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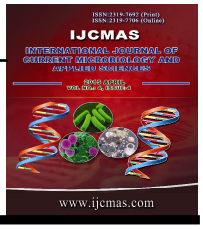

Original Research Article

Effect of fortification by full fat and defatted flaxseed Flour sensory properties of wheat bread and lipid profile laste

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Keywords

Flaxseed, bread, sensory evaluation, chemical composition, digestibility and hyper-cholesterolemic

A B S T R A C T

The research work was carried out to evaluate the acceptability of flaxseed utilization in the bread. The scores assigned to all the sensory parameters of breads affected significantly with the variation in levels of flaxseed fortified in wheat flours. The 10 and 15% replacements of both flaxseed flours resulted in acceptable product. Flaxseed flours (partially defatted and full fat) fortified wheat flour significantly improved the chemical composition (crude fat, crude fiber and crude protein), results showed that addition of flaxseed to wheat flour by (10%FFF+15%DFF) increased the protein, fiber, and ash, their content increased significantly with the increasing level of FFF and DFF supplementation in wheat flour. Meanwhile, the content of carbohydrates was decreased in the bread. Carbohydrates increased and fat decreased for bread 15% DFF. Also results revealed that, the diets containing flaxseed increased the level of serum HDL-C while serum TC, TG, LDL-C and VLDL-C significantly decreased.

Introduction

Flaxseed or linseed (*Linum usitatissimum L.*) has been used as food and medicines in many countries. It has been used in various forms such as flour, oil and seed. Flaxseed and flaxseed oil is considered as healthy due to presence of various bioactive compounds in it**.** Flaxseed has a history of food use in Europe and Asia. In the US, until the early 1990s flaxseed had been incorporated at low levels in some brands of cereal, bread, and other bakery products. However, during the past decade, potential health benefits associated with consumption of flaxseed and flaxseed meal have become more prominent (Jenkins *et al*., 1999 and Yamashita *et al*., 2003). Flax seed as a nutritional additive for the preparation of certain dietary items like baked products, ready to eat cereals and fiber bars having good health impacts has been widely recognized in all parts of the world (Rendon- Villalobos *et al*., 2009). Acceptance of flaxseed as a dietary functional food ingredient in cakes assessed at structured nine point hedonic scale

revealed consumer acceptance up to 30% supplementation level (Moraes *et al*., 2010). The baked products can be supplemented with whole flaxseed grains to achieve an attractive and appealing form with enhancement in the texture of final product. The grinding of flaxseed before its addition to products can be more beneficial to obtain the prospective health benefits from its active components like dietary fiber, lignin and Omega-3 fatty acids. Several researchers has investigated that ground flaxseed or whole flaxseed grains can be replaced wheat flour used for the production of pancake, muffins, breads and cookies (Muir and Westcott, 2000; Manthey *et al*., 2002).

There is a small difference in using the terms flaxseed and linseed. Flaxseed is used to describe flax when consumed as food by humans while linseed is used to describe flax when it is used in the industry and feed purpose (Morris 2008 and Thompson, 1995). Oils rich in unsaturated and poly unsaturated fatty acids and tocopherol are preferred to be added in infant's formulas and different food products to gain maximum nutraceutical and health related properties (Moyad, 2005; Bozan and Temelli, 2008). The seed is a healthy source of oil containing poly-unsaturated fatty acids, digestible proteins, and lignin. Major nutritional components of flaxseed include ALA rich oil, protein, minerals and a greater proportion of non nutritional lignin-rich dietary fiber. On the dry weight basis, flaxseed contains 20% protein, 27% total dietary fiber 41% oil, 4% ash and 8% moisture (Madhusudhan, 2009). Flaxseed oil is naturally high in anti-oxidant like tocopherols and beta-carotene, traditional flaxseed oil gets easily oxidized after being extracted and purified (Holstun and Zetocha 1994). It is therefore not surprising that flaxseed is the most prominent oilseed studied to date as a functional food, since it is a leading source of ALA (52% of total fatty acids) and of phenolic compounds known as lignin (Oomah and Mazza, 2000). There is evidence that whole flaxseed may lower serum cholesterol in both normal and hyperlipidemic subjects (Bierenbau *et al*., 1993). The major fatty acid in flaxseed were linolenic acid C18:3 (44.0%), oleic acid C18:1 (26.20%) and linoleic acid C18:2 (18.36%) (Horshid *et al*., 2010). While, Harper *et al*., (2006) found that, the fatty acids composition in flaxseed oil were C18:3 (58.9%), C18:2 (17.9%), C18:1 (16.9%), C16:0 (6.91%) and C18:0 (3.90%), respectively. Daily dietary consumption patterns of flaxseed in hypercholesterolemic patients have been reported to lower down the total cholesterol to a significant extent (Dahl *et al*., 2005; Bassett *et al*., 2009). Special diets can be formulated using flaxseed oil to meet the standard health requirements. (Rendon- Villalobos *et al*., 2009). Lignan reduces "bad" LDL-cholesterol (LDL-C) levels thus having a good impact on cardiovascular health. Lipid profile improving effect of flaxseed has long been studied and many literatures have related its effect to high fiber, ALA and lignin content of flaxseed (Arjmandi *et al*., 1998

The combined findings from nine clinical trials suggest that whole or ground flaxseed (15-50 g/day) can modestly reduce total and LDL cholesterol by 1.6 to 18% in both hypercholesterolemic and normocholesterolemic patients without significant changes in HDL or triglyceride levels (Bloedon and Szapary 2004). Flaxseed can be incorporated into the diet through oil, milled or ground flaxseed and animals fed flax meal (Vaisey-Genser and Morris, 1997). The U.S. Food and Drug Administration allow the inclusion of up to 12% (by weight) of flaxseed in food.

