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Original Article

ANTIHYPERLIPIDEMIC ACTIVITY OF METHANOLIC EXTRACT OF LEAVES OF BAMBUSA BAMBOS DRUCE AGAINST POLOXAMER-407 INDUCED HYPERLIPIDEMIA IN RATS

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ABSTRACT

Objective: The aim of present study was to investigate a dose-dependent antihyperlipidemic effect of methanolic extract of *Bambusa bambos* Druce. (Family: Graminae) (MEBB) in Poloxamer-407 induced hyperlipidemia in rats at three different dose levels i. e. 100 mg/kg, 200 mg/kg and 300 mg/kg.

Methods: Animals were divided randomly into 6 groups: Group I (Vehicle Control), Group II (Disease Control), Group III (100 mg/kg MEBB), Group IV (200 mg/kg MEBB), Group V (300 mg/kg MEBB) and Group VI (Standard Control).

Results: Groups treated with the MEBB showed significant ($p<0.01$) decrease in serum total cholesterol, triglycerides, LDL-cholesterol VLDL-cholesterol, hepatic cholesterol and hepatic triglycerides with the concomitant increase in serum HDL-cholesterol levels. The extract also showed significant ($p<0.01$) antioxidant activity in liver tissue homogenate. Atherogenic indices also showed significant ($p<0.05$, $p<0.01$) decrease in treatment groups as compared to disease control group. The most effective dose was found to be 300mg/kg MEBB.

Conclusion: These results suggest that methanolic extract of Bambusa bambos leaves possess significant antihyperlipidemic activity.

Keywords: Antihyperlipidemic activity, Poloxamer-407, Bambusa bambos, Atherosclerosis.

INTRODUCTION

Hyperlipidemia characterised by hypercholesterolemia represents a determinant for development of atherosclerosis and is an important risk factor for cardiovascular diseases [1, 2]. According to World Health Organisation (WHO) report, blood cholesterol contributes to approximately 56% cases of cardiovascular diseases worldwide and about 4.4 million deaths each year [3]. Hyperlipidemia is a metabolic disorder, specifically characterised by alterations in serum lipid and lipoprotein profile due to increased concentrations of Total Cholesterol (TC), Low Density Lipoprotein Cholesterol (LDL-C), Very Low Density Lipoprotein Cholesterol (VLDL-C) and Triglycerides (TG) with a concomitant decrease in concentrations of High Density Lipoprotein Cholesterol (HDL-C) in blood circulation.

The use of herbal drugs or phytochemicals has been rapidly increasing worldwide because they are less damaging than synthetic drugs thus improving patient compliance even on long term use.

Bambusa bambos Druce. (Family: Graminae) is a species of clumping bamboo commonly known as "Indian thorny bamboo" in English and "Vanshi" in Sanskrit. It mainly occurs throughout India, Srilanka, Malaya, Peru and Myanmar [4]. According to Ayurveda text, the plant is claimed to be *medhoghna* (removing or destroying excessive fat) [5]. Charakha prescribed decoction of leaves or seeds in treatment of excessive fat [6]. Fruit and seeds act on medhadhatu and are useful in fat metabolism and obesity [7]. The other traditional uses of the plant are as emmenagogue, anti-inflammatory, astringent, anti-spasmodic, tonic and to check cattle in diarrhoea [4,6]. Though the plant and its extracts have been used extensively in traditional medicine, no such scientific evidence for its antihyperlipidemic activity is available in established scientific journals of repute. Hence the present study is aimed at investigating the antihyperlipidemic potential of leaves of *Bambusa bambos*.

MATERIALS AND METHODS

Collection of plant material

The leaves of *Bambusa bambos* (4kgs) were procured from Keshav Shrusti, Thane, India in the month of July. The leaves were authenticated by Dr. Harshad Pandit, Department of Botany, Guru Nanak Khalsa College, Mumbai, India and a voucher specimen (accession number: ak/170912) was deposited for future reference.

