Evaluation of hypolipidemic and hepatoprotective effects of *Linum Usitatissimum* L. Seeds on hypercholesterolemic rats

Running Title: Hypolipidemic and hepatoprotective effects of *Linum Usitatissimum* L. Seeds

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ABSTRACT

Background: Hyperlipidaemia is a chronic progressive disease, which encompasses various genetic and acquired conditions resulting in inappropriately elevated lipid levels in humans. Several preclinical and clinical studies have demonstrated that dietary supplementation with *Linum Usitatissimum*have beneficial cardiovascular effects like antihypertensive action, antiatherogenic effects, lowering of cholesterol, anti-inflammatory action and inhibition of arrhythmia. There are few studies on hepatoprotective effects of flax seeds in hyperlipidemic animal models. Aim and objectives:Hence, this study was undertaken to evaluate the hypolipidemic and hepatoprotective effect of Linum seeds.**Methods:**The study was conducted for the period of two months in Department of Pharmacology KIMS, Bhubaneswar. Total 36 Albino wistar rats were taken and divided into 6 groups containing 6 rats each. Group II was hypercholesterolemic (HC) control, Group III was treatment control and Group IV to VI rats were given different doses of extract. Blood samples were taken at the end of two months.**Results:**All the values are expressed as Mean±SD (standard deviation). Significant decrease in lipid parameters and liver enzymes is seen at the end of two months in all 3 groups receiving the extract,where the highest dose of extract used, is near to the standard drug.**Conclusion:**From this study, it can be concluded that the addition of the flaxseeds to the diet may alleviate the rise in circulating cholesterol levels induced by the high cholesterol diet through its content of alfa-linolenic acid (ALA) and lignins. The hepatoprotective role of flaxseed in hypercholesterolaemia has also been demonstrated in this this study.

Keywords:*Linum Usitatissimum* L. Seeds, Hyperlipidemia, Hepatoprotective effect, high fat diet

1. INTRODUCTION

Hyperlipidaemia is a chronic progressive disease, which encompasses various genetic and acquired conditions resulting in inappropriately elevated lipid levels in humans. This condition is common throughout the world and the numbers continue to rise at an alarming rate. Lipids traditionally include cholesterol, lipoproteins, chylomicrons, very low-density lipoprotein (VLDL), low-density lipoprotein (LDL), apolipoproteins, and high-density lipoprotein (HDL)[1].

Hyperlipidaemia, particularly elevated LDL cholesterol, is one of the most prevalent risk factors contributing to the development of atherosclerosis, leading to subsequent vascular disease.In contrast, HDLcholesterol helps to regulate cholesterol levels by preventing imbalances that would increase the risk of atherosclerotic vascular disease. Recent studies have reported that high cholesterol is present in 25-30% of urban and 15-20% ruralpopulation in India[2]. The most common dyslipidaemia in India are borderline high LDL cholesterol, low HDL cholesterol and high triglycerides[2].Complications from undertreated or untreated hyperlipidaemia includecoronary artery disease, peripheral artery disease, cerebrovascular accidents, aneurysms, type II diabetes, high blood pressureand even death[3].Hyperlipidaemia is also a known risk factor for fatty infiltration of the liver, which can progress to cirrhosis and liver failure. The hepatic involvement is detected early through elevated liver enzyme tests[4].The initial treatment modalities of hyperlipidaemia are diet, lifestyle modification and one of the lipid-lowering agents like statins, fibrates, nicotinic acid may be added if required. Statin therapy is very beneficial for a majority of the patients but there are associated complications like myopathy, renal injury, arthralgia, extremity pains, nausea, myalgia, elevated liver enzymes/hepatotoxicity, diarrhoea, and rhabdomyolysis. Upto 5 to 20% of patients taking a statin have reported a muscle-related intolerance[5].