The objective of the present study was to prepare bread supplemented with the full fat and defatted flaxseed as a functional food for improves organoleptic properties, the nutritive value and evaluation hypercholesterolemic rats.

Materials and Methods

Materials

Bread ingredients including, full and defatted flaxseeds (2013 year of production) were obtained from Agricultural Research Center, Oil Crops Department, Giza, Egypt. Wheat flour (82%) extraction, sodium chloride, sugar, skimmed milk, linseed oil, instant dry yeast, was purchased from the local market in Kafr elsheikh Governorate, Egypt.

Male albino rats of Sprague Dawley (110 ± 10g) strain were obtained from the Agricultural Research Center, Giza, Egypt. Cholesterol powder obtained from ALgomhoria Co. for Trading in Medicines, Chemicals and Medical Supplies, Cairo, Egypt.

Methods

Preparation of flaxseeds

The cleaning of flaxseed was performed manually to remove damaged seeds, dust particles, seeds of other grains/crops and other impurities such as metals and weeds. After cleaning, the seeds ground using a laboratory mill and screened through a 0.25 mm sieve. Fluor obtained from raw flaxseed was divided into two batches; first one was left as a full fat and another one was defatted by solvent extraction in a Soxhlet apparatus (Tecator Inc., Colorado, USA) for 8 h using n-hexane. The defatted Fluor from flaxseeds were spread on aluminum trays and dried in a hot-air fan oven (70°C, 30 min) to expel residual hexane and stored in air-tight plastic containers, then full fat seed and defatted seed were kept stored at 4°C until used.

Preparation of the toast bread

The toast bread were prepared with Partially 10% full fat flaxseed flour, 15 % defatted flaxseed flour and 10% full fat flaxseed flour + 15 % defatted flaxseed flour fortified straight grade flours following the straight dough method as described in AACC (2000) method No. 10-10B.The ingredients were mixed mechanically for 5-10 minutes to form dough and allowed to ferment at 30°C and 75% R.H. for 180 minutes. First and second punches were made after 120 and 150 minutes, respectively. The dough was divided into 3 parts and final proofing was done for 45 minutes at 95 °F (35 °C) and 85% R.H. The toast bread was baked at 232 °C for 45 minutes, the different ingredients used in preparation toast bread are shown in Table (1), provided that several formulae were tested and the best one presented.

Analytical methods

Sensory evaluation

Sensory evaluation was performed using 20 graduate students and staff members in Home Economic Dept., Fac. of Specific Education, Kafr elsheikh Univ., Egypt. Sensory evaluation was performed 24 hours after baking to evaluate loaf appearance, crust color, crumb color, taste/flavor and overall acceptability of the bread sample. The bread samples were sliced into pieces of uniform thickness and served with water. To perform the evaluation, Panelists evaluated bread samples on a 9 point hedonic scale quality analysis with 9 = liked extremely, 8 = liked very much, 7 = liked, 6 = liked mildly, 5 = neither liked nor disliked, 4 =

disliked mildly, 3 = disliked, 2 = disliked very much and 1 = disliked extremely according to Larmond (1997). The objective sensory quality of bread is described by its sensory profile which is constituted by sensory attributes according to (Lawless and Heyman 1999). These attributes tend to be perceived in the following order: appearance, aroma, texture and flavor (Meilgaard *et al*., 1991).

Grosse of chemical composition

The contents of moisture, crude oil, crude protein (N x 6.25), crude fiber and total ash of toast bread fortified with full and defatted flaxseed were determined as described in A.O.A.C. (2000). The carbohydrate content was calculated by difference.

Caloric value was calculated from the sum of the percentages of crude protein and total carbohydrates multiplied by a factor of 4 (kcal.g-1) plus the crude fat content multiplied by 9 (kcal. g-1), according to (Zambrano *et al.*, 2004).

Determination of fatty acids composition

Fatty acid contents were performed in the Central Laboratory of the Faculty of Agriculture, Alexandria University. Samples were hydrolyzed using GC Chromatography Model, Shimadzu-4CM (PFE), equipped with FID detector and glass column 2.5 X 3 mm i.d, under the following conditions that first used by (Radwan, 1978).

Determination of antioxidants in full fat and defatted flaxseeds

Alpha-tochopherol and β-carotene were performed in the Central Laboratory Unit for Advanced Environmental and Biological Analysis. High Institute of Public Health, Alexandria University.