Preparation of plant extract

The leaves of the plant were dried in shade, powdered and passed through 40 mesh sieve. Dried powder (50g) was extracted with methanol in Soxhlet apparatus for 72Hrs. The extract was evaporated under vacuum in a rotary evaporator and dried completely in a desiccator and weighed. The yield of the extract was 4.5g. For dosing, the methanolic extract of *Bambusa bambos* leaves (MEBB) was uniformly suspended in 0.5% Carboxy methyl cellulose (CMC) dissolved in water and administered orally *(p. o.).*

Experimental animals

Adult male Sprague Dawley rats weighing 150-200g were purchased from Bharat Serums and Vaccines Ltd. Thane, Mumbai. The study was conducted after obtaining the clearance for the experimental protocol (IAEC/PR/2012/02) from Institutional Animal Ethics Committee (IAEC), Bharati Vidyapeeth's college of Pharmacy, Navi Mumbai, India. Rats were housed in animal house of Bharati Vidyapeeth's college of Pharmacy, Navi Mumbai, India. Three rats per cage were housed in polypropylene cages (32.5 ×21×14) cm lined with raw husk which was renewed every 48 hours. The animal house was maintained at an average temperature (24.0°C$\pm$ 2°C) and 30-70% RH, with 12-Hr light-dark cycle (lights on 8.00a. m to 8.00p. m). Animals received humane care and were fed with commercial pellet diet (Amrut Laboratory, Mumbai, India) and tap water ad-libitum. The animals were acclimatized for one week before the start of the experiment. The experiments were carried out in accordance with the guidelines set by Committee for Purpose of Control and Supervision on Experiments on Animals (CPCSEA)

In vivo antihyperlipidemic activity-Poloxamer-407 induced hyperlipidemia in rats [8]

The antihyperlipidemic effect of MEBB was examined in Poloxamer-407 induced hyperlipidemia in rats.

Animals were divided into six groups, each consisting of six rats and were treated as follows

Group I and group II received 0.5% CMC solution in water (5 ml/kg/day). Group III was administered Atorvastatin 10 mg/kg/day. Groups IV, V and VI received MEBB at doses of 100, 200 and 300 mg/kg/day. The vehicle (0.5%CMC), Atorvastatin and test drugs were administered orally for 12 consecutive days.

20%w/w Poloxamer-407 solution for i.p. injection was prepared by dissolving the powder in cold saline and placing the solution on ice overnight to facilitate dissolution of polymer according to the "cold method" of incorporation [9]. On 13th day, experimental hyperlipidemia was induced by intraperitonial injection of Poloxamer-407 (1 gm/kg). Animals were fasted for 16Hr prior to induction of hyperlipidemia. Blood was withdrawn from retro-orbital plexus at 0Hr, 24Hr and 48Hr after administration of Poloxamer-407.

The blood samples were allowed to clot at room temperature for 20-25 min and centrifuged for 20 min at 3000 rpm at 4°C. The supernatant clear serum thus obtained was stored at -20°C until the completion of biochemical analysis. After 48Hr of administration of Poloxamer-407, animals were sacrificed by CO_2 overdose. Livers were excised immediately and washed with ice cold saline, blotted with dry filter paper and liver weight was recorded. Livers were processed further for determination of antioxidant activity in liver tissue homogenate and hepatic lipids.

Biochemical analysis

All samples were used for following biochemical investigations. The blood serum under this model has been analysed for the marker parameters such as total cholesterol (TC), High density lipoprotein cholesterol (HDL-C) and triglycerides (TG). Serum total cholesterol and triglycerides were estimated by enzymatic methods of CHOD-PAP and GPO-Trinder method respectively [10, 11]. Estimation of HDL-C was done by precipitation method [12]. All parameters were analysed by ERBA Autoanalyser (Spectrophotometric) with standard biochemical kits (ERBA Diagnostic Mannheim GmbH, Germany). Serum concentrations of Very low density lipoprotein-Cholesterol (VLDL-C) and Low density lipoprotein-Cholesterol was calculated using Friedewald's formula [13].

