, VLDL, LDL, and AI were reduced to 102.73 ± 3.02 , 105.05 ± 9.63 , 21.01 ± 1.93 , 44.23 ± 2.63 mg/dL, and 1.77 ± 0.14 , respectively, while HDL-C increased to 37.49 ± 2.00 mg/dL. These findings indicate that EAESC ameliorates high-fat diet-associated alterations in serum lipid parameters and demonstrates a dose-related hypolipidemic effect. The observed activity may be associated with the bioactive constituents present in *S. commune*.

Keywords: *Schizophyllum commune*, hypolipidemic activity, high-fat diet, Triglycerides, LDL, VLDL.

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INTRODUCTION:

Cardiovascular diseases (CVD), principally ischemic heart disease and stroke, remain the leading cause of mortality worldwide and a major contributor to disability and rising healthcare costs [1-4]. In 2010 alone, CVD was a primary cause of 15.6 million global deaths [1]. An important first step in tackling this global CVD epidemic is to determine the magnitude of the disease and risk factor burden at the regional and local population levels and to inform strategies for CVD and CVD risk factor prevention, treatment, and control [5,6]. Hyperlipidemia is outlined as an elevation of cholesterol, cholesterol esters, phospholipids or triglycerides. Abnormalities of plasma lipids can result in predisposition to coronary, cerebrovascular, and peripheral vascular arterial diseases.⁷

Medicinal mushrooms (MM) have an established history of use in traditional ancient therapies. Contemporary research has validated and documented much of the ancient knowledge. The interdisciplinary field of science that studies MMs has been developed and increasingly demonstrates potent and unique properties of compounds extracted from a range of mushroom species in the last three decades. Modern clinical practice in Japan, China, Korea, Russia, and several other countries rely on mushroom-derived preparations⁸⁻¹³.

Schizophyllum commune (Fr.) is a species of basidiomycetes belonging to *Schizophyllaceae* of *Agaricales*. It is one of the most widely distributed fleshy fungi. Schizophyllan, phenolics, flavonoids, terpenoid compounds, saponin, tannins, alkaloids, and ergosterol have also been found in *S. commune*. *S. commune*, which shows several bioactivities such as immunomodulator, anticancer, antitumor, anti-inflammatory, and antioxidant, wound healing, anti-pigmentation activities¹⁴.

MATERIALS AND METHODOLOGY:**Selecting and identifying plants:**

Schizophyllum commune mushroom was collected from northeast Karnataka, Bengaluru Karnataka. The mushroom material was identified and authenticated by Dr. Thejesh Kumar M.P. M.Sc., Ph. D, Department of Botany (PG), Bharathi College, Bharathinagara.

Preparation of ethyl acetate extract

The finely powdered *S. commune* mushroom was subjected to hot continuous extraction using ethyl acetate in a Soxhlet extractor. The resulting extract was used for the subsequent experimental investigation.

EXPERIMENTAL ANIMALS.

Healthy Wistar albino male rats of approximately same age (10 to 12 weeks), weighing between 100-120 g were taken for evaluating hypolipidemic activity. The animals were procured from Vertebrates, Magadi road, Bangalore. The animals were acclimatized by keeping them in animal house facility of Bharathi College of Pharmacy, Bharathinagara. They were housed in polypropylene cages containing bedding material as husk and maintained under standard husbandry conditions and 12 hrs light and 12 hrs dark cycle. They were fed with commercial pelleted rat chow with water *ad libitum*. The animals were maintained in accordance with the CCSEA guidelines.

Ethical approval

The hypolipidemic activity study was conducted after obtaining the approval from Institutional Animal Ethics Committee (IAEC) with a reference no. **BCP/IAEC/2025/04** dated **12/07/2025**.

Preparation of high fat diet:

Prepared mixture of 3:1 ratio of Vanaspati ghee and coconut oil (v/v) was used to induce the hyperlipidemia in experimental animals.

Preparation of suspension:

Schizophyllum commune and standard drug was suspended in 2% Acacia and administered orally to the animals because mushroom extract was insoluble in water.

Selection of mushroom extract dose:¹⁵

Since *Schizophyllum commune* is an edible mushroom and it is reported that the LD₅₀ value of *Schizophyllum commune* extract is 5000 mg/kg. We are considering the 1/10th of LD₅₀ value as high dose i.e. 500 mg/kg and 1/20th of LD₅₀ value as low dose i.e. 250mg/kg. To carry out the hypolipidemic activity of *Schizophyllum commune* extract.