1-α –tochopherol

Alpha-tochopherol was extracted and determined using HPLC according to (Chase *et al*., 1994).

2-β-carotene

β-carotene was extracted and determined using Shimadzu HPLC according to the methods described by (Tee et al.,1996).

A method was described for the simultaneous analysis of α-tocopherol, and β-carotene in n-hexane extracts of ground seeds. The technique uses normal-phase HPLC in a silica gel column with n-hexane-2-propanol (97:3, v/v) as the mobile phase. The compounds are eluted from the column in the order α-tocopherol and β-carotene (internal standard). The compounds are quantified by use of the following detectors connected in series: β-carotene by a filter photometer, and α-tocopherol by a spectrophotofluorimeter. The time taken for separation is less than 6 min but a further 14 min is allowed for elution of the more polar compounds before the next run

Determination of lignin

Lignin content in the investigated ground seeds were determined according to (Fahmi, 1984) as follows:

One-gram sample was extracted at first with a mixture of method and benzene (1:1). In a wide mouthed bottle of 150ml capacity, 50ml of 38% pure HCL (not less than 38%) was added. The mixture was left for two minutes, and then 50ml of concentrated sulfuric acid was added. After shaking for one hour the mixture was left for 24th at 22 °C and then 30ml of distilled water were add. The content was poured in a beaker of one-liter capacity. The bottle was washed

with 415 ml of distilled water and the wash was added to beaker and filtered and washed with hot distilled water. The obtained lignin was dried at 105 °C for 3hrs and weighted.

In –vitro protein digestibility

For the investigation of in – vitro digestibility, the method used by Salgo' *et al*., (1985) as follow: A digestive enzymes (Trypsin) system was used in pH – drop method. The sample containing 200 mg of protein was suspended in 25 ml of distilled water and the PH was adjusted to 8.0 at a temperature of 37±0.1 ºC. While mixing continuously, 0.5 ml of Trypsin from bovine pancreas 7500 units per mg, solution of 8.0 mg / ml concentration (freshly prepared and put in ice) was injected in to the test solution. The pH was measured exactly after 10 min. of incubation at 37 ºC and the true and apparent digestibility was calculated from the following equations:

%True digestibility (TD) = 425.78- 47.64 PH10
% Apparent digestibility (AD) = 392.51- 44.84 PH 10
Where: PH 10 = the PH value after 10 mi

Biological assay

Animals and experimental design

This work was reviewed and approved by the animal Care and Wel-fare Committee Kafr elsheikh University. Forty tow male albino rats of Sprague Dawley (110 ± 10g) strain were housed in stainless steel, screen cages and allowed free access to food and water. Cages were maintained at 25°C and 50% relative humidity with a 12h cycle of light and dark. The basal diet composition was as mentioned in Table (2) (Cadden *et al*., 1983). The control, full fat, defatted flaxseed and full fat +defatted flaxseed toast bread were dried, finely ground and then incorporated into the basal diet at 666.7 g/kg at the expense of corn starch content. Diets were stored at -20°C to prevent spoilage and provided to animals daily. After the adaptation period, the preliminary body weights were initially recorded and rats were then divided into 6 experimental groups (7 rats each). All animals were fed a basal diet for one week, then rats were fed for six weeks according to the following scheme:-

G1: Rats fed on basal diet negative control;
G2: Rats fed on hypercholesterolemic diet positive control;
G3: Rats fed on control toast bread;
G4: Rats fed on hypercholesterolemic diet + toast bread with 10 % full fat and flaxseeds;
G5: Rats fed on hypercholesterolemic diet + toast bread with 15 % defatted flaxseeds;
G6: Rats fed on hypercholesterolemic diet + toast bread with 10 % + 15% full fat + defatted flaxseeds;

Food intake was recorded daily and body weights were recorded weekly over the sex weeks experimental period. At the end of the experiment period, all rats were fasted overnight anaesthetized by diethylether and sacrificed; blood samples were withdrawn in tow test tubes. The whole blood collecting in tube containing Ethylenediaminetetraacetic acid (EDTA) was used for estimation of some biochemical parameters; the other tubes of blood were left for coagulation then centrifuged at 3000 rpm for 15 minutes to obtain serum for further analysis. The obtained serum was kept in deep freezer at -20 °C for the subsequent analysis. Each diet was prepared to give equal nutritional value as control casein diet .Rats were weekly weighted through the feeding period (45 days), food intake, body weight gain (BWG) and food efficiency ratio (FER) were calculated at the end of experiment according to Chapman *et al*., (1959).

BWG = Final weight –Initial weight
FER = Body weight gain (g/day) / Food intake (g/day)

Biological analysis of serum

Triglycerides (TG) were determined according to the method of (Fossati and Prencipe, 1982).Total cholesterol (TC) was determined in serum according to the method of (Allain *et al*., 1974). High Density Lipoprotein cholesterol (HDL-cholesterol) was estimated in serum as described by Lopez-Virella *et al*., (1977). Low Density Lipoprotein cholesterol (LDL-cholesterol) was calculated as described by Lee and Nieman (1996) as following:
Low density lipoprotein cholesterol (LDL-cholesterol) was calculated according to the method of Lee and Nieman (1996) using the following:

VLDL = TG / 5 LDL = TC - (VLDL + HDL)

A therogenic index of plasma (AIP)

The atherogenic ratio of TC/HDL-C as well as the AIP, calculated as log(TG/HDL-C), with TG and HDL-C, were measured in molar concentrations (Dobiasova and Frohlich, 2001).

