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PHARMACOLOGICAL EVALUATION OF ANTI ATHEROSLEROTIC ACTIVITY OF POLYGONUMGLABRUM IN ANIMAL MODELS

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ABSTRACT

Objective: To investigate the anti-atherosclerotic activity of ethanol extract of Polygonum galbrum in male Wistar rats. **Material and methods:** In that model regarding atherosclerosis, 30 adult male wistar rats were always broken in 5 groups. Group-1 then Group-2 served namely untreated yet model controls respectively, whilst Group-3, four or 5 were the redress agencies which have been simultaneously treated including standard, 200 and four hundred mg/kg eliminate respectively alongside along High Fat Diet. On remaining day, blood samples because biochemical parameters, have been obtained below inhaled diether anaesthesia. **Results:** HFD induced atherosclerosis as evidenced by marked elevation in cholesterol, Triglycerides, LDL, VLDL and decrease in HDL ranges. Co-management of extract with HFD decreased upward thrust ldl cholesterol, Triglycerides, LDL, VLDL and increase in HDL tiers. **Conclusion:** It changed into observed that the ethanol extract of Polygonum galbrum conferred Anti- atherosclerotic interest by way of biochemical remark towards HFD triggered atherosclerosis in rats. Within the near destiny could constitute a result in discovery of a singular drug for remedy of drug brought on atherosclerosis.

KEYWORDS

Polygonumglabrum and Anti- atherosclerotic activity.

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INTRODUCTON

Herbal drugs generally observed as seasonerism or phytotherapy, is that the use of herbs for his or her therapeutic or healthful price. Associate herb could be a plant or natural object valued for its healthful, aromatic quality. Herb plants manufacture and contain a spread of chemical substances that impact the body. Herbalists use the leaves, flowers, stems, berries, and roots of plants to forestall, relieve, and

treat unwellness. From a "scientific" perspective, several seasoner treatments are thought-about experimental. The truth is, however, that seasoner drugs incorporates a long and revered history. Several acquainted medications of the 20th century were developed from ancient healing traditions that treated health issues with specific plants. Today, science has isolated the healthful properties of an oversized variety of botanicals, and their healing elements are extracted and analyzed. Several plant elements are currently synthesized in massive laboratories to be used in pharmaceutical preparations. As an example, periwinkle plant derivative (an antitumour drug), digitalis (a heart regulator), and bronchodilator (a medicine accustomed decrease metastasis congestion) were all originally discovered through analysis on plants[1,2].

Rather than employing a whole plant, pharmacologists determine, isolate, extract, and synthesize individual elements, so capturing the active properties. This may produce issues, however. Additionally to active ingredients, plants contain minerals, vitamins, volatile oils, glycosides, alkaloids, bio-flavanoids, and alternative substances that are necessary in supporting a selected herb's healthful properties. These parts additionally offer a vital natural safeguard Isolated or synthesized active compounds will become virulent in comparatively tiny doses; it always takes a far bigger quantity of an entire herb, with all of its elements, to succeed in a virulent level. Herbs are medicines, however, and that they will have powerful effects. They must not tee taken gently. The suggestions for flavourer treatments during this book don't seem to be supposed to substitute for consultation with a professional health care professional, however rather to support and assist you in understanding and dealing along with your physician's recommendation[3,4].

HERBAL MEDICINES TODAY

The World Health Organization (WHO) estimates that four billion individuals, eightieth of the planet population, presently use flavoring drugs for a few side of primary health care. Flavoring drugs could be a major part altogether autochthonic peoples' ancient drugs and a typical component in Ayurveda, homeopathic, naturopathic, ancient oriental, and Native yank Indian drugs. United Nations agency notes that of 119 plant-derived pharmaceutical medicines, regarding seventy four are utilized in trendy drugs in ways in which related to directly with their ancient uses as plant medicines by native cultures. Major pharmaceutical corporations ar presently conducting in depth analysis on plant materials gathered from the rain forests and alternative places for his or her potential medicative worth[5,11,12].

Excess Lipids Introduction

Excess lipids a broad term, conjointly referred to as excess proteins, may be a upset, specifically characterised by alterations occurring in bodily fluid supermolecule and compound protein profile thanks to magnified concentrations of Total cholesterin (TC), Low Density compound protein cholesterin (LDL-C),Very rarity compound protein cholesterin (VLDL-C) and Triglycerides (TG) with a concaminant decrease within the concentrations of High Density compound protein cholesterin (HDL-C) within the blood circulation. It's a typical disorder in developed countries and is that the major explanation for coronary cardiopathy. It results from abnormalities in supermolecule metabolism or plasma supermolecule transport or a disorder within the synthesis and degradation of plasma lipoproteins. The term "dyslipidaemia" currently a days is progressively getting used to explain abnormal changes in super molecule profile, replacement the previous term high lipids. Excess lipids means that abnormally high levels of fats within the blood. These fats embody cholesterin and triglycerides. These are vital for our bodies to perform however once they are high, they'll cause cardiopathy and stroke. Excess lipids is manifested as hypercholesteremia and/or hypertriglycerolemia. However, hypercholesteremia is that the commonest excess lipids. The lipids that are concerned in hypercholesteremia are cholesterin, a vital part of semipermeable membrane and a precursor of steroid synthesis and triglycerides, a crucial energy supply. They're transported in blood as lipoproteins. The

consequence of high lipids is that with time it will cause coronary-artery disease, and so the chance of coronary cardiopathy and stroke is magnified. However, in step with the newer scientific read, the cholesterin level alone isn't the full story. The chance of cardiopathy in future conjointly depends on several different factors that influence the health of a person's blood vessels and circulation[6,7].