Experimental Protocol Induced Hyperlipidemia

The 30 male Wistar rats were randomly divided into 5 groups each containing six animals (n=6). The experimental hyperlipidemia was induced by Vanaspati ghee and coconut oil (V/V) in the ratio of 3:1 at 3ml/kg body weight through oral route for 21 days induced then start the treatment by administration of ethyl acetate extract of *Schizophyllum commune* (EAESC) and standard for next 21 days, 10 min after the induced.

Group 1: Administered vehicle distilled water p.o., served as normal control.

Group 2: Administered High fat diet 3ml/kg p.o., served as hyperlipidemic control.

Group 3: Administered High fat diet 3ml/kg p.o. + Atorvastatin 10mg/kg b.w., suspended in distilled water p.o.

Group 4: Administered High fat diet 3ml/kg p.o. + EAESC 250 mg/kg b.w., suspended in distilled water p.o.

Group 5: Administered High fat diet 3ml/kg p.o. + EAESC 500 mg/kg b.w., suspended in distilled water p.o.

The study was carried out up to 6 weeks. Group 1: Animals received standard pellet diet and purified water for 24 hrs over a period of 6 weeks and was considered as normal control group. Group 2: Animals received standard pellet diet and high fat diet each day over a period of 6 weeks and was considered as hyperlipidemic control group. Group 3 animals received high fat diet and standard pellet diet each day over a period of 6 weeks. After 3 weeks, Atorvastatin (10 mg/kg) was given for next 3 weeks and considered as standard treatment group. Group 4 and Group 5 animals received standard pellet diet and high fat diet each day over a period of 6 weeks. After 3 weeks, ethyl acetate extract of *Schizophyllum commune* at low dose (250 mg/kg) and high dose (500 mg/kg) were administered to hyperlipidemic animals and considered as extract treatment groups.¹⁶

Blood collection and serum preparation

At the end of the experimental period, the animals were fasted overnight, blood was collected and serum was separated by centrifuging at 3000 rpm for 15 min and analyzed for various biochemical parameters like TGs, TC, HDL-C, LDL, VLDL and Atherogenic Index (AI).

Assay kits and reagents

The assay kits for cholesterol, HDL-C, triglycerides, were obtained from SLV Scientific Supplies #941, Suvarna nagar, M.K. Halli, Near Chamundeshwari temple, Gaddige main roa, Mysuru-570026, Karnataka, India. All other reagents used were of analytical grade.

➤ Estimation of total cholesterol

Serum total cholesterol was estimated by the enzymatic cholesterol oxidase-peroxidase (CHOD-POD) method. In this method, cholesterol esters are hydrolysed by cholesterol esterase to cholesterol and fatty acids. Cholesterol is subsequently oxidized by cholesterol oxidase, producing hydrogen peroxide, which reacts with 4-aminoantipyrine and phenol in the presence of peroxidase to form a colored quinoneimine product. The absorbance was measured at 505 nm. The cholesterol concentration was calculated using the standard supplied with the diagnostic kit^[17].

➤ Estimation of triglycerides

Serum triglycerides were estimated using the enzymatic glycerol phosphate oxidase-peroxidase (GPO-POD) method. Triglycerides are enzymatically hydrolysed to glycerol and fatty acids, followed by conversion of glycerol through a series of enzymatic reactions to generate hydrogen peroxide. The hydrogen peroxide participates in a color reaction to form a measurable quinoneimine dye. Absorbance was measured at 505 nm after incubation at 37°C^[17,18]. Triglyceride concentration was calculated using the supplied standard according to the following equation:

$$\text{Triglycerides (mg/dL)} = (\text{Absorbance of test} / \text{Absorbance of standard}) \times 200$$

➤ Estimation of HDL-C

Serum HDL-C was estimated using the PEGME/PVS direct HDL-C method. In this method, LDL, VLDL and chylomicrons are selectively masked by polyvinyl sulfonic acid (PVS) and polyethylene glycol methyl ether (PEGME), allowing selective measurement of HDL-associated cholesterol. The reaction was incubated at 37°C for 5 min, and absorbance was measured at 578 nm against the reagent blank. HDL-C concentration was calculated using the calibrator supplied with the diagnostic kit^[19].