Statistical analysis

Data was expressed by mean ± SD. Data analysis was performed using Minitab software version 15.1.1.0 (Minitab Inc. USA). Before and after treatment differences were measured using paired t-test and the probability of a Type error was set at 5%. Between groups differences were analyzed by one-way analysis of variance (ANOVA), followed by one-way multiple comparison of Tukey and p values lower than 0.05 were considered as statistically significant differences.

Result and Discussion

Sensory evaluation

Effect of Incorporation of full fat and defatted flaxseed flour on the sensory scores (9 Point Hedonic Scale) toast Bread .The results concerning sensory evaluation of toast bread produced from different selected levels of full fat and defatted flaxseed flour used are shown in Table (1). It could be noticed from Table (3) that toast bread treatments made from 100% wheat flour (extraction 82%) was characterized with high acceptability for all parameters.

There is no significant ($p \leq 0.05$) different were observed with regard to sensory evaluation (taste, flavor, appearance and overall acceptability) of wheat toast bread when prepared after incorporation of selected (10%full fat and 15%defatted flaxseed flour). (Hussain *et al.*, 2011) concluded that addition of up to 12% full fat and partially defatted flaxseed flour in the pan breads does not negatively affect the sensory scores. While toast bread prepared by combination (10% FFF+15 % DFF) It is still acceptable in previous parameters as compared to the control toast bread but it was liked mildly by the panel members.

Similarly, organoleptically acceptable cookies can be prepared by supplementing 20% flax in foods as an ingredient (Hussain *et al*., 2006). Also, flaxseed can be used to improve the nutritive value of bakery products as well as for improving sensory properties (Kaur *et al*., 2013).Significant decrease in assigning scores to all the sensory attributes at toast breads exceeding flaxseed supplementation in (82%) wheat flour extraction to 25%. In the present study,

assignment of fewer score (neither liked nor disliked) by the panelists to the crust color and crumb color of (supplementation15% DFF and 10% FFF +15% DFF) toast breads attributed to the darker color of flaxseed flour imparted to resultant breads as the flaxseed meal (flour) is darker than (82%) wheat flour extraction. The results of the present study are inconformity with the work of (Frank and Sarah 2006) who found that addition of 15% flaxseed flour in bread negatively affected the volume, crust color and crumb color of breads.

The work of other researchers also supported the findings of the present study as (Koca and Anil 2007) showed that crumb darkness increased by increasing the level of flaxseed flours levels. Naz, (2000) observed that breads exceeding15% flaxseed supplementation in wheat flour resulted in lower scores for texture, crumb color, grain, and volume and crust colors.

Gross chemical composition

Chemical composition of produced toast bread from the obtained results in Table (4), it could be noticed that the chemical composition of toast bread which were produced from 100% wheat flour (82% extraction) were, protein (10.16%), ether extract (3.52 %), ash (1.68%), crude fiber (2.87%) and total carbohydrates (81.76%), respectively. While the chemical composition of produced toast bread with 10 % FFF,15% DFF and the incorporation(10 % FFF+15% DFF) substitution, it could be observed that increase in all constituents of chemical composition the protein content, fat, ash and fiber except the total carbohydrate and Kcal/100g were decreased. The protein content increased significantly with the increasing level of FFF and DFF supplementation in wheat flour. The crude fiber content of produced toast bread also varied significantly among different composite flours supplemented with DFF and FFF. The highest content (4.30%) was found significantly with bread (10%FFF+15%DFF) these results agreement with those (Hussain *et al*., 2011). The changes in chemical composition of toast bread enriched with15% defatted flaxseed flour was decreased at the level of fat to 3.52 when compared with 10% full fat flaxseed which recorded 6.79 % fat. Moreover, the results indicated that, the toast bread enriched with flaxseed types retained moisture more efficiently than the control and did not differ significantly among the composite flour .These results in accordance with the results of (Pohjanheimo *et al.,* 2006 and Horshid *et al.*, 2010) who showed that, analysis of chemical composition indicated that the flaxseed bread treatment contained higher protein, fat, crude fiber and ash. Finally it could be noticed that significantly different were observed at $p < 0.05$ between all treatments and control due to the supplementation of full fat and defatted flaxseed flour in wheat flour.