The atherogenic indices calculated were

Atherosclerosis Index (A. I) [14] = LDL-C / HDL-C

Cardiac Risk Ratio (C. R. R) [15] =TC/HDL-C

Atherogenic Coefficient (A. C) [16] = TC-HDL-C/HDL-C

Antioxidant activity in liver tissue homogenate

About 1g of liver tissue was homogenised in 10 ml of 0.1M phosphate buffer pH 7.4 to form 10%w/v liver tissue homogenate. The prepared liver tissue homogenate were centrifuged at 3500 rpm for 15 min. and supernatant was used for the determination of various antioxidant parameters like lipid peroxidation [17], reduced glutathione [18] and catalase [19].

Estimation of hepatic lipids

Hepatic lipids were extracted using method of Folch et al. (1957) [20]. Briefly, 1 gm of liver was homogenised in 20 ml of solvent mixture (Chloroform: Methanol in ratio 2:1). The homogenate was filtered and washed with 4 ml of saline solution. The mixture was vortexed for few seconds and then centrifuged at 2000 rpm for separation of the two phases. The lower phase was used for estimation of hepatic lipids.

Statistical analysis

All the experimental results were expressed as mean ±SEM. Data were analysed by One-way Analysis of Variance (ANOVA) followed by Dunnett's test ($^{n.s}$p>0.05, p*<0.05, p**<0.01)

RESULTS

Biochemical analysis

Effect of MEBB on serum cholesterol

The groups treated with MEBB and Standard Control (Atorvastatin) showed significant (p<0.01) decrease in Poloxamer-407 induced elevation of serum total cholesterol when compared to disease control (Table 1). 100 mg/kg MEBB reduced serum cholesterol level by 13.45% and 8.47% at 24Hr and 48Hr respectively. 200 mg/kg MEBB reduced serum cholesterol level by 23.65% and 25.94% at 24Hr and 48Hr respectively. 300 mg/kg MEBB reduced serum cholesterol level by 41.94% and 50.01% at 24Hr and 48Hr respectively. Standard control (Atorvastatin 10 mg/kg) reduced serum cholesterol by 49.52% and 59.65% at 24Hr and 48Hr respectively.

Effect of MEBB on serum triglycerides

The groups treated with 200 mg/kg MEBB, 300 mg/kg MEBB and Standard control (Atorvastatin) demonstrated significant (p<0.01) decrease in Poloxamer-407 induced elevation of serum triglyceride when compared to disease control (Table 1). 100 mg/kg MEBB did not show significant reduction in serum triglyceride level when compared to disease control. 200 mg/kg MEBB reduced serum triglyceride level by 26.86% and 30.71% at 24Hr and 48Hr respectively. 300 mg/kg MEBB reduced serum triglyceride level by 48.18% and 58.06% at 24Hr and 48Hr respectively. Standard control (Atorvastatin 10 mg/kg) reduced serum triglyceride by 53.38% and 65.57% at 24Hr and 48Hr respectively.

Effect of MEBB on serum HDL-Cholesterol (HDL-C)

The groups treated with MEBB and Standard Control (Atorvastatin) showed significant (p<0.01) increase in Poloxamer-407 induced depletion of serum HDL-C when compared to disease control (Table 1). 100 mg/kg MEBB increased serum HDL-C by 25.22% and 25.99% at 24Hr and 48Hr respectively. 200 mg/kg MEBB increased serum HDL-C level by 62.80% and62.21% at 24Hr and 48Hr respectively. 300 mg/kg MEBB increased serum HDL-C level by 93.74% and 93.58% at 24Hr and 48Hr respectively. Standard control (Atorvastatin 10 mg/kg) increased serum HDL- C levels by 116.05% and 113.82% at 24Hr and 48Hr respectively.

Table 1: Effect of MEBB on serum Total cholesterol, Triglycerides and HDL-C in poloxamer-407 induced hyperlipidemia in rats