Classes of Lipoprotein

Since blood and different body fluids square measure watery, thus fats want a special transport system to travel round the body. They're carried from one place to a different intermixture with macromolecule particles, referred to as lipoproteins. There are four forms of lipoproteins, every having terribly distinct job. These lipoproteins square measure delineated as follows.

Chylomicrons

Chylomicrons square measure created by the intestines for carrying new fatto the body's cells. These carry largely triglycerides. Chylomicrons carry exogenous lipids to liver, adipose, internal organ and striated muscle tissue wherever their lipid parts square measure free by the activity of the catalyst referred to as conjugated protein enzyme. Consequently, molecule remnants square measure left behind that square measure concerned by the liver. The density of these particles is less than 0.95 g/ml for chylomicrons and 1.006 g/ml for chylomicron remnants.

Very-Low-Density Lipoproteins (VLDL)

Very Low Density Lipoproteins are made by the liver and intestine, to carry fats around the body. These carry mostly triglycerides.

Low-Density Lipoproteins (LDL)

Low Density Lipoproteins square measure created by the liver to move steroid alcohol to the body's cells and tissues. LDL might type deposits on the walls of arteries and different blood vessels. So they're thought-about because the lazy or dangerous steroid alcohol[8-10].

MATERIAL AND METHODS

Materials

Plant material

The plant material used for the study is

The ethanolic extract of *Polygonum glabrum* plant.

Collection of plant material

The aerial part of *Polygonum glabrum* was collected from Tirumala hills, Tirupati, Andhra Pradesh, India.

Methods

Preparation of the plant extract

Preparation of *Polygonum glabrum* extract

The collected whole plant was dried at temperature, powdered by a mechanical grinder, sieved through 40mesh.

About 100g of small-grained materials were extracted with plant product (90%) exploitation soxhlet equipment.

The extraction was distributed till the extractive becomes colourless.

The extracts is then targeted and dried underneath reduced pressure.

The solvent free solid mass so obtained is dissolved in tween eighty and used for the experiment.

Preliminary phytochemical analysis

The crude and ordered extracts were tested for the subsequent phytoconstituents, sugar, alkaloids, glycosides, tannins, flavonoids, phytosterols, fats and oils by customary procedures as represented by khandelwal and kokate.

The extracts were subjected to the subsequent chemical check for the identification of varied active constituents.

Test for alkaloids

Dragondroff's test

To 1ml of the extract, add 1ml of Dragondroff's reagent, an orange red precipitate indicates the presence of alkaloids.

Mayer's test

To 1ml of the extract, add 2ml of Mayer's reagent, a cream coloured precipitate reveal the presence of alkaloids.

Wagner's test

To 1ml of the extract, add 2ml of Wagner's reagent, the formation of reddish brown precipitate indicates the presence of alkaloids.

Hager's test

To 1ml of the extract, add 3ml of Hager's reagent the formation of yellow precipitate confirms the presence of alkaloids.

Test for carbohydrates

Molisch test

To 2ml of the extract, add 1ml of α-naphthol resolution so add targeted concentrated sulphuric acid through the edges of the tube, purple or cherry violet ring at the junction of the 2 reveals the presence of carbohydrates.

Fehling's test

To 1ml of the extract, add an equal quantity of Fehling's solution A and B and warmth. The formation of the burnt sienna precipitate indicates the presence of carbohydrates.

Benedict's test

To 5ml of Benedict's chemical agent add 1ml of extract resolution and boil for 2 minutes and funky. Formation of a red precipitate shows the presence of carbohydrates.

Barfoed's test

To 5ml of Barfoed's chemical agent, add 1ml of the extract resolution and warmth to boil, a red precipitate of copper oxide was fashioned and confirms the presence of carbohydrates within the check extract.

Test for steroids and sterols

Libermann Burchard test

Dissolve the extract in 2ml of chloroform in an exceedingly dry tube. Add 10 drops of acetic anhydride and 2 drops of targeted concentrated sulphuric acid. The answer becomes red, then blue and eventually blue inexperienced, indicating the presence of steroids.

Salkowaski test

Dissolve the extract in chloroform and add volume of concentrate sulphuric acid. Formation of blue red to redness colourise chloroform layer and whereas the acid layer assumes marked inexperienced efflorescence, represents the steroid and sterol parts within the tested extract.

Test for glycosides

Legal test

Dissolve the extract in alkali and add freshly ready sodium metal nitroprusside solution to form it base-forming. The formation of pink to red color shows the presence of organic compound.

Baljet test

To 1ml of the check extract add 1ml metal sodium picrate resolution and also the yellow to orange color reveals the presence of glycoside.

Borntrager's test

Add a couple of cubic centimetre of diluted sulphuric acid to 1ml of the extract solution. Boil, filter and also the filtrate extract with chloroform. Separate the chloroform layer and treat with 1ml ammonia. The formation of red color shows the presence of anthraquinone organic compound.

Keller Mililani test

Dissolve the extract in ethanoic acid containing traces of ferrous chloride and transfer to a tube containing sulphuric acid. At the junction, formation of a reddish brown color, that step by step becomes blue, confirms the presence of deoxy sugar hooked up to the a glycon a part of organic compound.

Test for saponins

Foam test

About 1ml of alcoholic extract, dilute on an individual basis with 20ml of H_2O and shake in an exceedingly graduate for quarter-hour. A 1cm layer of froth indicates the presence of saponins.

To 1ml of the extract, add alcoholic vanillin chemical compound solution and a couple of drops of concentrated sulphuric acid. A deep violet color confirms the presence of saponins.