It is well known that essential fatty acids like omega-3 and omega-6 are important for control of cholesterol homeostasis. Long-chain omega-6 fatty acids include linoleic, gamma-linolenic, and arachidonic acids. Omega-3 fatty acids comprise of long-chain alpha-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA). Omega-3 fatty acids have anti-inflammatory, antiarrhythmicand anti-thrombotic properties whereas omega-6 fatty acids, though proposed to be lipid lowering agents, are proinflammatory and prothrombotic[6].The typical Western diet includes corn, cotton seed, sunflower and safflower oils which is rich in omega-6 fatty acids due to the abundance of linoleic acid.Conversely, omega-3 fatty acids account for only a small percentage of the daily dietary fat intakewalnuts, flaxseed, and canola[7]. This has drastically shifted the dietary ratio of omega-6 to omega-3 fatty acids from an estimated 1:1 in the early human diet to approximately 10:1 in the typical modern American diet. Indian diets are principally vegetarian and relatively low in fat. Also, the main sources of fat are of plant origin rather than animal originresulting in a diet that is relatively low in saturated FA, high in omega -6 polyunsaturated fatty acids (PUFA), and very low in omega-3 PUFA.The dietary recommendations by the Indian Council of Medical Research (ICMR) have always emphasized on increasing omega-3 PUFA intake in Indian diets; but, it is not quite clear how this can be achieved[8].

Linum Usitatissimum L. Seedsis one of the richest plant sources of ALA (alpha linolenic acid), an omega-3 polyunsaturated fatty acid (PUFA). Several preclinical and clinical studies have demonstratedthat dietary supplementation with flaxseed have beneficial cardiovascular effects like antihypertensive action, antiatherogeniceffects, lowering of cholesterol, anti-inflammatory action and inhibitionof arrhythmias[9]. However, studies ofhepatoprotective effect of *Linum Usitatissimum* L. Seedsextract on hypercholesterolaemic models are sparse.

Aim of the Study: Hence, this study was undertaken to evaluate the effect of flaxseeds extract on hyperlipidaemia and liver function in albino rats fed on high fat diet and to compare the activity with standard drug Rosuvastatin in animal models.

2. MATERIALS AND METHODS

2.1Experimental Animals:

The study was conducted on healthy adult albino wistarrats in the Department of Pharmacology in collaboration with Department of Biochemistry at Kalinga Institute of Medical Sciences, Bhubaneswar. The experiments were done from January 2021 to March 2021. All experimental protocols were approved by Institutional Animal Ethics Committee vide No. 1730/PO/Re/13/CPCSEA/13/19. Healthy malealbino wistarrats weighing between 150-200g were used in this study for assessment of hypolipidemic and hepatoprotective effects. They were housed in standard polypropylene cages and kept under controlled room temperature (24 ± 2^0 C) and humidity (55 ± 5%) with a 12hr light/dark cycle. The animals were fed with standard laboratory diet and water ad libitum. The animals were acclimatized to the surroundings for one week before being grouped for the experiment. In addition, all the precautions were taken to minimize pain and discomfort to the animals.

2.2Preparation Of Flax Seeds Extracts:

Linum Usitatissimum L. Seedswere purchased from the local market and shade dried for 2-3 days. The seeds were then powdered by mechanical grinder and stored properly in air tight containers. The powdered form offlaxseeds was used for the preparation of aqueous extract. About 100 g of the powdered seed material was soaked in 2 litres of distilled water and shaken thrice daily. The extract obtained after this was concentrated under reduced pressure in rotary evaporator at 50-60 °C leaving a brown residue. The aqueous extracts of theseeds obtained was transferred to a petri dish and kept over water bath (50 °C) until the solvent got completely evaporated. It was stored in air tight glass containers in refrigerator at 2-8 °C for further use in experiments. Toxicity studies done previously in rats at 2000mg/kg dose showed no mortality of the animals[10]. Hence, 100mg/kg, 200mg/kg and 400mg/kg doses of *Linum Usitatissimum* Seed Extract (LUSE) were selected and prepared by reconstituting the extract in distilled water[10].