	Baseline (0Hr)			24Hr			48Hr		
Group	**TC (mg/dl)**	**TG (mg/dl)**	**HDL-C (mg/dl)**	**TC (mg/dl)**	**TG (mg/dl)**	**HDL-C (mg/dl)**	**TC (mg/dl)**	**TG (mg/dl)**	**HDL-C (mg/dl)**
Vehicle control	51.29± 4.72	62.65± 3.84	19.87± 0.36	45.81± 1.26	60.95± 0.055	20.15± 0.29	45.13± 1.14	61.78± 0.53	19.57± 0.29
Disease Control	56.75± 3.41	60.60± 2.88	20.47± 0.66	467.00± 9.09	428.33± 0.67	12.77± 0.39	398.00± 3.49	354.35± 5.49	13.39± 0.35
Test I (100 mg/kg MEBB)	61.61± 5.31	64.93± 0.92	18.71± 0.40	404.20± 9.83**	421.31± 0.89$^{n.s}$	15.99± 0.096**	364.27± 4.01**	344.81± 3.86$^{n.s}$	16.87± 0.28**
Test II (200 mg/kg MEBB)	62.96± 0.64	74.21± 1.03	18.73± 0.43	356.56± 4.99**	313.30± 3.80**	20.79± 0.23**	294.75± 11.54**	245.53± 9.20**	21.72± 0.18**
Test III (300 mg/kg MEBB)	58.09± 5.49	63.32± 0.54	18.99± 0.37	271.16± 4.33**	221.97± 4.23**	24.74± 0.30**	198.97± 3.83**	148.61± 2.30**	25.92± 0.31**
Standard control (Atorvastatin 10 mg/kg)	66.01± 0.61	48.52± 1.18	19.12± 0.29	235.71± 5.10**	199.68± 2.43**	27.59± 0.24**	160.59± 3.54**	121.99± 4.09**	28.63± 0.36**

N=6 animals in each group. Values (mean ± SEM) are expressed in mg/dl.

Treatment groups are compared with disease control group; One way ANOVA followed by Dunnett's test ($^{n.s}$p>0.05, p*<0.05, p**<0.01), n. s = non-significant.

TC: Total cholesterol, TG: Triglycerides, HDL-C: High density lipoprotein cholesterol

Effect of MEBB on serum LDL-Cholesterol (LDL-C)

The groups treated with MEBB and Standard control (Atorvastatin) showed significant (p<0.01) decrease in Poloxamer-407 induced elevation of serum LDL-C when compared with disease control (Table 2). 100 mg/kg MEBB reduced LDL-C levels by 19.49% and 11.84% at 24Hr and 48Hr respectively. 200 mg/kg MEBB reduced serum LDL-levels by 29.13% and 27.37% at 24Hr and 48Hr respectively. 300 mg/kg MEBB reduced serum LDL-C levels by 46.65% and 53.70% at 24Hr and 48Hr respectively. Standard control (Atorvastatin) reduced serum LDL-C levels by 55.77% and 66.24% at 24Hr and 48Hr respectively.

Effect of MEBB on serum VLDL-Cholesterol (VLDL-C)

The groups treated with 200 mg/kg MEBB, 300 mg/kg MEBB and Standard control (Atorvastatin) showed significant (p<0.01) reduction in Poloxamer-407 induced elevation of serum VLDL-C levels when compared to disease control (Table 2). 100 mg/kg MEBB did not show significant reduction in serum VLDL-C level when compared to disease control. 200 mg/kg MEBB reduced serum VLDL-C levels by 26.86% and 30.70% at 24Hr and 48Hr respectively. 300 mg/kg MEBB reduced serum VLDL-C by 49.35% and 58.06% at 24Hr and 48Hr respectively.

Standard control (Atorvastatin) reduced serum VLDL-C levels by 53.38% and 65.57% at 24Hr and 48Hr respectively.

Table 2: Effect of MEBB on serum LDL-C and VLDL-C in poloxamer-407 induced hyperlipidemia in rats