Test for flavonoids

Shinoda test

To 1ml of the extract, add metal turnings and 1-2 drops of targeted acid. Formation of pink or red color shows the presence of flavonoids.

To 1ml of extract, add 1ml of ferrous chloride, the formation of brown color confirms the presence of flavonoids.

Test for triterpenoids

Dissolve two or three granules of tin metal in 2ml of thionyl chloride solution. Then add 1ml of the extract into test tube. The formation of a pink colour indicates the presence of triterpenoids.

Detection of phenolics and tannins

Ferric chloride test

The extract was treated with few drops of neutral ferric chloride solution. The formation of blue black color indicates the presence of phenolic resin nucleus.

Gelatin test

To the extract, 1% gelatin solution containing common salt sodium chloride was more. The formation of white precipitate indicates the presence of tannins.

Lead acetate test

The extract was treated with few drops of lead acetate solution. Formation of yellow precipitate indicates the presence of flavonoids.

Test for protein and amino acid

Biuret test

To 1ml of the extract ad 1ml of 40% sodium hydroxide solution and 2 drops of 1% copper sulphate solution. Formation of violet color indicates the presence of protein macromolecule.

Ninhydrine test

Add 2 drops of freshly ready 0.2% Ninhydrine chemical agent to the extract resolution and warmth. Development of a purple color reveals the presence of proteins and amino acids.

Xanthoprotein test

To 1ml of the extract add 1ml of concentrated nitric acid. The formation of white precipitate confirms the presence of amino acid.

Test for fixed oils

Spot test

Press atiny quantity of extract between 2 filter paper. Oil stains on paper indicates the presence of fatty oil.

Acute toxicity studies

Animal selection

A complete of thirty male Wistar rats were obtained from the animal facility and administration and used for the study. All rats were certified with healthiness at the time of receiving. Age of the animals at the beginning of the treatment was about 8 to 12 weeks.

Acclimatization

Owen Wister rats were allowed to adapt to experimental area conditions for a amount of ten days before randomisation and treatment. Throughout the acclimation amount the rats square measure ascertained for the clinical signs.

Environmental conditions

The rats were maintained within the separate polypropene cages. within the experimental area, temperature of 23±2°C, controlled wetness (50-55%), twelve hrs of artificial lightening and twelve hrs of darkness cycle were maintained. The experimental area was clean and mopped with a disinfectant daily.

Housing conditions

The rats were housed supported the cluster size per polycarbonate cage. Every cage was fastened with a polypropene bottle with chrome steel nozzle. Feed was provided impromptu throughout the study. The litter was modified daily.

Feeding conditions

Rats were given 150gms of feed and sterilized water. Rat feed and provided water was modified on different days. The amounts of the feed consumed by the rats were calculated on the ordered days.

Atherogenic diet composition[13]

Dosing of animals

The animals were treated with the check and also the customary medicine orally supported the body weights of the animals. The animals were treated with the extracts for concerning fourteen days. Throughout dosing of animals, the body weights of the animals and also the food consumed by the animals were taken on ordered days.

Grouping of animals: Ant atherosclerosis

The animals were divided into four groups. Each group contains five animals.

Grouping is as follows:

Group 1: Normal Group (Tween 80)

Group 2: Control Group (HFD)

Group 2: Extract I- *Polygonum glabrum* + HFD (200 mg/kg)

Group 3: Extract II- *Polygonum glabrum* + HFD (400 mg/kg)

Group 4: Standard-Atorvastatin + HFD (10 mg/kg)

Dosing of animals

The animals were dosed with the test and the standard drugs orally based on the body weights of the animals. The animals were dosed with the

extracts for about 14 days. During dosing of animals, the body weights of the animals and the food consumed by the animals were taken on successive days.

The rats were treated with test and standard drugs by oral gavage for 14 days.

After this time i.e., 20 hrs after the last application of the test compounds the animals are anaesthetized with anaesthetic ether and 1.2ml of blood is withdrawn by retro orbital puncture.

The blood samples will be collected on the 14th day for estimating biochemical parameters

The blood samples were taken from the rats after overnight fasting.

Biochemical parameters were determined after treatment.

The serum was labeled with the animal number and the estimations were made. The serum enzymes SGOT, SGPT and ALP cholesterol HDL, LDL, VLDL and triglyceride level and the lipid profile (total level) and total protein was determined enzymatically on prietest bio chemistry analyser. SOD, GSH, MDA were determined by using UV Spectrophotometer

RESULTS

Plant Extract

The contemporary aerial components of *Polygonum glabrum* were dried below shade and powdered in grinder. 300gm of powder was extracted with methanol (MEDB) by soxhletion in soxhlet apparatus. The yield of extract was 30.27 gm and the percentage yield was 15.13%. The extract was hold on in air tight instrumentality and additional used for pharmacological screening.

Preliminary Phytochemical Screening

Phytochemical screening of *Polygonum glabrum* was done treatment with Methanol, the extract showed the presence of Alkaloids, carbohydrates, glycosides, saponins, flavonoids and tannins. Results are shown in the following table.

Acute toxicity studies

The acute toxicity studies of *Polygonum glabrum* was applied as per OECD pointers 423. There was no gross evidence of any abnormalities ascertained up to a amount of 4-6hrs and no mortality was ascertained at the utmost tolerated dose (MTD) level of 2000mg/kg bw. Per oral. The utmost tested dose was 2000mg/kg body weight. Further pharmacological screenings were applied with two dose ranges i.e. 1/10 of MTD (200 mg/kg bw p.o.), 1/5 of MTD (400mg/kg bw p.o.). They were taken as Test doses T_1 and T_2 respectively.