Fatty acid contents

The fatty acids composition of incorporation full fat and defatted flaxseed flour on wheat toast bread. The results obtained are tabulated in Table (5) and Fig.1. It could be observed that flaxseed toast bread was found to have high content of unsaturated fatty acids. The lowest contents of palmitic acid (0.185%) and stearic acid (0.045%) were recorded in control bread and bread+15%DFF respectively. While the highest contents of palmitic and stearic acid were recorded (0.451%) and (0.126%) in toast bread +(10%FFF+15%DFF) respectively these results agreement with those of (Hussain *et al*., 2011) who reported that unleavened flat breads containing 12%

FFF possessed the highest content of palmitic (0.595%) and stearic acid (0.152%). The major fatty acids in all flaxseed toast bread were linolenic acid C18:3, oleic acid C18:1 and linoleic acid C18:2.The results showed that bakery products prepared from flaxseed supplemented composite flours is realized the highest contents of linoleic and linolenic acid which was found as the most plentiful polyunsaturated fatty acid in these products. Toast bread having (10%FFF+15%DFF) achieves the highest linoleic acid (1.167%) and linolenic acid (2.158%) among all bakery products. Higher amounts of flaxseed flour supplementation in bakery products beside their potential health benefits in the form of its functional components like α-Linolenic acid, lignin and antioxidants might affect organoleptic properties (Aliani *et al*., 2011).

The results of present study (Fig. 1) are in line with the findings of (Gambus *et al*., 2004) who found that addition of 10-13% flaxseed in bread resulted in the about 800-1000 time enhancement of linolenic acid content of bread as compared to control bread. (Ziemlanski, 1997) also observed an increase in linolenic acid content of wheat bread and pastry supplemented with linseed meal.

The linolenic acid is the most important fatty acid which is polyunsaturatedand has major concern on health from nutritional point of view. The medical research has shown that the excessive level of linoleic acid relative to linolenic acid may increase the probabilities of a number of diseases (Hibbeln, 2006). On the other hand, lowest contents of these essential fatty acids are well evident in the toast bread prepared from 15% defatted flaxseed supplemented composite flour when compared to toast bread prepared from 10% full fat flaxseed these results agreement with those of Hussain *et al*. (2012).

Antioxidants in full fat and defatted flaxseeds

Natural antioxidants namely α-tochopherol, β-carotene and lignan as polyphenols were extracted from the studied full fat and defatted flaxseeds, and the results are presented in Table 6. These results indicate that full fat contained higst amount of α-tochopherol, β-carotene and lignan (8.70 mg/100g, 1686 IU/100g and 25.00 mg/g), respectively, which can be used for health promoting than defatted flaxseeds (5.33mg/100 g, 0764.84 IU/100 g and 12.28 mg/g).

These results are in line with (Johnsson *et al*. 2000) who reported SDG content in the range of 11.7 to 24.1 mg/g and 6.1 to 13.3 mg/g in defatted flaxseed flour and whole flaxseed, respectively. While, (Oomah *et al*., 1997; Velasco and Goffman, 2000) reported that the total tocopherol content in flaxseed is 9.3-16.9 mg/100 g seed. Other tocopherols are α-tocopherol, with a content of 0.0-9.1 mg/100 g oil (Choo *et al.,* 2007; Schwartz *et al*., 2008).

In –vitro protein digestibility

The digestibility is important criterions that determines the availability of physiologically active amino acids and peptides and is affected by processing treatments (Duodu *et al*., 2003). The result in Table (7) indicated that the highest digestibility value using trypsin was found in control toast bread (89.33) that for true digestibility and (76.90) for apparent digestibility and the lowest value in digestibility was toast bread+10%FFF (72.20) in true digestibility and (63.42) in apparent digestibility, significant difference

(P<0.05) was observed between all treatments. It was cleared from Table (7) that digestibility was high in control toast bread followed by toast bread+15%DFF (85.63), it could be related to the effect of drying treatment, since it is well known that heat treatment increase protein digestibility.

While, (Madhusudhan and Singh1985) reported 61% IPD for pepsin–pancreatin digestion of defatted flaxseed meal. This agreement with Bishnoi and Khetarpaul (1994) found improvement in protein digestibility (in vitro) by the common methods of domestic processing and cooking including soaking and ordinary cooking of legume grains. Baking and boiling increased the IPD of flaxseed at the end of both digestion phases (Wanasundara *et al*., 1999); therefore, intestinal phase enzyme activity was not hampered. Flax meals, after oil extraction contains 35.5% protein, 3.5% fat, 6% ash and 70% total digestible nutrients (Lay and Dybing, 1989).

Biological evaluation of incorporation of full fat and defatted flaxseed flour on the wheat toast bread

Nutritional parameters of normal and hypercholesterolemic rats fed on full fat and defatted flaxseed flour on composition toast bread are shown in Table (8). The results revealed that non-significant changes were found in final body weight and body weight gain, food intake and feed efficiency ratio of rats fed on FFF, DFF and mixture of them diets were estimated to follow up the healthy feed parameters during the experimental period, whereas previous measurements were significantly lower in hypercholesterolemic rats in comparison with the normal rats (p<0.05). Rat's weight positively increased during the treatment period in all groups except positive control group. The rats groups feed bread+(10%FFF+15%DFF) had the highest body weight gain (54.50 g). For food daily intake and food efficiency ratio, it was clearly that it increased in all treated groups compared with control positive group. Results showed significant (p≤0.05) different between two controls and rats groups fed on full fat and defatted flaxseed flour diets.