	Baseline (0Hr)		24Hr		48Hr	
Group	**LDL-C (mg/dl)**	**VLDL-C (mg/dl)**	**LDL-C (mg/dl)**	**VLDL-C (mg/dl)**	**LDL-C (mg/dl)**	**VLDL-C (mg/dl)**
Vehicle control	19.67±4.11	12.53±0.77	17.79±1.07	12.19±0.01	16.13±0.92	12.36±0.11
DiseaseControl	25.40±3.08	12.12±0.58	389.30±5.76	85.67±0.14	318.63±1.98	70.87±1.09
Test I(100 mg/kg MEBB)	30.05±6.00	12.99±0.19	313.41±2.93**	84.26±0.18$^{n.s}$	290.89±3.19**	68.89±0.64$^{n.s}$
Test II(200 mg/kg MEBB)	29.33±1.40	14.85±0.21	275.90±4.81**	62.66±0.76**	231.42±3.37**	49.11±1.84**
Test III(300 mg/kg MEBB)	26.32±6.25	12.67±0.11	207.71±1.36**	43.39±0.85**	147.51±2.53**	29.72±0.46**
Standard control(Atorvastatin 10 mg/kg)	36.75±0.69	9.71±0.24	172.30±3.03**	39.94±0.49**	107.56±3.10**	24.40±0.81**

N=6 animals in each group. Values (mean ± SEM) are expressed in mg/dl.

Treatment groups are compared with disease control group; One way ANOVA followed by Dunnett's test ($^{n.s}$p>0.05, p*<0.05, p**<0.01), n. s = non-significant.

LDL-C: Low density lipoprotein cholesterol, VLDL-C: Very low density lipoprotein cholesterol

Table 3: Effect of MEBB on Atherogenic indices in Poloxamer-407 induced hyperlipidemia in rats

	Baseline (0Hr)			24Hr			48Hr		
Groups	**A. I**	**C. R.R**	**A. C**	**A. I**	**C. R. R**	**A. C**	**A. I**	**C. R. R**	**A. C**
Vehiclecontrol	1.04±0.22	2.69±0.27	1.69±0.27	0.95±0.01	2.59±0.14	1.59±0.14	0.86±0.07	2.51±0.08	1.51±0.08
Disease control	1.33±0.18	2.96±0.22	1.96±0.27	31.3±1.07	38.67±1.51	37.67±1.51	24.16±0.89	30.54±1.14	29.54±1.14
Test I(100 mg/kg MEBB)	1.64±0.40	3.33±0.45	2.33±0.45	19.60±0.30*(37.38)	25.88±0.34*(33.07)	24.88±0.24*(33.95)	16.66±0.46*(31.04)	21.78±0.57*(28.68)	20.78±0.57*(29.65)
Test II(200 mg/kg MEBB)	1.57±0.16	3.36±0.20	2.36±0.20	13.28±0.38**(57.57)	17.30±0.44**(55.26)	16.30±0.44**(56.73)	10.66±0.23**(55.88)	13.92±0.34**(54.42)	12.92±0.34**(56.26)
Test III(300 mg/kg MEBB)	1.40±0.40	3.06±0.43	2.06±0.43	8.40±0.15**(73.16)	11.23±0.22**(70.96)	10.23±0.22**(72.84)	5.70±0.16**(76.40)	7.99±0.31**(73.84)	6.99±0.31**(76.33)
StandardControl(Atorvastatin 10 mg/kg)	1.880.07	3.37±0.08	2.27±0.08	6.24±0.16**(80.06)	8.76±0.26**(73.84)	7.76±0.26**(79.40)	3.76±0.15**(84.44)	5.62±0.19**(81.60)	4.62±0.19**(63.83)

N=6 animals in each group. Values are expressed as (mean ± SEM)

Treatment groups are compared with disease control group; One way ANOVA followed by Dunnett's test ($^{n.s}$p>0.05, p*<0.05, p**<0.01), n. s=non-significant. Number in parenthesis indicates % decrease in the respective indices when compared to disease control group.

A. I: Atherosclerosis index, C. R. R: Cardiac Risk Ratio, A. C: Atherogenic coefficient

Effect of MEBB on Atherogenic indices

The groups treated with MEBB and Standard control (Atorvastatin) showed significant (p<0.05, p<0.01) decrease in Atherosclerosis index (A. I), Cardiac Risk Ratio (C. R. R) and Atherogenic Coefficient (A. C) when compared to disease control (Table 3).

Antioxidant activity in liver tissue homogenate

The groups treated with MEBB and Standard control (Atorvastatin) showed significant (p<0.01) reduction in Poloxamer-407 induced lipid peroxidation in liver (Table 3). The groups treated with MEBB and Standard control (Atorvastatin) also showed significant (p<0.01) elevation of Catalase in liver when compared to disease control (Table 3). Only 200mg/kg MEBB, 300 mg/kg MEBB and Standard control (Atorvastatin) demonstrated significant (p<0.01) increase in levels of reduced glutathione in liver when compared to disease control (Table 4).

