



## Original Research Article

## Evaluation of hypolipidemic effect of flaxseeds in experimentally induced dyslipidemia in albino rats

Neha Bhasin<sup>1,\*</sup>, Jyoti Batra<sup>1</sup>, Suman Lata<sup>2</sup>, Kavita Dhar<sup>3</sup>

<sup>1</sup>Dept. of Biochemistry, Santosh Medical College, Ghaziabad, Uttar Pradesh, India

<sup>2</sup>Dept. of Pharmacology, Muzaffarnagar Medical College, Muzaffarnagar, Uttar Pradesh

<sup>3</sup>Dept. of Pharmacology, Santosh Medical College, Ghaziabad, Uttar Pradesh, India



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## ABSTRACT

**Introduction:** The most important risk factor of IHD is hyperlipidaemia. The dietary manipulation with flaxseed powder has reduced the incidence of CVD by decreasing the lipid profile in human beings. So flaxseed powder has been used in the present study to analyse its hypolipidemic action in experimentally induced hyperlipidaemia in male albino rats.

**Materials and Methods:** This is an animal interventional study. It was conducted in 24 male albino rats divided into 4 groups with 6 animals in each. Gr I (control), Gr II to see the per se effect of flaxseed powder, Gr III (standard) kept on High cholesterol diet and Gr IV (test) kept on high cholesterol diet with flaxseed powder. The study was conducted for two weeks. Blood samples for estimation of TC, TGc, LDLc, VLDLc and HDLc were collected at the beginning and at the end of the study period from orbital vein.

**Results:** In the intervention group fed on flaxseed powder and HCD significantly reduced the blood lipids but not in the Gr II fed on flaxseed alone. The HCD fed Standard group developed a highly significant rise in lipid levels of the rats. This has revealed that incorporation of flaxseed in diet may protect against the development of IHD. The hypolipidemic effect of flaxseeds may be due to the presence of rich content of omega 3 fatty acids, ALA, Lignans and antioxidant properties and phytoestrogens.

**Conclusion:** So, we conclude that flaxseed powder administration has highly significantly reduced the blood lipid levels in rats. The addition of flaxseed in diet has a beneficial effect on lipids and protects against cardiovascular disorders due to omega 3 fatty acids, ALA and Lignans.

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### 1. Introduction

With a spectacular rise in living standards economic upliftment and industrial growth, there had been an increase in stress and strain in daily life. This rise in economy has in turn resulted in an increased expenditure on luxury items and junk food and has resulted in a sedentary lifestyle among the people, which has raised the number of premature deaths from obesity, hyperlipidaemia, diabetes mellitus, hypertension and ischemic heart diseases (IHD).<sup>1</sup> Hyperlipidaemia is one of the most important risk factor of atherosclerosis and thus of IHD.<sup>2,3</sup> Besides medical treatment nutritional manipulations in the form of addition

of certain indigenous substances to diet may decrease the blood lipid levels and prevention of the occurrence of cardiovascular disease (CVD). The increased use of omega ( $\omega$ )-3 fatty acids is a good example of one such dietary change that may produce significant benefits.<sup>4</sup> Sea food products are rich dietary sources of  $\omega$ -3 fatty acids while Flaxseed is an alternative to sea food. It is one of the richest sources of the plant-based  $\omega$ -3 fatty acid, alpha-linolenic acid (ALA) and lignans and have a positive impact on CVD.<sup>5-7</sup>

This plant is a member of genus linium, family linaceae and it grows all over the world and in India it is grown during the winter season. Its botanical name is Linium Usitatissimum. The plant is rich in fibre, which his used for

\* Corresponding author.

E-mail address: [nehabhasin130@gmail.com](mailto:nehabhasin130@gmail.com) (N. Bhasin).

preparation of cloth called Lenin so it is named Linium. Flax seed oil is known as linseed oil and is used as an edible oil.

Flaxseeds consumption daily containing omega 3 fatty acids is now commonly advocated as hypolipidemic, antiatherogenic and cardioprotective agent. So, it was well-thought-out to be evaluated for its hypolipidemic activity in experimentally induced hyperlipidaemia in rats.

## 2. Materials and Methods

The experimental study was done on animal in the Department of Pharmacology at Muzaffarnagar Medical College, Muzaffarnagar in collaboration with the Biochemistry Department of Santosh Medical College, Ghaziabad. The study was conducted from February 2019 to April 2019. The ethical approval was taken from the Institutional Animal Ethical Committee (IAEC) before the commencement of the study.