➤ Calculation of LDL-C

LDL-C was estimated using the Friedewald equation:

$$\text{LDL-C (mg/dL)} = \text{TC} - \text{HDL-C} - (\text{TG}/5)$$

where TC, HDL-C and TG are expressed in mg/dL.

➤ Calculation of VLDL-C

VLDL-C was calculated from serum triglyceride concentration using the following equation:

$$\text{VLDL-C (mg/dL)} = \text{TG}/5$$

➤ Calculation of atherogenic index

The atherogenic index was calculated using the following formula:

$$\text{AI} = \frac{\text{TC} - \text{HDL-C}}{\text{HDL-C}}$$

where: TC = Total cholesterol, HDL-C = High-density lipoprotein cholesterol

Statistical Analysis

The experimental data were expressed as mean ± standard error of the mean (SEM) for six animals per group. Statistical analysis was performed using one-way analysis of variance (ANOVA), followed by Dunnett's multiple comparison test. A *P* value of <0.05 was considered statistically significant. The methodology classified *P*<0.01 as moderately significant, *P*<0.001 as highly significant, and *P*<0.0001 as very highly significant.

RESULT AND DISCUSSION:

Biochemical parameters(mg/dL)						
Groups	TC	HDL	TGs	VLDL	LDL	AI
Normal Control	101.09±7.27	40.11±2.92	97.54±4.92	19.51±0.98	41.47±7.68	1.60±0.26
HFD Control	141.01±2.44###	24.41±1.66##	144.90±5.84###	28.98±1.17##	87.62±3.79###	4.94±0.50###
Standard	99.34±0.68***	41.22±2.89**	102.63±3.43***	20.53±0.69**	37.59±5.21***	1.46±0.20***
S.commune Low dose	114.17±10.01*	34.12±2.55*	116.88±8.92*	23.38±1.78*	56.67±11.91*	2.48±0.48**
S.commune High dose	102.73±3.02**	37.49±2.00**	105.05±9.63**	21.01±1.93**	44.23±2.63***	1.77±0.147**

Table: 1. Effects of ethyl acetate extract of *Schizophyllum commune* on serum lipids at the end of 6 th week of study.

The biochemical analysis of serum lipid parameters in different experimental groups revealed significant alterations in lipid metabolism following administration of the ethyl acetate extract of *Schizophyllum commune*. The results are summarized in Table 1. The hyperlipidemic control (HC) group exhibited a marked elevation in serum triglycerides (TG), total cholesterol (TC), low-density lipoprotein (LDL-C), and very low-density lipoprotein (VLDL) levels, accompanied by a significant reduction in high-density lipoprotein cholesterol (HDL-C) when compared with the normal control group. These findings confirm the successful induction of hyperlipidemia in the experimental model.

Treatment with the *ethyl acetate extract of Schizophyllum commune* produced a dose-dependent improvement in serum lipid parameters in hyperlipidemic rats. The HFD control group showed marked dyslipidemia, with elevated TC (141.01 ± 2.44 mg/dL), TG (144.90 ± 5.84 mg/dL), VLDL (28.98 ± 1.17 mg/dL), LDL (87.62 ± 3.79 mg/dL), and atherogenic index (4.94 ± 0.50),

together with a reduced HDL-C level (24.41 ± 1.66 mg/dL), compared with the normal control group. Treatment with the standard drug produced near-normalization of the lipid profile, with reductions in TC (99.34 ± 0.68 mg/dL), TG (102.63 ± 3.43 mg/dL), VLDL (20.53 ± 0.69 mg/dL), LDL (37.59 ± 5.21 mg/dL), and AI (1.46 ± 0.20), accompanied by an increase in HDL-C (41.22 ± 2.89 mg/dL). This response confirms the effectiveness of the experimental model and provides a reference for evaluating the lipid-modulating effect of the *S. commune* extract.

The low-dose *S. commune* extract treatment showed a moderate improvement in the lipid profile compared with the HFD control. TC, TG, VLDL, and LDL were reduced to 114.17 ± 10.01, 116.88 ± 8.92, 23.38 ± 1.78, and 56.67 ± 11.91 mg/dL, respectively. HDL-C increased to 34.12 ± 2.55 mg/dL, while the atherogenic index decreased to 2.48 ± 0.48. These changes indicate that the low dose exerted a beneficial hypolipidemic effect, although the values did not reach the levels observed in the normal and standard-treated groups.