T.C, total cholesterol; T.G, triglycerides; LDL-C, low density lipoprotein cholesterol; HDL-C, high density lipoprotein cholesterol Analysis of blood lipid profiles. The comparison of blood lipid profiles of positive control group and treated groups with flaxseed revealed increase in total cholesterol (TC), triacylglycerol (TG) and low density lipoprotein cholesterol (LDL-C) and a mild decrease in high density lipoprotein cholesterol (HDL-C) but these changes were significant Table (9).

A significant reduction (p < 0.05) of total cholesterol was observed all flaxseed diet groups especially toast bread+(10%FFF+15%DFF) group having the highest reduction (98.06 Mg/dl) followed by diet groups(toast bread+10%FFF) these results are in line with (Abdel-Rahman *et al*., 2010) who investigated that, similarly a dietary intake of 10% flaxseed for a period of 30 days has indicated a substantial reduction in blood cholesterol level of rat models . (Yamashita *et al*., 2003) reported that, the flaxseed oil lowered plasma total cholesterol than defatted flaxseed and sesame seed.(Bierenbaum *et al*., 1993) found that flaxseed supplementation in the form of either flaxseed containing bread or 15 g of ground flaxseed resulted in significant reductions in serum total cholesterol in human subjects with hyperlipidemia. . The components of flaxseed to which health benefits have been ascribed as LDL lowering, include its high contents of lignans and vegetable protein, (Jenkins *et al*., 1999).

Table.1 Formula of toast bread prepared using full and defatted flaxseeds

Ingredient	Control (g)	Bread with full and defatted flaxseeds flour		
		Full flaxseeds flour (g)	Defatted flaxseeds flour (g)	Full and defatted flaxseeds flour (g)
Wheat flour (82%)	500.0	490.0	485.0	475.0
Sugar	25.0	25.0	25.0	25.0
linseed oil	25 .0	25.0	25.0	25.0
Skimmed milk	12.5	12.5	12.5	12.5
Improver	6.0	6.0	6.0	6.0
Salt	8.25	8.25	8.25	8.25
Yeast	6.0	6.0	6.0	6.0
Full and defatted flaxseeds flour	-	10.0	15.0	25.0
Water	as required	as required	as required	as required

Table.2 Diet composition

Ingredients	The basal diet (g)	Bread diet (g)
Corn starch	723	56.3
Dried bread	-	666.7
Casein	122	122
Corn oil	50	50
Cellulose	50	50
Mineral mixture	40	40
Vitamin mixture	10	10
DL-methionine	3	3
Choline chloride	2	2

Table.3 Effect of incorporation of full fat and defatted flaxseed flour on the sensory scores (9 point hedonic scale) of toast bread

Properties Toast bread	Taste	Crust color	Flavor	crumb color	Appearance	Over all Acceptability
Control toast bread	$8.69^{a}\pm0.67$	$8^{a}\pm0.72$	$8.2^{a}\pm0.6$	$8.1^{a}\pm0.77$	$8.55^{a}\pm0.57$	$8.4^{a}\pm0.88$
Toast bread +10%FFF	$8.25^{a}\pm0.52$	$7.2^{b}\pm0.55$	$7.55^{ab}\pm0.89$	$7.09^{b}\pm0.57$	$7.85^{ab}\pm0.41$	$7.8^{ab}\pm0.75$
Toast bread +15% DF	$7.81^{ab}\pm0.53$	$6.4^{c}\pm0.69$	$7.68^{ab}\pm0.72$	$6.15^{c}\pm0.52$	$7.45^{ab}\pm0.44$	$7.52^{ab}\pm0.77$
Toast bread +(10%FFF+ 15%DF)	$6.8^{c}\pm0.45$	$5.75^{cd}\pm0.39$	$.6.69^{c}\pm0.54$	$5.55^{cd}\pm0.44$	$6.83^{c}\pm0.38$	$6.02^{c}\pm0.68$

Each value is the mean ± SD

Mean values in each column having different subscript (a, b, c, d) are significantly different at $p < 0.05$.

Table.4 Gross chemical composition of incorporation of full fat and defatted flaxseed flour on the wheat toast bread (on dry weight bases g/100g)

	Constituents %						
Toast bread	Moisture	Crude Protein	Fat	Ash	Fibers	Carbohydrates	Kcal/100g
Control toast bread	10.17^{c} ± 0.82	10.16^{d} ±0.86	3.52^{c} ±0.39	1.68^{d} ±o.32	2.87^{d} ±0.35	81.76^{a} ±4.00	39.36^{b} ±56.00
Toast bread +10%FFF	11.06^{ab} 0.59±	12.18^{c} ±0.97	6.79^{a} ±0.90	2.46^{c} ±0.40	3.01^{c} ±0.47	75.56^{b} ± 6.00	412.07^{a} ± 65.00
Toast bread +15% DF	11.85^{a} ± 0.65	15.71^{b} ± 0.89	3.05^{c} ± 0.40	3.12^{b} ±0.52	3.55^{b} ±0.42	74.57^{b} ±5.00	388.57^{c} ± 44.00
Toast bread +(10%FFF+ 15%DF)	12.24^{a} ± 0.80	16.74^{a} ± 0.98	5.21^{b} ±.0.70	4.08^{a} ±0.79	4.3^{a} ±0.81	69.67^{c} ± 3.00	392.53^{bc} ± 48.00

Each value is the mean ± SD

Mean values in each column having different subscript (a, b, c, d…..) are significantly different at $p < 0.05$.