The study was conducted on 24 male albino rats weighing between 200-220 grams. The rats were familiarized to the experimental conditions for 1 week and were grouped and housed in standard polypropylene rat cages (three rats per cage) during the experiment. They were maintained at 25°C and a 12 hrs interval light/dark cycle and provided standard laboratory animal feed and water and libitum throughout the experimental period of 2 weeks.

Cholesterol AR grade was procured from the SRL fine chemicals. Flaxseed were bought from the local market.

The subjects were divided into 4 groups containing 6 rats in each:

Group I served as Control and was fed on commercial pallet diet and water at lib for 2 weeks.

Group II to see the per se effect of flaxseed powder was fed on normal commercial pallet diet and flaxseed powder (3g/kg body weight)

Group III served as Standard and was given high cholesterol diet daily containing 2% cholesterol and 15% saturated fatty oil to the usual pallet diet and water at lib for two weeks.

Group IV served as Test and were supplied with flaxseed powder (3g/kg body weight) daily in addition to the high cholesterol and usual commercial pallet diet for two weeks.

The blood samples were collected after overnight fasting from the orbital vein by making a puncture near medial epicanthus of the eye with the help of a capillary tube from each rat separately before starting the experiment and was again withdrawn after 2 weeks for the estimation of blood lipids namely Total cholesterol (TC), Serum Triglycerides (TG), Low Density Lipoprotein Cholesterol (LDLc), High Density Lipoprotein Cholesterol (HDLc) and Very Low Density Lipoproteins (VLDL).

The total cholesterol was measured by: Cholesterol Oxidase/ Peroxidase Method (CHOD – POD Method),<sup>8</sup> HDL Cholesterol was measured by Direct Enzymatic Method,<sup>9</sup> LDL Cholesterol was measured by Friedewald

Equation:  $LDL = TC - (HDL + TG/5)$ ,<sup>10</sup>

Serum Triglycerides were measured by: Glycerol – 3 – Phosphate Oxidase Method (GPO).<sup>11</sup>

The statistical analysis was done using Two Way ANOVA and Tukey (post hoc analysis).

## 3. Results

In this study the analysis of blood samples of rats of all the four groups (Gr I, Gr II, Gr III and Gr IV) for lipid profile was done at the beginning of the study and after two weeks of feeding of HCD (high cholesterol diet) or HCD and Flaxseed Powder. It revealed no significant change in Gr I and Gr II rats fed on normal commercial pallet diet (NCPD) alone and NCPD with flaxseed powder after two weeks of feeding. While a highly significant increase in lipid profile was noted after two weeks in Gr III subjects fed on HCD (high cholesterol diet) along with NCPD. The Gr IV subjects fed on flax seed powder with HCD and NCPD showed a significant prevention from hyperlipidaemia.

There was no significant difference in the initial values of TC, TGC, LDLc, VLDLc and HDLc of all the four groups (Table 1). But after two weeks there was a significant change in Gr III and IV subjects. In Gr I (fed on NCPD) and II (fed on flaxseed powder and NCPD) after two weeks the blood samples revealed no significant change in the lipid levels of TC, TGC, LDLc, VLDLc and HDLc in comparison to the initial values showing that flaxseed administration per se does not affect the normal lipid levels.

Feeding of HCD (High Cholesterol Diet) to the Gr III rats for two weeks induced a highly significant hyperlipidaemia as revealed by a rise in TC (204mg/dL±25.62), TGC (260mg/dL±28.6), LDLc (156mg/dL±16.5) and VLDLc (52mg/dL±7.3) in comparison to the lipid level of Gr I rats (Table 1, Figures 1, 2, 3, 4 and 5), but no significant change in HDLc (25mg/dL±4.6). Administration of flaxseed powder along with HCD to Gr IV animals for two weeks produced a highly significant decrease in the TC (158mg/dL±17.2), TGC (109mg/dL±16.8), LDLc (128mg/dL±11.7), VLDLc (21.8mg/dL±5.3) in comparison to Gr III animals and a significant increase in HDLc (36mg/dL±6.2) level was observed (Table 1, Figures 1, 2, 3, 4 and 5). This shows that the flaxseed powder administration reduces only the raised lipid levels but does not affect the normal lipid profile.