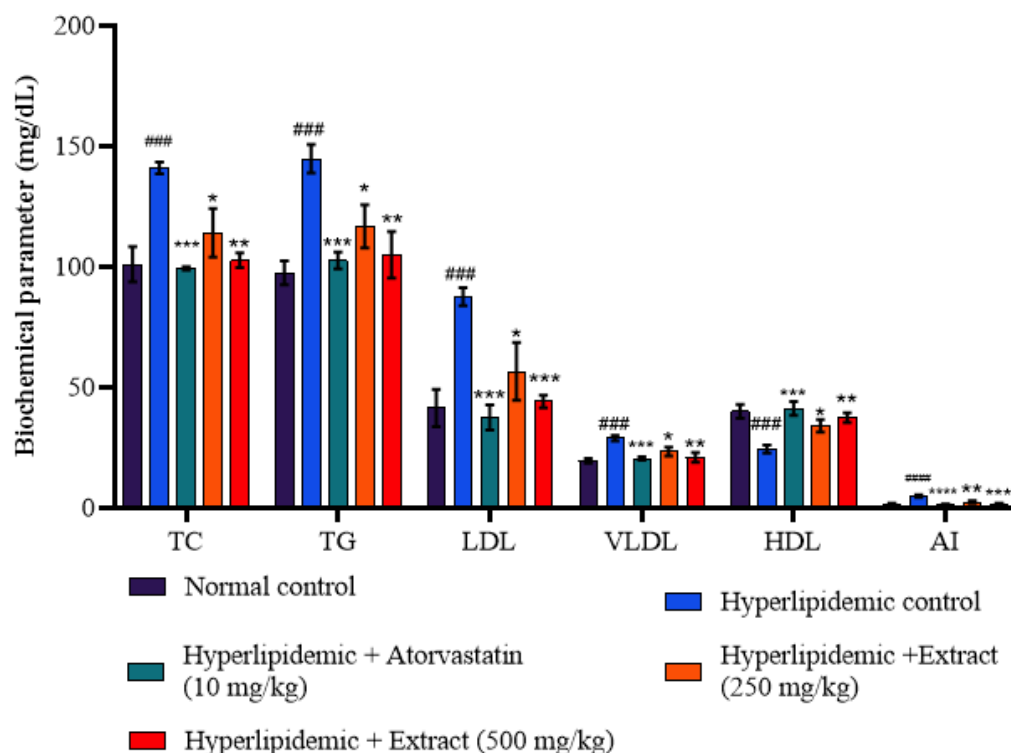


Figure 1: Graph showing mean serum lipid parameters in 5 groups at the end of 6 th week.

The high-dose *S. commune* extract produced a more pronounced improvement in serum lipid parameters. TC, TG, VLDL, and LDL decreased to 102.73 ± 3.02 , 105.05 ± 9.63 , 21.01 ± 1.93 , and 44.23 ± 2.63 mg/dL, respectively, while HDL-C increased to 37.49 ± 2.00 mg/dL. The atherogenic index was also substantially reduced to 1.77 ± 0.147 , indicating an improvement in the overall lipid profile. The greater improvement observed with the high dose compared with the low dose suggests a dose-related lipid-modulating effect of the *S. commune* extract.

Overall, the findings indicate that ethyl acetate extract of *Schizophyllum commune* ameliorated HFD-associated alterations in serum lipid parameters, as evidenced by reductions in TC, TG, VLDL, LDL and AI, together with an increase in HDL-C. The high-dose treatment produced changes closer to those of the normal and standard-treated groups, suggesting a potential hypolipidemic and cardioprotective effect of the extract. These effects may be associated with bioactive constituents present in *S. commune*.

CONCLUSION:

The present study demonstrated that the ethyl acetate extract of *Schizophyllum commune* possesses promising hypolipidemic activities. In the in-vivo high-fat diet model, administration of the ethyl acetate extract at 250 and 500 mg/kg improved the altered serum lipid profile by reducing total cholesterol, triglycerides, LDL-C,

VLDL-C and atherogenic index, while increasing HDL-C. The 500 mg/kg dose generally produced greater improvement than the 250 mg/kg dose. These findings indicate that *S. commune* ethyl acetate extract has potential hypolipidemic activity in the experimental model used. Overall, the findings support the potential of *Schizophyllum commune* as a natural source with lipid-modulating.

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