Table.5 Effect of incorporation of full fat and defatted flaxseed flour on the fatty acid composition of toast bread

Fatty acid/Formulation Toast bread	Palmittic acid (16:0) (%)	Stearic acid (18:0) (%)	Oleic acid (18:1) (%)	Linoleic acid (18:2) (%)	Linolenic acid (18:3) (%)
Control toast bread	0.185	0.081	0.225	0.133	0.152
Toast bread +10%FFF	0.416	0.116	1.189	0.980	2.083
Toast bread +15%DFF	0.212	0.055	0.622	0.507	0.987
Toast bread +10%FFF +15%DFF	0.451	0.126	1.2171	.1.167	2.158

Table.6 Antioxidants extracted from of full fat and defatted flaxseed

Antioxidants / **Flaxseed Type**	**α–tochopherol (mg/100g)**	**β-carotene (IU/100g)**	**Lignan (mg/g)**
Full fat flaxseed	8.70	1686.00	25.00
Defatted flaxseed	5.33	0764.84	12.28

Table.7 In–vitro enzymatic digestibility of incorporation of full fat and defatted flaxseed flour on the wheat toast bread

Treatments	Trypsin	
	True digestibility	Apparent digestibility
Control toast bread	89.33^{a} ±1.00	76.90^{a}±1.00
Toast bread+10%FFF	72.20^{d} ±1.00	63.42^{d}±0.01
Toast bread+15%DFF	85.63^{b} ±0.01	75.76^{b}±0.01
Toast bread+ (10%FFF+15%DF)	77.01^{c} ±1.04	65.21^{c}±0.01

Each value is the mean ± SD

Mean values in each column having different subscript (a, b, c, d) are significantly different at $p < 0.05$.

Table.8 Initial weight, final weight, Body weight gain, food intake and feed efficiency ratio of rats fed on full fat and defatted flaxseed flour on composition toast bread

Parameters / Rat groups	Initial weight (g)	Final weight (g)	Body Weight gain (BWG) (g)	Food intake (FI) gm	Feed efficiency Ratio (FER) %
G (1) Negative control	103.44^{c}±0.10	129.8^{g}±0.10	26.3^{e}±0.01	13.73^{c}±1.00	1.92^{b}±1.00
G(2) Positive control	102.50^{c}±1.00	83.38^{h}±0.10	19.17^{f}±1.00	10.10^{d}±1.00	1.90^{b}±1.00
G(3) Control toast bread	104.70^{b}±1.00	149.90^{d}±1.00	45.20^{d}±1.00	16.95^{a}±1.00	2.66^{a}±1.00
G(4) Toast bread +10%FFF	100.60^{d}±0.10	152.85^{b}±0.10	52.25^{b}±1.00	17.90^{a}±1.00	2.91^{a}±1.00
G(5)Toast bread +15%DFF	106.20^{a}±1.00	159.40^{a}±1.00	53.20^{a}±1.00	17.60^{a}±1.00	3.02^{a}±1.00
G(6)Toast bread +(10%FFF+15%DFF)	103.75^{c}±1.00	158.25^{a}±0.01	54.50^{a}±1.00	18.80^{a}±1.00	2.90^{a}±0.01

Each value is the mean ± SD

Mean values in each column having different subscript (a, b, c, d) are significantly different at $p < 0.05$

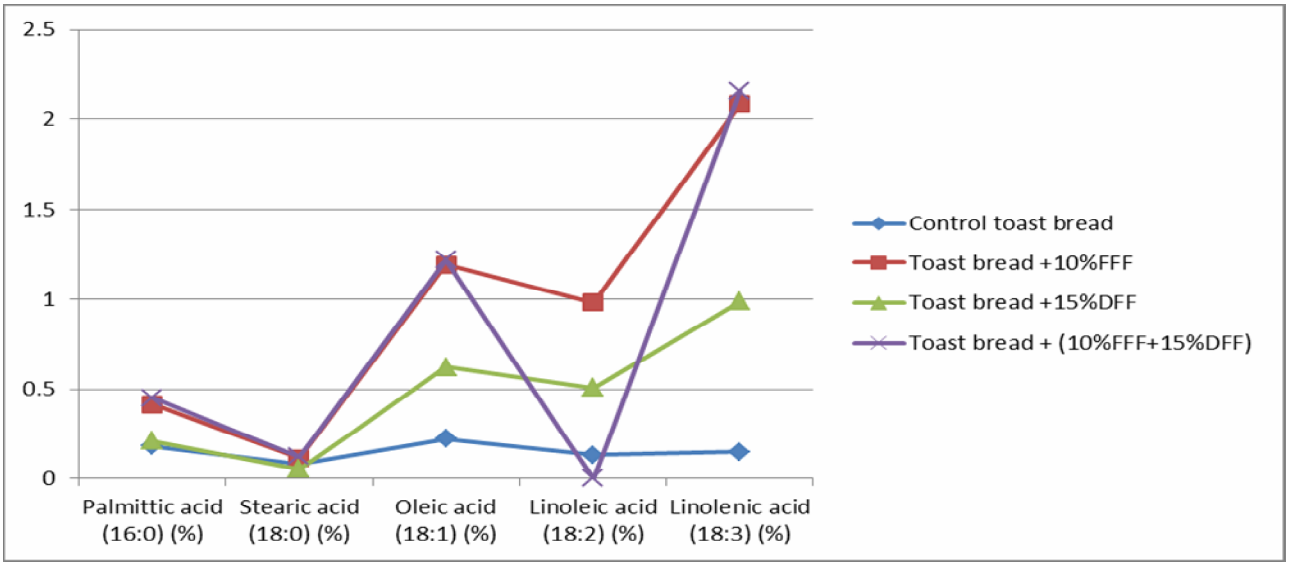
Table.9 Serum lipids profile of rats fed on full fat and defatted flaxseed flour on composition toast bread