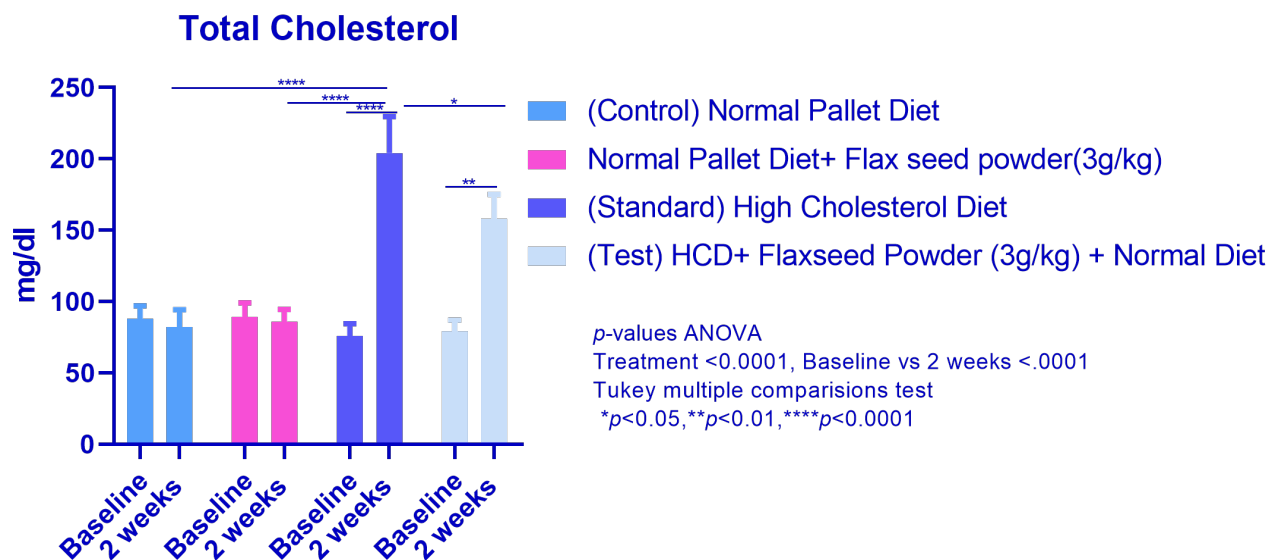
## 4. Discussion

The present study has been undertaken to demonstrate the effect of flaxseeds powder on lipid profile in hyperlipidemic as well as on normal rats (per se effect). To prevent the occurrence of hyperlipidaemia and thus of CVD with nutritional interventions is an important therapeutic strategy. Now a days the use of flaxseeds is common to reduce the risk of CVD because its rich ALA and omega 3 fatty acids

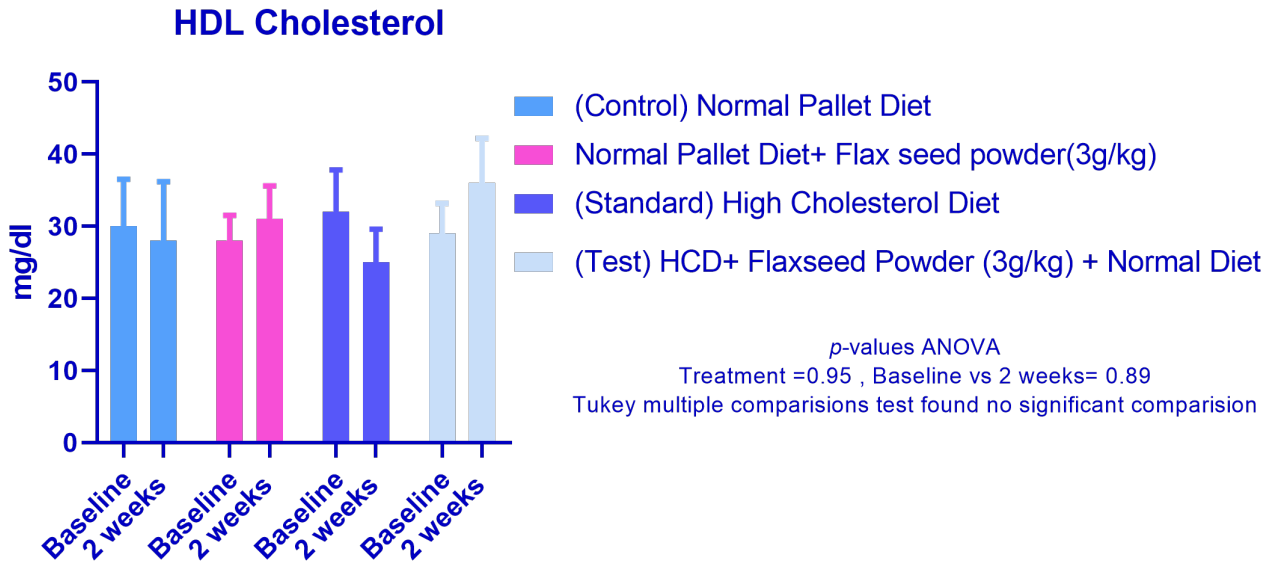
**Table 1:** Effect of high cholesterol diet and flaxseed powder administration for two weeks on lipid profile in rats (N=6)