Effect of ethanolic extract of *polygonum glabrum* on HFD, atherosclerosis profile in rats

Biological parameters

Body Weights

HFD and MSG fed rats significantly gained weight compared to the normal rats. Oral administration of EEPG had reduced the weight gained. EEPG at a dose of 200 mg/kg b.w. p.o. significantly decreased ($p<0.05$) weights, while EEPG at 400 mg/kg b.w. p.o. and orlistat had decreased the body weight significantly ($p<0.01$) at the end of week 4. Results are presented in the following Table No.4, depicted in Graph No.1.

Liver Weights

The livers in negative control group were enlarged and produced a yellow color, indicating liver steatosis and increased weights. The livers were appeared yellow and bulky due to presence of fat. On the other hand group given with orlistat reversed the conditions of liver to remain normal and healthy. Significant reduction ($p<0.05$) was seen with EEPG at 200mg/kg whereas significant reduction ($p<0.01$) was observed with EEPG at 400mg/kg and orlistat. Presented in Table No.5 and depicted in Graph No.2.

Biochemical parameters

Serum Lipid Profile

Rats fed with high fat diet (HFD) showed impairment in normal lipid profile, leading to increased total cholesterol, triglyceride, LDL-C, VLDL-C while HDL-C was decreased. EEPG at 200mg/kg bw showed significant reduction ($p<0.05$), while, EEPG at 400mg/kg bw significantly decreased ($p<0.01$) the total cholesterol levels were highly significant reduction of $p<0.001$ was observed with orlistat at 50mg/kg bw.

Significant reduction of triglycerides, $p<0.05$ was seen with EEPG 200 mg/kg bw and the values were

found to be <0.01 with EEPG 400 mg/kg bw whereas highly significant reduction p<0.001 was seen with orlistat at 50mg/kg bw.

LDL and VLDL were significantly reduced p<0.05 with EEPG at 200mg/kg bw but with EEPG 400 mg/kg bw and orlistat at 50mg/kg bw the value of LDL was found to be p<0.01. Whereas HDL-C levels were significantly increased with EEPG 400 mg/kg bw and orlistat at 50mg/kg bw p<0.01 when compared to normal and untreated groups.

Liver Function Test

Animals treated with high fat diet (HFD) showed increased levels of marker enzymes SGOT, SGPT and ALP but upon administration of EEPG significantly reduced the levels. P<0.05 was seen with EEPG 200 mg/kg bw and p<0.01 was seen with EEPG 400 mg/kg bw and orlistat at 50mg/kg bw p<0.01. P<0.01 value was seen in the levels of ALP with administration of EEPG at 200mg/kg bw and EEPG 400 mg/kg bw but p<0.001 was seen with orlistat at 50mg/kg bw.

Hepatic morphology and histopathology

The livers of untreated groups were found to be yellow in color and appeared bulky, whereas livers of the treated groups were found to be normal and less bulky. Histological analysis is shown in the figure, where liver of untreated rats exhibited a typical signs of fatty mass i.e. Showing accumulation of fat droplets through the liver acini. When treated with EEPG and Orlistat smaller degree of lipid accumulation and fewer pathological signs were observed in a dose dependent manner.

Cardiac Risk Indicator

Atherogenic index of plasma and % protection of HFD treated animals were calculated from their Triglycerides and HDL-C values. There was a dose dependent reduction in atherogenic index of plasma with EEPG administered at all two doses (200 and 400 mg/kg bw p.o.) and Orlistat exhibited maximum reduction in AIP which is as follows:

In HFD Model

T_1 - 2.93

T_2 - 2.64

Orlistat - 2.35.

Table No.1: Atherogenic diet composition

S.No	Composition	Normal diet (%)	Atherogenic diet (%)
1	Protein (Milk powder)	12	10
2	Carbohydrates (Wheat flour)	71	61
3	Sugar	05	05
4	Fat (Butter)	05	16
5	Salts	04	04
6	Vitamins	01	02
7	Fibers	02	01
8	Cholesterol	--	01
9	Total Weight	100g	100 g

Table No.2: Preliminary Phytochemical Analysis

S.No	TEST	RESULT
1	**ALKALOIDAL TEST** a. Dragondroffs test b. Mayer's test c. Wagner's test d. Hager's test	 Positive Positive Positive Positive
2	**CARBOHYDRATES TEST** a. Molish's test b. Fehling's test c. Benedict's test d. Baeford's test	 Positive Positive Positive Positive
3	**STEROIDS TEST** a. Libermann Buchard test b. Salwoski test	 Negative Negative
4	**GLYCOSIDES TEST** a. Legal test b. Baljet test c. Killerkilaini test d. Borntagers test	 Positive Positive Positive Positive
5	**SAPONINS TEST** a. Foam test	 Positive
6	**FLAVONOIDS TEST** a. Shinoda test	 Positive
7	**TRITERPINOIDAL TEST**	Negative
8	**TANNINS TEST** a. Ferric chloride test b. Gelatin test c. Lead acetate test	 Positive Positive Positive
9	**PROTIEN and AMINOACIDS TEST** a. Buret's test b. Ninhydrin test c. Xanthoprotic test	 Negative Negative Negative

Table No.3: Acute toxicity study results

Alertness	**Behavioral Responds**	↓
Stereotypy		-
Irritability		↓
Fearfulness		↑
Touch responds		↑
Analgesia		N
Spontaneous activity		↓
Grooming		↑
Restfulness		↑
Inclined plane test		↑
Body Temperature		↑
Righting responds	**Neurological Responds**	-
Limb tone		N
Grip strength		+
twitching		-
Abdominal tone		+
Pinnal reflex		N
Corneal reflex		N
Straub tail		+
Tremors		-
Convulsions		-
Catalepsy		-
Writhing	**Autonomic Responds**	+
Defecation		↑
Urination		↑
Piloerection		+
SMA		N
Respiration		↑
Pupil size		N
Cyanosis		N
Heart rate		N
Ataxia		+
Ptosis		-
Salivation		-
Lachrymation		-