2.3Hyperlipidemic diet:

High fat dietwas prepared by mixing Indian vanaspati ghee and coconut oil in the ratio of 3:1 (v/v). It was administered to the experimental rats at a dose of 3 ml/kg body weight per dayby mixing it with the standard laboratory rat diet[11]. This comprised of the high fat diet (HFD) animal model based on the traditional fatty diet consumed by the Indian population.

2.4Study Design:

About 36 albino wistar rats of either sex and those having the body weights, biochemical parameters within normal limits were selected for the study. The selected animals were dividedrandomly into 6 equal groups (Group I – VI). The study was conducted for a period of

2 months during which the animals were administered the drugs along with continuous feeding.

Group I– served asVehicle control, received standard laboratory rat diet and distilled waterorallydailyGroups II - VI were fed with highfat diet (HFD) for 2 months along with the drugs, out of which
Group II–served as Hypercholesterolemic (HC) control, received distilled water(5ml/kg orally) dailyGroup III– served as Treatment control, receivedstandard drug Rosuvastatin(10 mg/kg, orally) daily Group IV, V, VI–served as Test drug groups, received graded doses of flax seed extract (LUSE) 100mg/kg, 200mg/kg and 400mg/kg orally daily,respectivelyfor a period of 2 months.

The body weights of the experimental animals were measured at the end of 2 months. Blood samples for biochemical analysis were taken from retro bulbar venous plexus of eachrat at the end of the experiment[12].

2.5Biochemical Analysis:

3 ml of retro-orbital blood sample was collected after overnight fasting. Serum was then separated after centrifuging it at 3000rpm for 20 minutes. Serum was then analyzed for the parameters of lipid profile and liver function tests. Total cholesterol (TC) was done by Cholesterol oxidase peroxidase, end point method[13]. Serum Triglyceride (TG) was done by Glycerol phosphate oxidase method based upon the methodology of Mc Gowen and Fossati et al[14,15].Calculation of VLDL, LDL and HDL was done by the formula developed by Friedwald et al[16].

Serum SGOT (Serum glutamate Oxaloacetate transaminase), SGPT(Serum glutamate pyruvate Transaminase) and ALP (Alkaline phosphatase) was done by IFCC, kinetic method[17]. All the biochemical analysis was done by semi-automated analyser, Photometer 5010 by DiasysIndia and Reagents used were from Erba Diagnostics (Mannheim).

2.6Statistical Analysis:

Data obtained were expressed as mean ± SD (standard deviation) values. The values were analysed by using one-way ANOVA followed by *post-hoc* tests using the software sociostatistics.com. The chosen level of significance was $p < 0.05$ and interpretation was done accordingly.

3. RESULTS

This experiment was undertaken to evaluate the hypolipidemic and hepatoprotective effectof flax seed extractin albino rats whose basal body weights andbiochemical parameters were within normal limits. The rats were divided into six groups (I-VI). Group I was the vehicle treated group, Groups II-VI were the hypercholesterolaemic (HC) rats which were administered the standard drug Rosuvastatin (Group III) and the Groups IV-VI received graded doses of *Linum Usitatissimum* Seed Extract (LUSE), group II did not receive any drug. There was an increase in bodyweights of the HC rats as well as the LUSE treated rats who were also on high fat diet.

- **Effect of LUSE administration(100 mg/kg, 200mg/kg and 400mg/kg)on serum lipid profile(mg/dl) in normaland hypercholesterolaemic rats:**