Experimental Group (N=6)	Total Cholesterol (mg/dL)		Triglyceride Cholesterol (mg/dL)		LDL Cholesterol (mg/dL)		VLDL Cholesterol (mg/dL)		HDL Cholesterol (mg/dL)	
	Initial	2 Weeks	Initial	2 Weeks	Initial	2 Weeks	Initial	2 Weeks	Initial	2 Weeks
<b>Gr I (Control)</b> Normal Pallet Diet	88±9.08	82±12.3	72±16.4	74±17.6	96±8.9	103±12.5	14.5±3.9	14.8±4.4	30±6.5	28±8.2
<b>Gr II</b> Normal Pallet Diet+ Flax seed powder (3g/kg)	89±10.1	86±8.5	79.5±8.9	74.8±9.6	104±10.5	96±11.2	15.9±4.5	12.8±3.8	28±3.5	31±4.6
<b>Gr III (Standard)</b> High Cholesterol Diet	76±8.5	204±25.62	68±12.9	260±28.6	81±9.1	156±16.5	13.6±5.1	52±7.3	32±5.8	25±4.6
<b>Gr III (Test)</b> HCD+ Flaxseed Powder (3g/kg) + Normal Diet	79±8.06	158±17.2	65±14.8	109±16.8	86±10.4	128±11.7	13±4.1	21.8±5.3	29±4.2	36±6.2

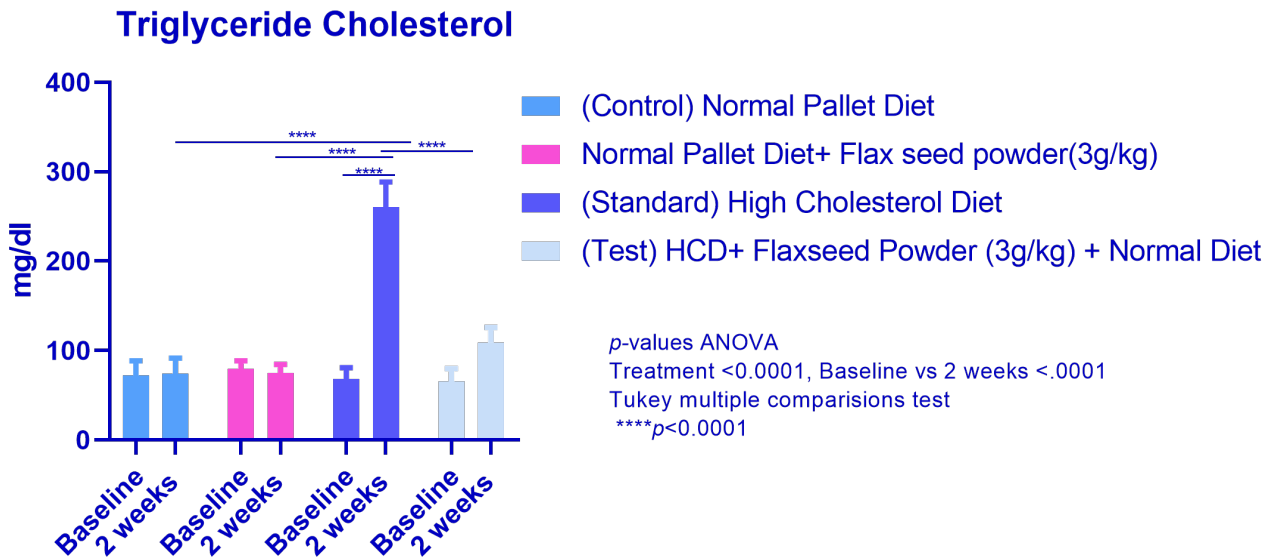
Data are presented as (Mean ± S.E), S.E = Standard error.