(↑) Increased (↓) Decreased (+) Presence (-) Absence (N) None

Table No.4: Effect of EEPG on body weights of rats (HFD MODEL)

S.No		Differences in body weights (gm) (Mean ± SEM)			
	Group (n=5)	**Week 1**	**Week 2**	**Week 3**	**Week 4**
1	Group I Normal control group	33.2 ± 1.92	36.8 ± 0.9	38.2 ± 1.9	41.20 ± 1.0
2	Group II Negative control group HFD	33.4 ± 1.89	77.6 ± 3.5	102.3 ± 4.0	112.6 ± 3.9
3	Group III Positive control group Orlistat 50mg/kg b.w. p.o	33.4 ± 1.86	68.4 ± 3.8	92.6 ± 4.5	84.4 ± 4.6
4	Group IV T_1 – EEPG 200mg/kg b.w. p.o	33.2 ± 4.5	79.6 ± 3.1	99.1 ± 4.3	95.3 ± 4.1
5	Group V T_2 – EEPG 400mg/kg b.w. p.o	33.8 ± 1.6	77.4 ± 5.4	97.4 ± 2.8	89.54 ± 4.8

Table No.5: Effects of EEPG on liver weights of rats (HFD MODEL)

S.No	Groups (n = 5)	Liver weights (g) (Mean ± SEM)
1	Group I Normal control	5.92 ± .23
2	Group II Negative control HFD	6.79 ± 0.15
3	Group III-Positive control Orlistat 50mg/kg b.w. p.o.	6.15 ± 0.23**
4	Group IV T_1 – MEDB-200mg/kg b.w. p.o	6.57 ± 0.17*
5	Group V T_2 – MEDB-400mg/kg b.w. p.o.	6.29 ± 0.16**

Table No.6: Effect of EEPG on Total Cholesterol and Triglyceride levels in HFD rats

S.No	Groups (n = 5)	Total Cholesterol (mg/dl) Mean ± SEM	Triglycerides (mg/dl) Mean ± SEM
1	Group I Normal control	82.13 ± 2.98	71.05 ± 1.98
2	Group II Negative control (HFD)	138.43 ± 2.13	141.87 ± 3.12
3	Group III Positive control Orlistat 50mg/kg b.w. p.o	96.98 ± 2.04***	78.91 ± 3.89***
4	Group IV T_1 – EEPG 200mg/kg b.w. p.o	125.43 ± 3.65*	109.98 ± 3.16*
5	Group V T_2 – EEPG 400mg/kg b.w. p.o.	118.5 ± 2.91**	89.63 ± 3.87**

Table No.7: Effect of EEPG on HDL, LDL AND VLDL levels in rats

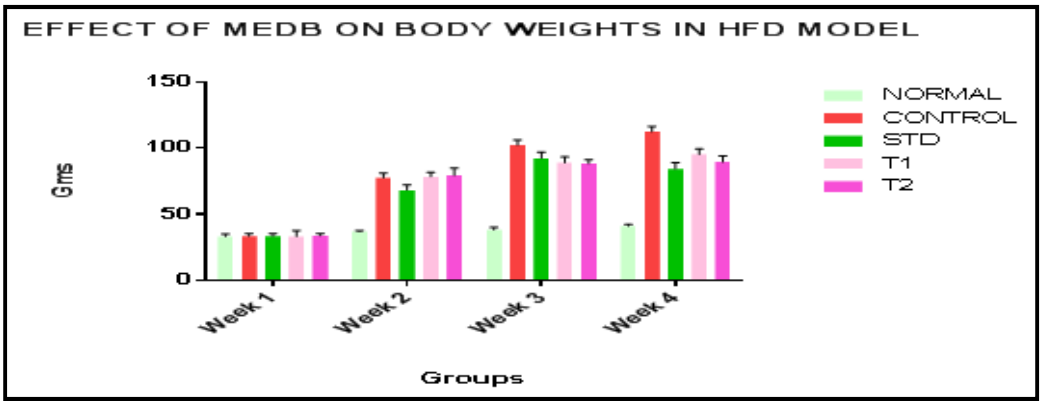
S.No	Groups (n = 5)	HDL (mg/dl) Mean ± SEM	LDL (mg/dl) Mean ± SEM	VLDL (mg/dl) Mean ± SEM
1	Group I Normal control	32.62 ± 2.12	34.54 ± 2.01	15.39 ± 1.07
2	Group II Negative control HFD	23.87 ± 3.39	88.09 ± 3.12	27.59 ± 3.39
3	Group III Positive control Orlistat 50mg/kg b.w. p.o	30.45 ± 3.97**	49.67 ± 3.96**	17.29 ± 1.87**
4	Group IV T_1 – EEPG 200mg/kg b.w. p.o	27.42 ± 1.89*	74.98 ± 2.12*	23.24 ± 1.18*
5	Group V T_2 – EEPG 400mg/kg b.w. p.o	28.91 ± 2.98**	71.02 ± 4.14**	19.36 ± 2.25**