The values (mg/dl) of total cholesterol (TC), triglycerides (TG), very lowdensity lipoproteins (VLDL), low density lipoproteins (LDL) and high density lipoproteins (HDL) obtained from the serum of the experimental rats at the end of the experiment were expressed as Mean ± SD and tabulated as shown in Table 1.The vehicle treated rats (Group I) showed no significant changes in their TC, TG, VLDL, LDL and HDL levels at the end of the experiment. There was a significant increase in the levels of TC, TG, VLDL, LDL and a significant decrease in HDL of the HC rats (Group II) at the end of 2 months and this change is also significant ($p < 0.001$) when compared to the vehicle control rats.It is observed that in the treatment control Group III which received the standard lipid lowering drug - Rosuvastatin, the lipid profile parameters TC, TG, VLDL, LDL were significantly less than the HC Group II and HDL levels improved significantly as compared to Group II.In the LUSE treated rats i.e, Groups IV, V and VI, there was a significant decrease in the levels of TC, VLDL, LDL seen with increase in dose of the LUSE from 100 mg/kg to 400 mg/kg and a highly significant decrease at 400 mg/kg, demonstrating a graded dose response. The effect seen with 400 mg/kg dose of LUSE was almost near to the effect with Rosuvastatin treatment in rats. Whereas, significant decrease in TG levels was observed at the highest dose of 400mg/kg LUSEin Group VI only and not with the lower doses of LUSE in groups IV and V. The HDL levels of the test groups did not show any significant changes as compared to the HC group II.

Table 1: Effect of LUSE administration (100 mg/kg, 200mg/kg and 400mg/kg)on serum lipid profile(mg/dl) in normal and hypercholesterolaemic rats

Group	Treatment	TC (mg/dl)	TG (mg/dl)	VLDL (mg/dl)	LDL (mg/dl)	HDL (mg/dl)
I	Vehicle control	91.16 ± 2.11	66.59 ± 1.77	14.08 ± 0.43	42 ± 3.09	58.5 ± 2.87
II	Hypercholesterolemic control	147.5 ± 3.30[a]	101.84± 2.55[a]	21.09 ± 0.52[a]	68.83 ± 2.43[a]	25 ± 3.31[a]
III	Treatment control (Rosuvastatin 10mg/kg)	93.67 ± 1.58***	69.48 ± 1.55***	15.28 ± 1.43***	37.17 ± 2.60***	59.81 ± 2.40***
IV	LUSE-100mg/kg	128.17 ± 3.22*	99.69 ± 2.32	18.16 ± 2.13*	59.83 ± 2.68*	32.17 ± 3.41
V	LUSE-200mg/kg	111.83 ± 2.75**	92.67 ± 1.45	17.88 ± 0.23**	49.87 ± 2.26**	36 ± 3.33
VI	LUSE-400mg/kg	107.67 ± 4.52***	86.29 ± 2.54*	16.32 ± 0.82***	48.83 ± 4.80***	39.5 ± 1.72

Total Cholesterol (TC), Triglycerides (TG), Very low density lipoproteins (VLDL), Low density lipoproteins (LDL) and High density lipoproteins (HDL), *Linum Usitatissimum* Seed Extract (LUSE)

All the values are mean ± SD, n=6, data analyzed by ANOVA and post hoc test. [a]*P*<0.001 when compared to group I (vehicle control), **P*value <0.05– significant, ***P* value <0.01- highly significant, ****P* value <0.001- very highly significant when compared to Group II (Hypercholesterolemic control).

- **Effect of LUSE administration(100 mg/kg, 200mg/kg and 400mg/kg)on serum liver enzymes(mg/dl) in normal and high cholesterolfed male rats:**

The level of clinical biochemistry such as Serum glutamate Oxaloacetate transaminase(SGOT), Serum glutamate pyruvate Transaminase (SGPT), alkalinephosphatase (ALP)was evaluated to determine the enzymatic activities ofthe livers, ~~kidneys and heart muscles~~ of the control groups andthe experimental groups. The values obtained were expressed as Mean ± SD and tabulated as shown in Table 2. The levels of SGOT, SGPT and ALP of the vehicle treated rats did not show any significant changes from the normal baseline levels at the end of the experiment, however these levels showed a significant rise in the HC Group II. It is seen that in the treatment control Group III which received the standard lipid lowering drug - Rosuvastatin, the SGOT, SGPT and ALP levels were significantly less than the HC Group II. In the LUSE treated rats i.e, Groups IV, V and VI, there was a significant decrease in the levels of SGOT, SGPT and ALP seen with increase in dose of the LUSE from 100 mg/kg to 400 mg/kg and a highly significant decrease at 400 mg/kg, demonstrating a graded dose response.