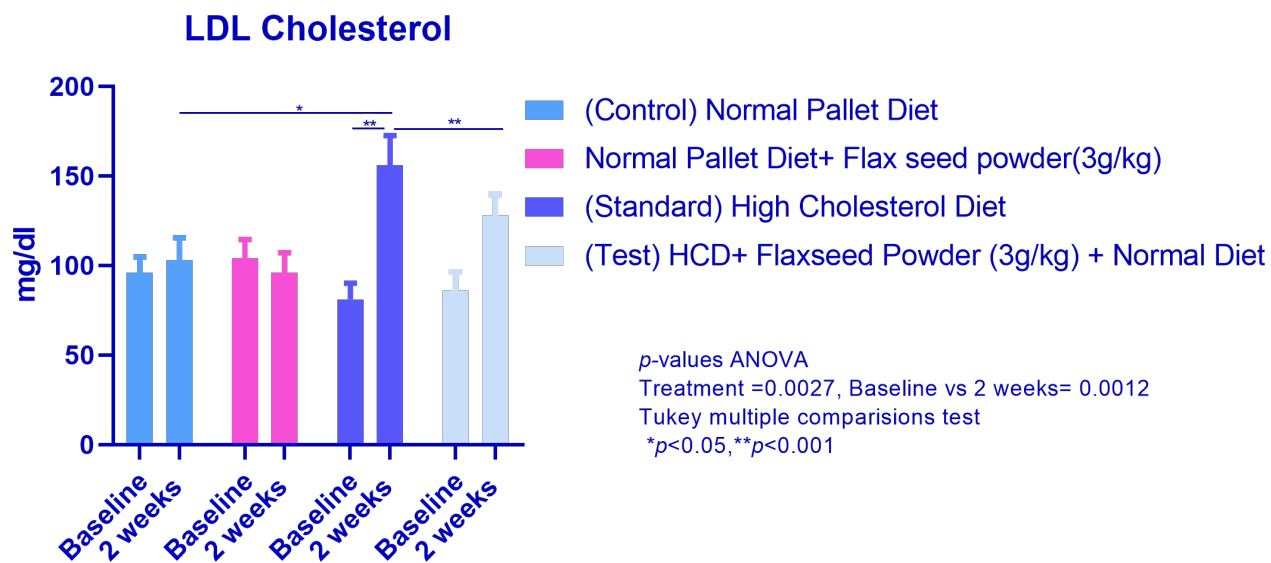
**Fig. 1:** Effect of High Cholesterol Diet with Flaxseed powder administration daily for 2 weeks on Total Serum Cholesterol Level in male albino rats. (N=6)



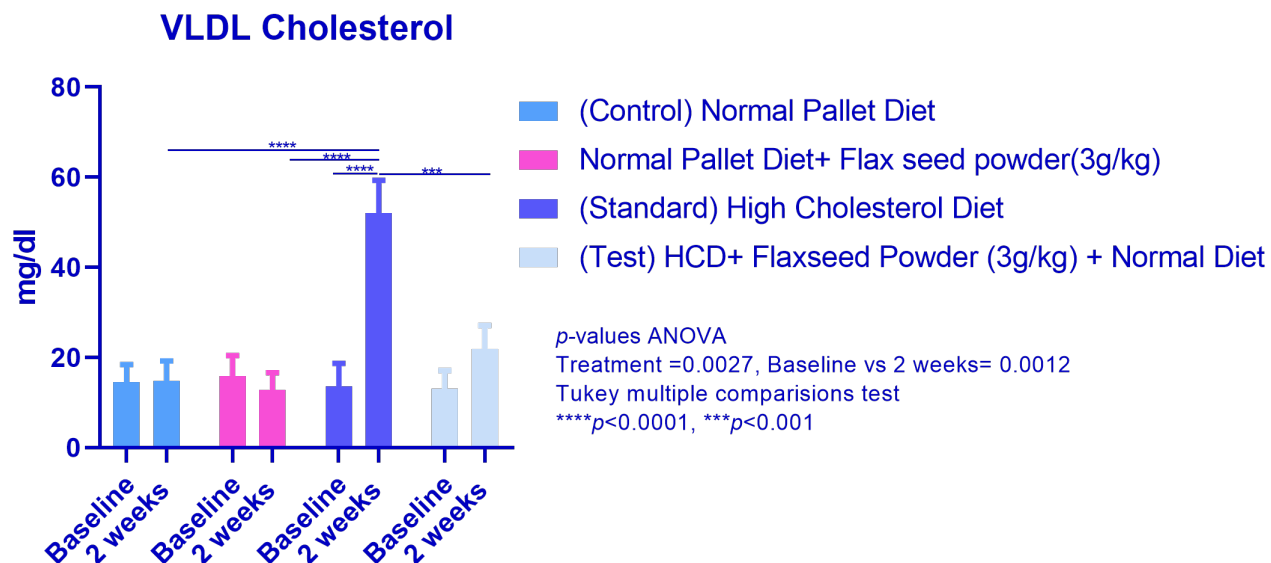
**Fig. 2:** Effect of High Cholesterol Diet with Flaxseed powder administration daily for 2 weeks on High Density Lipoproteins(HDLc) Level in male albino rats. (N=6)