Table No.8: Effect of EEPG on SGOT, SGPT AND ALP levels in rats

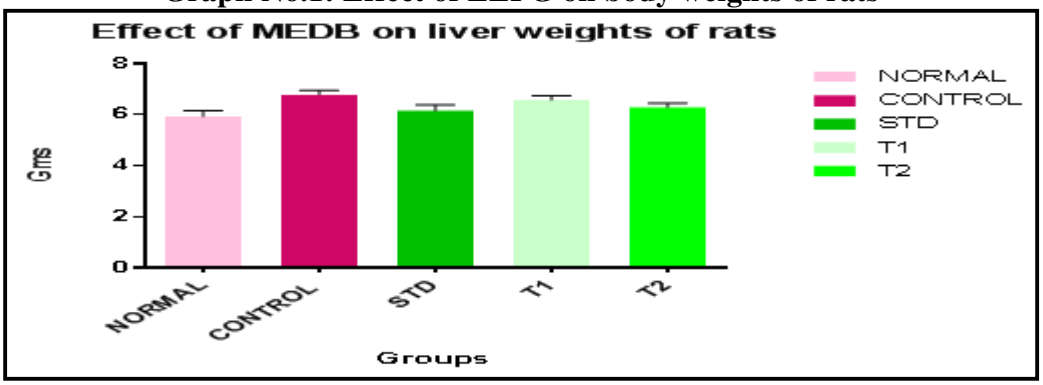
S.No	Groups (n = 5)	SGOT (IU/L) Mean ± SEM	SGPT (IU/L) Mean ± SEM	ALP (IU/L) Mean ± SEM
1	Group I Normal control	17.34 ± 3.67	22.42 ± 3.65	87.49 ± 4.93
2	Group II Negative control HFD	42.28 ± 2.87	52.85 ± 5.98	246.59 ± 2.98
3	Group III Positive control Orlistat 50mg/kg b.w. p.o	21.84 ± 2.91**	23.78 ± 5.92**	97.31 ± 5.24***
4	Group IV T_1 – EEPG 200mg/kg b.w. p.o	31.59 ± 3.66*	37.39 ± 4.03*	159.93 ± 3.61**
5	Group V T_2 – EEPG 400mg/kg b.w. p.o	24.56 ± 3.75**	32.78 ± 5.02**	123.09 ± 4.63**

Table No.9: Atherogenic index and percentage protection with EEPG: (HFD MODEL)

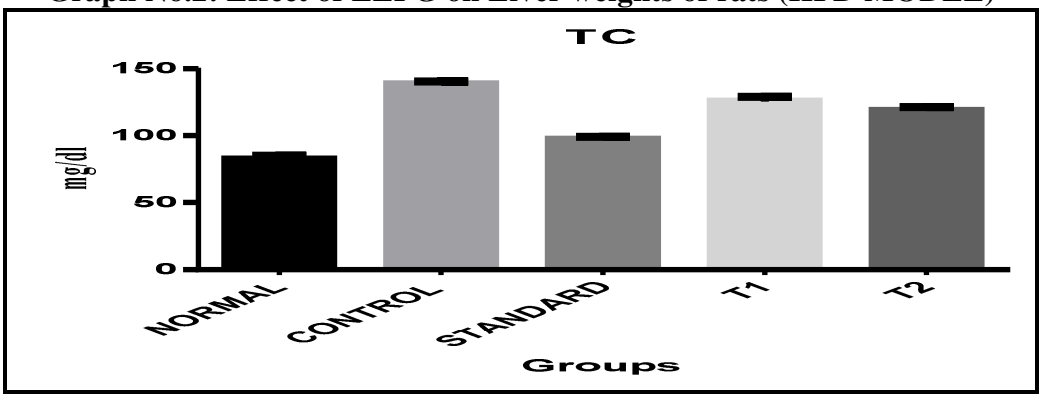
S.No	Group (n=5)	Atherogenic Index of Plasma (AIP)	Percentage protection
1	Group I Normal control	2.09	
2	Group II Negative control HFD	5.63	
3	Group III Positive control Orlistat 50mg/kg b.w. p.o	2.35	59.7 %
4	Group IV T_1 – MEDB 200mg/kg b.w. p.o	2.93	49.2 %
5	Group V T_2 – EEPG 400mg/kg b.w. p.o	2.64	55.32 %



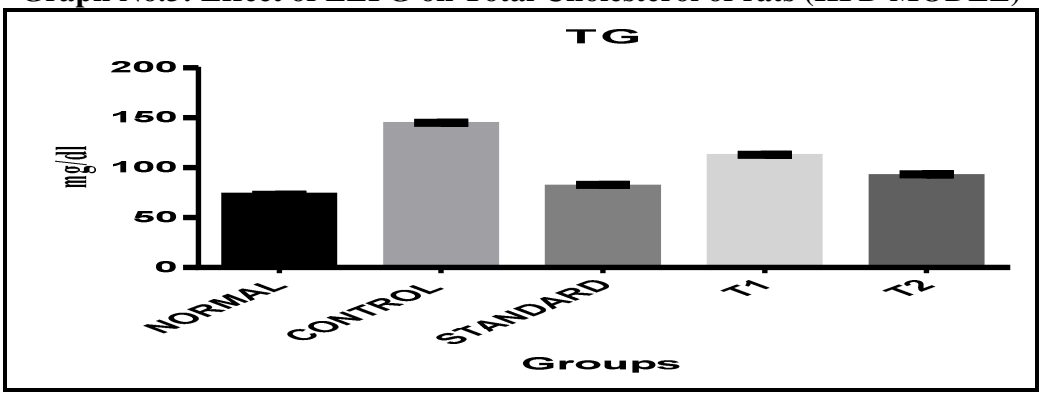
Graph No.1: Effect of EEPG on body weights of rats