Table 2: Effect of LUSE administration (mg/kg) on serum liver enzymes in normal and high cholesterol fed male rats (mg/dl)

Group	Treatment	SGOT (IU/L)	SGPT (IU/L)	ALP (IU/L)
I	Vehicle control	102.67 ± 6.28	27.17 ± 3.66	83.83 ± 7.58
II	Hypercholesterolemic control	170.50± 5.38 [a]	92.67 ± 7.39 [a]	151± 5.10 [a]
III	Treatment control (Rosuvastatin 10mg/kg)	111.66 ± 3.98***	41.17 ± 2.32***	99 ± 3.04***
IV	LUSE-100mg/kg	136.67 ± 3.21*	67.67 ± 2.66*	136.67 ± 3.5*
V	LUSE-200mg/kg	122.16 ± 3.18**	55.1 ± 4.74**	121.17 ± 3.19**

VI	LUSE-400mg/kg	113 ± 3.74***	48.33 ± 3.77***	103 ± 3.89***

Serum glutamate Oxaloacetate transaminase (SGOT), Serum glutamate pyruvate Transaminase (SGPT), alkaline phosphatase (ALP), *Linum Usitatissimum* Seed Extract (LUSE)

All the values are mean ± SD, n=6, data analyzed by ANOVA and post hoc test. [a]*P*<0.001 when compared to group I (vehicle control), **P* value <0.05– significant, ***P* value <0.01- highly significant, ****P* value <0.001- very highly significant when compared to Group II (Hypercholesterolemic control)

Discussion

Coronary heart disease is one of the leading causes of death in the world, this isattributable to genetic factors, unhealthy diet and lifestyle.The principal metabolic causes of atherosclerosis include hyperlipidaemia, hypertension and diabetes mellitus.Most of the population in India consumes food which is rich in oil and/or ghee, which increases the susceptibility to atherosclerotic vascular disease.Coconut oil and vanaspati ghee is also consumed by majority of population leading to development of hyperlipidaemia. The inclusion of plant-based foods in the diet aregaining importance as lifestyle changes in the fightagainst coronary heart disease. Several preclinical and clinical studies have shownthebeneficial cardiovascular effects of dietary supplementation with flaxseed[9].This study was undertaken to further evaluatethe hypolipidemic and hepatoprotective effects of flax seed extract on a rat experimental model using high fat diet prepared by a combination of vanaspati ghee and coconut oil. The advantage of this model was that it simulates the common man's diet in India and could be easily developed within a shorter period of time[11].

The results obtained from the study showed a significant increase inthe serum levels of TC, TG, VLDL, LDL, while the HDL levels decreased significantly in the high fat diet group, similar to the study by Munshi, Renuka P et al.[11]. A major risk factor for the development of cardiovascular disease is increased lipid parameters and decreased plasmaconcentrations of HDL-C[18].Supplementation of cholesterol in diet results in a marked increase in the production of cholesteryl ester rich-VLDL by the liver and intestine. Thehigh levels of LDL found in hypercholesterolemicratsmay be due to down regulation in LDL receptors bycholesterol and saturated fatty acids present in the high fat diet[19]. It is well documented that elevated total cholesterol and LDL levels promote atherosclerosis and cardiovascular complications. Elevated serum triglycerides are considered as independent risk factor for cardiovascular disease[20].The high levels of TG observed in the experimental rats may be due to inhibition of 7a-hydroxylase activity[21].The results of our study demonstrate thatflaxseed extract administration in high fat diet fed rats causeda significant decrease of TC, VLDL, LDL at all doses and a significant decrease in TG levels at only the highest dose.Many animal and human studies as well have demonstrated no changes in serum HDL-cholesterol and triglyceride (TG) levels. A meta-analysis of 28 studies have concluded that flaxseed produces no significant changes in HDL and TG levels. The same absenceof flaxseed effect on HDL and TG levels was observed in an interventional clinical trial[9].It is observed that whole flaxseed and flaxseed powder can moderately reduce LDLby 5-10% without affecting HDL or TG. Conversely, flaxseed oil does not reliably lower LDL-C, but can reduce TG at high doses[17].In our study, flaxseed extract did not produce any significant changes in the HDL levels, which is similar to a study where defatted flaxseed (2-3% ALA, but similar in lignan content to traditional flaxseed) reduced TC and LDL-C in rabbits fed an atherogenic diet for eight weeks when compared to controls, but had no effect on HDL-C[22].