Parameters / Rat groups	TC (Mg/dl)	T.G (Mg/dl)	HDL (Mg/dl)	VLDL (Mg/dl)	LDL (Mg/dl)	Atherogenic Index (AI)
G(1) Negative control	91.20^{c}±3.0	88.54^{f}±2.0	45.05^{d}±1.1	17.71^{d}±1.0	28.44^{c}±1.2	2.02^{b}± 1.0
G(2) Positive control	132.97^{a}±3.3	143.51^{a}±3.0	37.13^{f}±1.3	28.71^{a}±1.1	67.13^{a}±2.3	3.58^{a} ± 1.1
G(3) Control toast bread	115.11^{b}±2.0	130.12^{b}±1.0	47.92^{c}±2.0	26.02^{b}±1.2	41.17^{b}±2.0	2.40^{bc}± 1.0
G(4) Toast bread+10%FFF	105.01^{c}±2.5	119.05^{c}±2.0	51.80^{a}±2.2	23.81^{c}±1.0	29.40^{c}±1.2	2.03^{b}±1.0
G(5) Toast bread+15%DFF	106.04^{c}±1.0	117.15^{d}±1.0	50.10^{ab}±1.2	23.43^{c}±1.0	32.51^{d}±1.2	1.99^{b}±1.0
G(6) Toast bread+(10%FFF+15%DFF)	98.06^{d}±1.3	114.30^{e}±1.2	52.18^{a}±2.4	22.86^{c}±1.0	23.02^{e}±1.0	1.88^{b}±1.0

* Each value is the mean ± SD

* Mean values in each column having different subscript (a, b, c, d, e) are significantly different at $p < 0.05$

Fig.1 Finally the linolenic acid is the most important fatty acid which is polyunsaturated and has major concern on health from nutritional point of view



The effect of different treatments on serum triglycerides after feeding of hypocholesterolemic rats for six weeks was clearly. All treatments decreased serum triglycerides levels at different degrees (88.54 to 143.51 mg / dl) ,there was significantly different between all rats groups that may be due to using different levels of flaxseed, TG significantly decreases in all diets groups treated with flaxseed. Gaafar (2005) reported that hypercholesterolemic rats fed diet containing cakes fortified with hulled flaxseed showed a significant reduction in triglyceride level. The significant reduction of TC and LDL-C however; HDL-C was observed significantly increased ($p < 0.05$) in all treated groups. No significant difference was observed between any flaxseed groups on HDL-C concentration.

By increasing the dosage of flaxseed in the diet the blood improving effect of flaxseed became more significant. Significantly increased HDL-C concentration similar to the report by (Daleprane et al., 2010). On the other hand, the atherogenic index is an indicator for the susceptibility for atherosclerosis (Kawase *et al.*, 2000). The highest content in atherogenic index was found in positive control as (3.58) and the lowest content in atherogenic index was found in diet groups (toast bread+(10%FFF+15%DFF) as (1.88) .No significant difference was observed between any flaxseed groups .The cholesterol-lowering effect of flaxseed is likely the main contributing factor to its anti atherogenic potential; however, since atherosclerosis was only inhibited in animals fed a higher flaxseed dose, another mechanism, likely cellular, may be responsible for this anti atherogenic action(Chantal *et al*., 2007).

It can be concluded from the present studies that inclusion of full fat and partially defatted flaxseed flours in the toast breads imparted significant improvement in the protein, fat and fiber contents. It is also observed that addition of 10% full fat and partially15% defatted flaxseed flour does not negatively affect the sensory scores. Keeping in view the health potentials of flaxseed, fortified bread can be easily recommended to the public who are really conscious about their health and nutrition. Breads fortified with the full or defatted flaxseed showed significant improvements in digestibility and plasma lipid profile of hypercholesterolemic rats have related its effect to high fiber, ALA and lignin content of flaxseed and were acceptable from the sensory point of view. Therefore such bread can be recommended as functional food to treat hypercholesterolemia or reduce the risk of atherosclerosis.

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