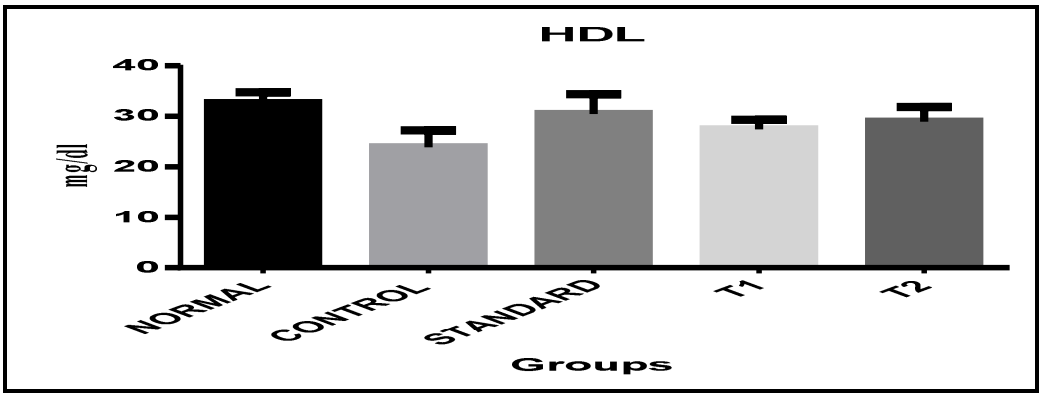
Graph No.2: Effect of EEPG on Liver weights of rats (HFD MODEL)



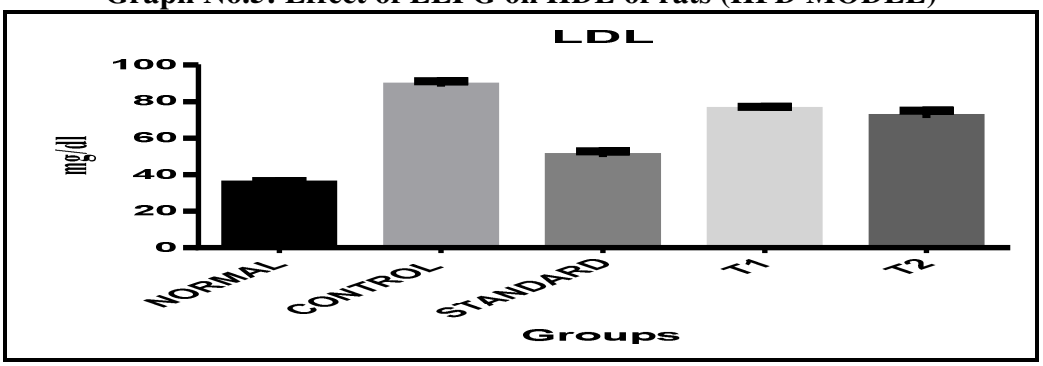
Graph No.3: Effect of EEPG on Total Cholesterol of rats (HFD MODEL)



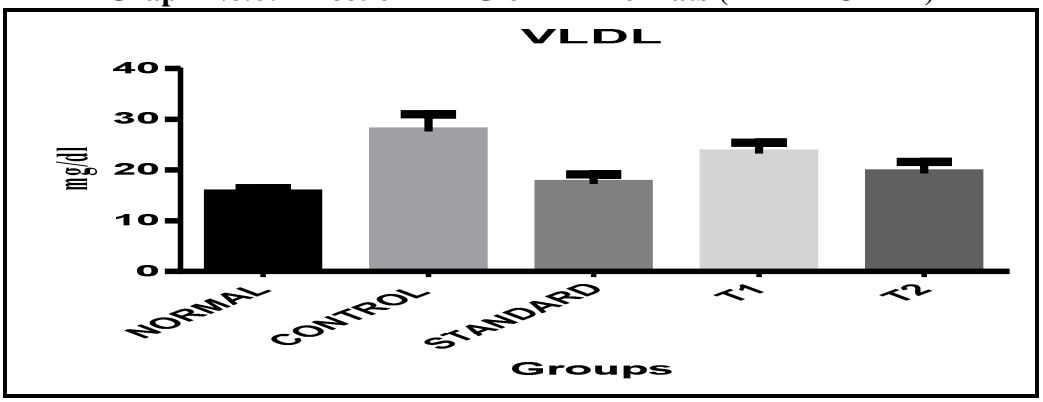
Graph No.4: Effect of EEPG on Triglycerides of rats (HFD MODEL)



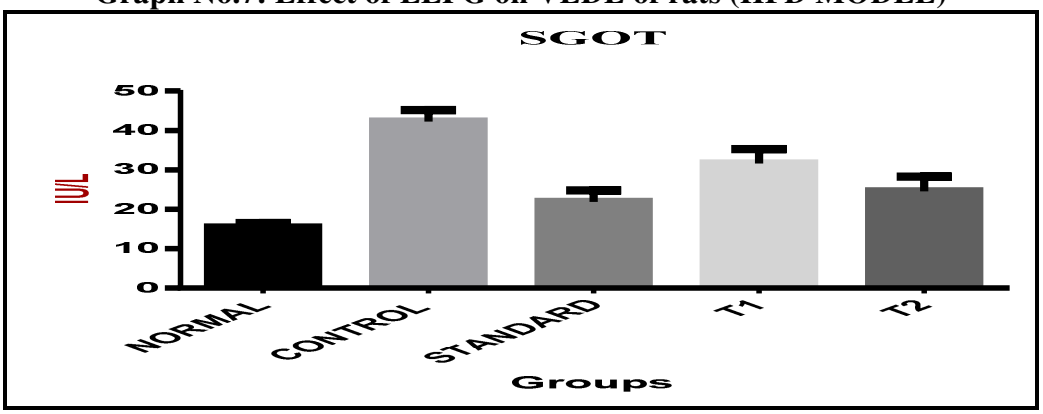
Graph No.5: Effect of EEPG on HDL of rats (HFD MODEL)