Flaxseed, *Linum usitatissimum*also known as linseed, is one of therichest plant sources of ALA (alpha linolenic acid) an omega-3polyunsaturated fatty acid (PUFA). It also contains phytoestrogen "lignans," speciallysecoisolariciresinol diglucoside (SGD) and both insoluble, soluble fibers. Alpha-linolenic acid (ALA) is responsiblefor higher cholesterol secretion into bile leading toa depletion of the intrahepatic pool of cholesteroland thus to an increase in cholesterol synthesis and turnover, this being the major cholesterol-lowering mechanisms of flaxseed.ALA rich diet also decreases hepatic lipid accumulation both by stimulating β-oxidation and by inhibiting fatty acid synthesis[19]. ALA is further metabolized to the longer chain fatty acids eicosapentaenoic acid (EPA), docosapentaenoic acid (DPA), and docosahexaenoic acid (DHA). Eicosapentaenoic acid replaces arachidonic acid in membrane phospholipids, this may reduce the oxidative stress by scavengingreactive oxygen species and subsequently decreasing the activation of proinflammatory transcription factors. DPA is known to inhibit cyclooxygenase-I activity and cellularTNF- α activity, whereas EPA and DHA are known to be involved in various anti-inflammatory and antiatherogenic activities through activation of various peroxisome proliferator-activated receptors[9]. Animal studies indicate lignans and associated substances are responsible for reduced LDL and total cholesterol with flaxseed consumption rather than ALA. Dietary flaxseed lignans are known to possess lipid-lowering and antioxidant properties[17]. It is suggested that the lignans precursor SDG may directly lower serum cholesterolas well as through modulation of enzymes involved in cholesterol metabolism, including 7a-hydroxylase and acyl CoA cholesterol transferase[22]. Flaxseed fibres may exert its lipid-lowering effect by producing satiety and thereby reducing caloric intake. They may act byenhancing gastric emptying, decreasing transit time, increasing the rate of bile acid excretionand by reducing bile acid reabsorption through increased faecal excretion of cholesterol. Fibers may also getconvertedto small-chain fatty acids like acetate and propionate. These small-chain fatty acids can affect multiple metabolic pathways like fatty acid synthesis, oxidation, lipolysis, and cholesterol synthesis. Increased levels of propionate also inhibit the conversion of acetate to lipids and 3-hydroxy- 3-methyglutaryl-CoA reductase, thus decreasinghepatic cholesterol synthesis[9].However, nutrientcomposition and bioavailability of flaxseeddepends on the form used.Three distinct forms of flaxseed areavailable: oil extract, whole, and milled(ground) seed.Flaxseed oil is composedof 73% PUFA (ALA and linoleic acid), 18% monounsaturated fatty acids, and9% saturated fatty acids.Soluble fiberand lignans are absent in the oil; however,oil has the highest ALA content and bioavailability.Nonetheless,flaxseed oil alone does not appear toimprove the lipid profile and associatedCVD risk. Other functional components associatedwith the lignin complex includecinnamic acid glucoside, an antioxidant,and hydroxymethylglutaric acid, whichhas hypolipidemic effects. Grinding flaxseed does not alter thecomposition, but it does improve ALAbioavailability, in part because wholeseeds may pass undigested[23], this justifies the use of ground flaxseed for the preparation of the LUSE extract.