Estimation of hepatic lipids

The groups treated with MEBB and Standard control (Atorvastatin) demonstrated significant (p<0.01) decrease in Hepatic total cholesterol and Hepatic triglycerides when compared to disease control (Table 5).

Hepatic cholesterol was decreased by 43.85%, 67.22%, 95.11% and 96.74% by 100 mg/kg MEBB, 200 mg/kg MEBB, 300 mg/kg MEBB and Standard control (Atorvastatin) respectively. Hepatic triglycerides were decreased by 36.58%, 63.85%, 81.71% and 94.68% by 100 mg/kg MEBB, 200 mg/kg MEBB, 300 mg/kg MEBB and Standard control (Atorvastatin) respectively.

Table 4: Antioxidant activity of MEBB in liver tissue homogenate

Group	Malondialdehyde (µMol/g wt. of wet liver tissue)	Reduced glutathione (mMol/g wt. of wet liver tissue)	Catalase (mMol/g wt. of wet liver tissue)
Vehicle control	3.18±0.33	2.41±0.01	79.42±0.68
Disease Control	20.97±0.12	1.74±0.0067	57.92±0.54
Test I(100 mg/kg MEBB)	16.51±0.18**(25.07)↓	1.86±0.01$^{n.s}$	60.86±0.27**(13.72) ↑
Test II(200 mg/kg MEBB)	8.00±0.13**(72.90) ↓	2.14±0.001**(59.70)↑	63.03±0.35**(23.77) ↑
Test III(300 mg/kg MEBB)	5.36±0.14**(87.75) ↓	2.26±0.0058**(77.61) ↑	68.07±0.32**(47.21) ↑
Standard Control(Atorvastatin 10 mg/kg)	4.75±0.21**(91.17) ↓	2.57±0.20**(123.88) ↑	71.43±0.23**(62.84) ↑

N=6 animals in each group. Values are expressed as (mean ± SEM)

Treatment groups are compared with disease control group; One way ANOVA followed by Dunnett's test ($^{n.s}$p>0.05, p*<0.05, p**<0.01), n. s. = non-significant.

Number in parenthesis indicates % increase or decrease in the respective parameters when compared to disease control group. ↓ denotes decrease; ↑ denotes increase in the respective parameters.

Table 5: Effect of MEBB on Hepatic lipids in Poloxamer-407 induced hyperlipidemia in rats

Group	Hepatic cholesterol (mg/g)	Hepatic triglycerides (mg/g)
Vehicle Control	7.26±0.16	10.14±0.09
Disease Control	31.82±1.13	42.67±0.63
Test I(100 mg/kg MEBB)	21.05±0.24**(43.85)	30.77±0.37**(36.58)
Test II(200 mg/kg MEBB)	15.31±0.20**(67.22)	21.90±0.54**(63.85)
Test III(300 mg/kg MEBB)	8.46±0.04**(95.11)	16.09±0.08**(81.71)
Standard Control(Atorvastatin 10 mg/kg)	8.06±0.06**(96.74)	11.87±0.36**(94.68)

N=6 animals in each group. Values are expressed as (mean ± SEM)

Treatment groups are compared with diseased control group; One way ANOVA followed by Dunnett's test ($^{n.s}$p>0.05, p*<0.05, p**<0.01), n. s = non-significant. Number in parenthesis indicates % decrease in the hepatic lipids when compared to disease control group.

DISCUSSION

The present study was conducted to determine the antihyperlipidemic activity of methanolic extract of leaves of *Bambusa bambos* in Poloxamer-407 induced hyperlipidemia in rats. Poloxamer-407 is a hydrophilic, non-toxic surfactant with low degree of toxicity. Wout and co-workers have reported that single intraperitonial injection of Poloxamer-407 causes significant elevations in serum cholesterol and triglyceride levels [21].