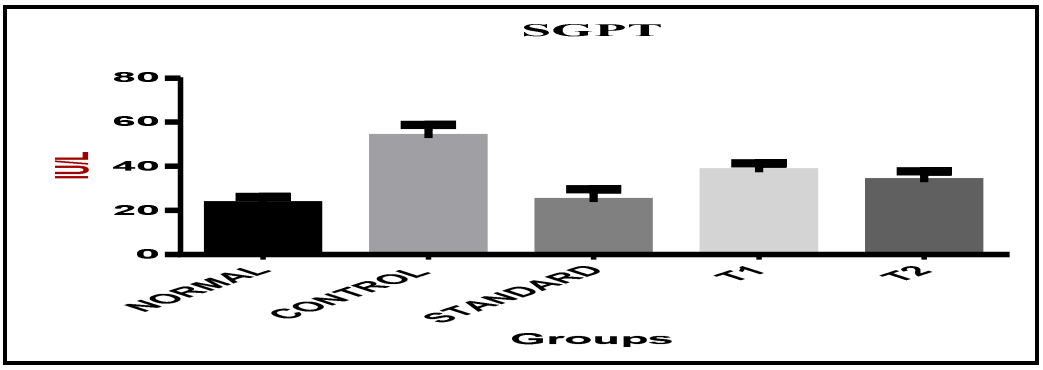
Graph No.6: Effect of EEPG on LDL of rats (HFD MODEL)



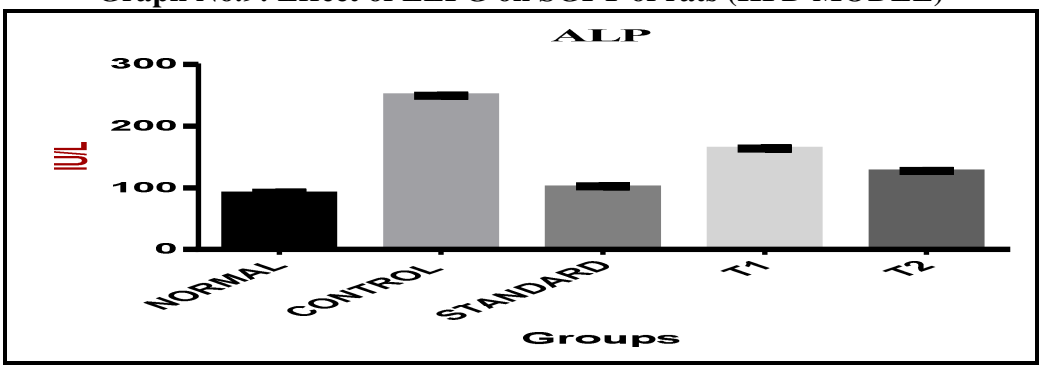
Graph No.7: Effect of EEPG on VLDL of rats (HFD MODEL)



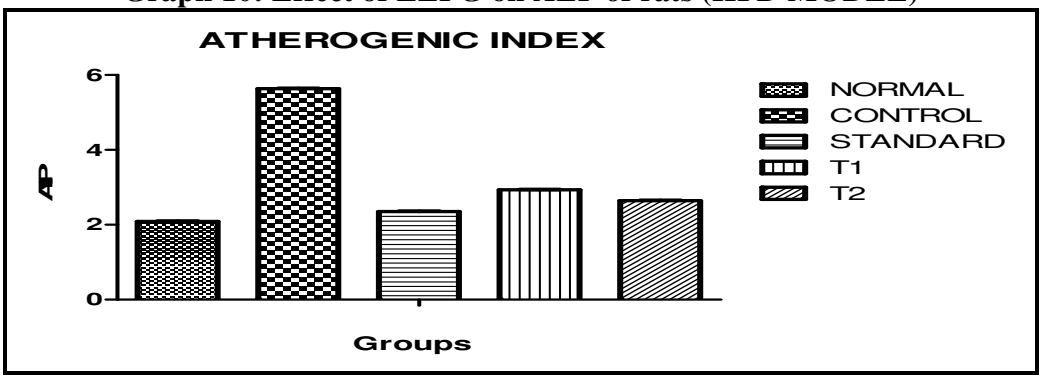
Graph No.8: Effect of EEPG on SGOT of rats (HFD MODEL)



Graph No.9: Effect of EEPG on SGPT of rats (HFD MODEL)



Graph 10: Effect of EEPG on ALP of rats (HFD MODEL)



Graph No.11: Effect of EEPG on Atherogenic Index of rats (HFD MODEL)

CONCLUSION

Phytochemical screening of the extract shows the presence of chemical constituents like Alkaloids, steroids, fixed oils, cardio tonic aglycones, flavonoids, saponins, carbohydrates, proteins, resins. Acute toxicity tests were performed in step with the OECD guide line No.423, LD50 worth was found to be 200mg/kg and 400mg/kg. Anti atherosclerotic activity was performed by victimisation the high fat diet iatrogenic methodology. In the present study an increase in plasma HDL-cholesterol with a concomitant percentage decrease from other lipid was observed. It can be concluded from the present data that the levels of total serum cholesterol, triglyceride and MDA which are actually raised in atherogenic diet, can be lowered significantly with Polygonum glabrum. And total proteins and antioxidant parameters SOD, GSH which are actually lowered in atherogenic diet can be raised significantly with Polygonum glabrum. From this we are able to conclude that the extract (Polygonum glabrum.) Showed the anti-atherosclerotic activity.

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CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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