Serum SGOT, SGPT, and ALP are the enzyme biomarkers used tomonitor the structural integrity of the liver, kidneys, heartand specifically help in theclinical diagnosis of liver damage[24]. In case of injury to the organs due to any reason, these enzymes leak into the blood stream. Therefore, AST, ALT and ALPlevels in rat serum were examined to detect the liver damage due to HF diet and to analyse the hepatoprotective effect of flax seed extract on the induced liver damage. The reference ranges of AST,ALT and ALP are 50 to 150 IU/L, 10 to 40 IU/L and 30 to 130IU/L respectively[25]. In the high fat diet fed rats, liver which is the primary organ to metabolise the excess cholesterol, is affected by oxidative stress. This occurs due to imbalance between the production of free radicals and decreased effectiveness of antioxidant defence system. Increased oxidative stress plays an important role in the

chronic inflammatory responses to hypercholesterolemia and atherosclerosis. Thus oxidative stress leads to cell damage related to free radicals and lipid peroxidation, including the destruction of the cell membrane structure and elevation of the liver enzymes in the hypercholesterolemic rats[26].

Our study showed that the flaxseed extract prevented elevation of liver enzymes in rats on HF diet, demonstrating a hepatoprotective effect on the proposed oxidative liver damage. This might be attributed to the fact that ALA needs specific in vivo conditions to produce its metabolite, namely dihydrolipoic acid, which has been also reported to be hepatoprotective[26]. Our observation is consistent with other studies on flax and pumpkin seed mixture which have suggested anti-atherogenic and hepatoprotective effects, probably mediated by unsaturated fatty acids present in seed mixture[27]. Another study reported thatflaxseed *chutney*possesses antioxidant and significant hepatoprotectiveproperties[28].

Soluble flaxseed gum, also called mucilage, is aseries of water-soluble polysaccharides from the husk of flaxseed. In vitro assays have demonstrated the powerful antioxidant activity ofsoluble flaxseedgum. In addition to polysaccharides, oligosaccharidesfrom flaxseed exhibit biological activity as well. These oligosaccharideshave several physiological functions, such as regulation ofgastrointestinal function, liver protection, and antitumor, reducingthe risk of cardiovascular disease. The monosaccharidecomposition of flaxseed oligosaccharide includes rhamnose,fructose, xylose,mannose, arabinose, glucose, and galactose.The flaxseed oligosaccharide exhibited a strong potential toscavenge hydroxyl radicals. The proposed antioxidantmechanism for the flaxseed oligosaccharide involves hydroxylradical inducing abstraction of the hydrogen atom bonded to acarbon on the carbohydrate. The resultant carbon centredradical then reacts with hydroxyl radical to form carbohydratehydroxylradical complexes[29,30]. It may be valuable to conduct further studies to assess the antioxidant activity of flax gum in cardiovascular diseases that may have a component of oxidation-like atherosclerosis.
Through this study, we observed thatraised TG, HDL levels, which are important predictors of cardiovascular diseaseare not reduced significantly by the flaxseed extract.Presently used hypolipidemic drugs are associated with various complications. Plant derived products like flaxseed are relatively devoid of such side effects, also flaxseed consumption did not interfere and was additive to the cholesterol-lowering effects of cholesterol-lowering medications like statins[9]. Hence, these can be combined with other drugs like statins whose dose can thus be reduced to achieve the desired therapeutic effect, especially in patients who are not able to tolerate the higher doses of statins due to the adverse effects. Flaxseed supplementation can be given in hypertensive, diabetic and obese individuals at risk of hyperlipidaemia. But, more studies are needed in larger populations to determine the dose of flaxseed as adjuvant therapy in patients and as supplements in high risk population.

5. CONCLUSION

From this study, it can be concluded that the addition of the flaxseeds to the diet may alleviate the rise in circulating cholesterol levels induced by the high cholesterol diet through its content of ALA and lignins. The hepatoprotective role of flaxseed in hypercholesterolaemia has been demonstrated in this study. Hence, flaxseed or flaxseed powder can be a useful part of a cholesterol-lowering diet, such as the Therapeutic Lifestyle Changes (TLC) diet, which was recently endorsed by the National Cholesterol Education Program.

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7. COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research is commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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