**Fig. 3:** Effect of High Cholesterol Diet with Flaxseed powder administration daily for 2 weeks on Triglyceride Cholesterol Level in male albino rats. (N=6)



**Fig. 4:** Effect of High Cholesterol Diet with Flaxseed powder administration daily for 2 weeks on Low Density Lipoproteins (LDLc) Level in male albino rats. (N=6)



**Fig. 5:** Effect of High Cholesterol Diet with Flaxseed powder administration daily for 2 weeks on Very Low Density Lipoproteins (VLDLc) Level in male albino rats. (N=6)

contents that has the potential to reduce TGs, blood pressure and CVD.<sup>12</sup>

The rats of Gr I and II did not show any significant change in lipid levels of TC, TGc, LDLc, VLDLc and HDLc after two weeks of feeding of usual rat feed or usual rat feed with flaxseed powder. This indicates that flax seed does not alter the normal lipid levels i.e. flaxseed per se has no effect on normal blood lipids (Table 1, Figures 1, 2, 3, 4 and 5). Feeding of high cholesterol diet (HCD) to Gr III rats for two weeks produced a very highly significant increase in TC, TGc, LDLc and VLDLc but no significant effect on HDLc. In Gr IV animals addition of flaxseed powder to HCD for two weeks significantly prevented the rise in TC, TGc, LDLc, VLDLc and a significant increase in HDLc. This result corroborates with the findings of a study which provided evidence that consumption of flaxseed significantly reduced TC and significantly increased HDLc concentration.<sup>13</sup> (Table 1, Figures 1, 2, 3, 4 and 5). A major risk factor for the development of CVDs is decreased plasma concentrations of HDL-C.<sup>14</sup> This shows a protective effect of flax seeds against CVD.

This shows a preventive action of flax seed powder against the development of hyperlipidaemia and atherosclerosis and thus CVD. These results are in accordance of Maryam Torkan et al.<sup>15</sup> and O M Ipatova et al.<sup>16</sup> Flaxseed is a class of phytoestrogens which is rich in dietary lignans, is known to be lipid-lowering and has antioxidant properties.<sup>17</sup> The hypolipidemic effect of flax seed may be due to the presence of omega 3 fatty acids and ALA. The ALA is metabolized to eicosapentaenoic acid, which replaces arachidonic acid in membrane phospholipids. The ingestion of flaxseed powder can alter the generation of eicosanoids and thus its procoagulant activity by reducing thromboxane A2 synthesis and other membrane-dependent responses to produce anti-allergic and anti-atherosclerosis effect.<sup>16</sup>

The addition of the flaxseeds to the diet may mitigate the rise in circulating cholesterol levels induced by the high cholesterol diet through its content of ALA.<sup>18</sup> This study is further supported by the findings of another study, which proposes that increased bile acid synthesis is one of the major cholesterol-lowering mechanisms of flaxseed.<sup>19</sup> Dietary flaxseed lignans, such as secoisolariciresinol diglucoside (SDG), also have demonstrated potent antioxidant and anti-atherogenic effects.<sup>20</sup>

## 5. Conclusion

It is well documented that the modification of lipid levels is a useful approach to reduce the incidence of cardiovascular diseases through prevention of development of atherosclerotic disease. So, the addition of flaxseed powder into diet is feasible, affordable, and well tolerated. It provides a good source of soluble fibre and ALA and thus reduction in CVD risk factors.

## 6. Source of Funding

Self funding.

## 7. Conflicts of Interest

None.

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**Suman Lata** Professor and Head

**Kavita Dhar** Associate Professor

### Author biography

**Neha Bhasin** Post Graduate Student

**Jyoti Batra** Professor and Dean Research

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