Studies demonstrate that Polxamer-407 activates 3-Hydroxy-3-methylglutaryl coA reductase [22] (HMG-coA reductase), a rate liming enzyme in synthesis of cholesterol from mevalonate thus increasing serum cholesterol levels [23]. Administration of Poloxamer-407 also inhibits Cholesterol 7α-hydroxylase [24]which catalyses synthesis of bile acid from cholesterol called "classic pathway" thereby limiting the clearance of cholesterol from the body [25,26]. Administration of MEBB showed significant (p<0.01) decrease in serum cholesterol levels as compared to disease control group. The observed effect may be due to inhibition of HMG-coA reductase and thus decreasing cholesterol synthesis or activation of Cholesterol 7α-hydroxylase and thereby increasing the synthesis of bile acid from cholesterol and its clearance from the body.

The predominant mechanism by which Poloxamer-407 produces increase in circulating triglyceride can be attributed to reduction in the rate at which triglycerides are hydrolysed to free fatty acids due to inhibition of heparin-releasable Lipoprotein lipase (LPL) [27]. Administration of 200 mg/kg and 300 mg/kg MEBB showed significant (p<0.01) decrease in serum triglyceride level as compared to disease control. The observed effect may be due to activation of LPL by MEBB.

MEBB also decreased serum LDL-C and VLDL-C levels significantly (p<0.01) in comparison with disease control group. Low LDL-C levels may be attributed to decrease in synthesis of its precursor i. e. VLDL-C. According to oxidative-modification hypothesis, LDL-C accumulates in the subordinated extracellular space of arteries and is oxidised to form oxidised LDL-C which is highly atherogenic [28-30]. Therefore low levels of LDL-C can retard the progression of atherosclerosis.

According to "HDL hypothesis" serum HDL-C levels are inversely linked with the risk of development of atherosclerosis [31]. HDL-C may exert is anti-atherosclerotic effect by scavenging excess cholesterol from peripheral vasculature and transporting it to liver, where it is excreted in biliary system ("reverse cholesterol transport) [32,33]. HDL-C inhibits the oxidation of LDL-C in artery wall and also scavenges cholesterol from foam cells thus limiting the inflammatory process that underlines atherosclerosis [34]. Atheroprotective nature of HDL-C can also be attributed to its wide spectrum of activities viz. antioxidant, anti-inflammatory, anti-apoptotic, anti-thrombotic, anti-infective and vasodilatory effects [35-37]. Therefore administration of MEBB may also exert anti-atherosclerotic effect by elevation of serum HDL-C levels.

Oxidative stress is one of the causative factors that link hyperlipidemia with the pathogenesis of atherosclerosis. This stress results from an imbalance between production of free radicals and effectiveness of antioxidant defence system [38]. The main targets for the free radicals are polyunsaturated fatty acids in membrane of lipids causing lipid peroxidation which may lead to disorganisation of cell structure and function [39]. Lipid peroxidation, a free radical mediated process has been accepted to be one of the primary causes in development of cholesterol induced diseases [40]. Malondialdehyde (MDA) a stable secondary product of lipid peroxidation cascade is widely used as a marker for measurement of lipid peroxidation [41]. Reduced glutathione and Catalase play unique role in cellular defence system by their ability to scavenge free radicals, as such depletion of reduced glutathione and catalase

increases vulnerability to free radicals [42-44]. Treatment with MEBB significantly ($p<0.01$) decreased hepatic MDA levels while increasing hepatic reduced glutathione and catalase levels. Hence administration of MEBB showed significant antioxidant activity in liver tissue homogenate and may have protective role against oxidative stress induced atherosclerosis.

Atherogenic indices are powerful indicators of the risk of development of heart disease: higher the value, higher the risk of development of cardiovascular disease and vice versa [45,46]. Treatment with MEBB lowered atherogenic indices significantly ($p<0.05$, $p<0.01$) thus decreasing the risk of development of cardiovascular disease. From the results obtained in this study, it can be concluded that methanolic extract of leaves of *Bambusa bambos* possess antihyperlipidemic activity. However further studies are required to elucidate the exact mechanism(s) of action.

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CONFLICT OF INTERESTS

Declared None

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