

# Flaxseed Composition and Phytochemical Screening Methods

Aarti K. Giram<sup>1</sup>, Shivani O. Gud<sup>2</sup>, Aishwarya Sakhare<sup>3</sup>,  
Samadhan M. Gaikwad<sup>4</sup>

<sup>1,2,4</sup>Student, Amepurva Forums Nirant Institute of Pharmacy

<sup>3</sup>Professor, Amepurva Forums Nirant Institute of Pharmacy

## ABSTRACT:

Flaxseed, often known as linseed (*Linum usitatissimum* L.), is derived from the annual flax plant. The human nutrition industry is the primary application for flaxseed, as it is becoming recognized as a significant functional food component because to the active ingredient content, which is shown to have health advantages. Flaxseed can be consumed in a variety of forms, such as ground, as oil, or combined with baked goods. Flaxseed consumption is supported by scientific evidence due to its high content of omega-3 and omega-6 rich oil, lignans,  $\alpha$ -linolenic acid, high-quality proteins, and fibers. These compounds are biologically active and can prevent various chronic diseases like diabetes, cardiovascular disease, cerebrovascular stroke, and many types of cancer. Additionally, benefits from flaxseed diet have been demonstrated in the field of animal nutrition, and as a result in healthier food from animal origin. In fact, the fatty acid profile of the meat and fat is directly affected by the source of fat in diet in swine and poultry, feeding omega-3 enriched diets by the addition of flaxseed would increase the omega-3 content in eggs and meat and thus enrich the products. The present review is focused on recent data on flaxseed chemical composition and its phytochemical screening methods result in healthier food from animal origin.

**Keywords:** Flaxseeds, chemical composition, screening method, identification, structure.

## INTRODUCTION:

Flaxseed are used in food industry, mainly they are used in bakery products and in feed due to the presence of nutraceutical components including  $\alpha$ -linolenic acid (ALA), lignans and fiber [6]. Flax can be classified into two groups-fibered flax (rich in fiber) and oily flax (rich in oil). The stem of flax is used to produce the fiber of linen and the flaxseed are used for oil and feed production [7]. Flax seed contain 35-45% oil, which contains 9-10% saturated fatty acids (pamitic and stearic acid), about 20% mono unsaturated fatty acid (mainly oleic acid) and 70% poly unsaturated fatty acid (linoleic acid and linolenic acid). Protein content of flaxseed varies from 20-30%. Protein of flaxseed is limited by lysine content but their biological value (77.4%) and digestibility (89.6%) is high [8]. It also contain 30% dietary fiber, 4% ash and 6% moisture [9.10]. There has been a growing interest for the probiotic properties of flaxseed and its beneficial effects in coronary heart disease, some kinds of cancer, neurological and hormonal disorders [11]. Mainly flaxseed are produced in Canada, Argentina, U. S. A, Chaina, India and Europe [12]. Flaxseed oil is rich  $\alpha$ -linolenic acid (55%), and omega 3 fatty acids, which is higher than any other

vegetable oils [13]. A-linolenic acid can be metabolized to form eicosa pentanoic acid and docosa hexanoic acid in human intestine, which helps in the reduction of lifestyle diseases [14]. Previous studies also have shown that flaxseed oil helps in the reduction of many diseases like hyper lipidaemia, atherosclerosis [15], mammary cancer [16] and cardiovascular disease [17]. Flaxseed oil is also used in the production of linoleum, paints, ink, cosmetics, coating, vernishes [18,19]. The medicinal importance of a plant is due to the presence of some special substances like alkaloids, glycosides, resins, volatile oil, gum and tannins etc. The active principles usually remain concentrated in the storage organs of the plants [20]. Considering all these facts the present study is designed to investigate the presence of various phytochemicals in the flaxseed, a plant which evokes various therapeutic effects.

Consumer's interest in healthy eating, in the last decades, shifted towards the potential health benefits of specific foods and food ingredients. Foods, in fact, are not intended to only satisfy hunger and to provide basic nutritional requirements but also to prevent nutrition-related diseases and to improve physical and mental well-being of the consumers. Consumer demand for foods with greater beneficial effects, led food industries in increasing the production of functional foods that now represents a significant share of new food products. The result was a growing demand for marketing authorizations and the regulatory authorities had to face the problem of the evaluation of "claims" proposed by companies, in the absence of clear and global rules. Therefore, the need of a European Union law, which regulates the marketing of functional foods, has led to the definition of Regulation CE 1924/2006, with the aim of protecting consumers, promoting fair trade promotion and encourages product innovation in food industry [2]. In the relationship between diet, health and well-being, functional foods play an outstanding role. Many definitions exist worldwide for functional foods, but there is no official or commonly accepted definition [3]. The European Commission's Concerted Action on Functional Food Science in Europe (FuFoSE), defined that a food product can only be considered functional if together with the basic nutritional impact it has beneficial effects on one or more functions of the human organism thus either improving the general and physical conditions or/and decreasing the risk of the evolution of diseases [4].

### **Structure And Chemical Composition :**

An embryo with two cotyledons that is enclosed by a thin endosperm and a smooth, frequently lustrous yellow to dark brown seed coat makes up mature flaxseed, which is oblong and flattened (hull) [21]. Flaxseed sprouts are edible, with a slightly spicy flavor. Whole flax seeds are chemically stable, but ground flaxseed can go rancid at room temperature in as little as 1 week, although there is contrary evidence. A longer amount of time will be prevented from ground flax going rancid by refrigeration and storing in airtight containers. When packed quickly without exposure to air or light, milled flax is impressively resilient to oxidation for 9 months at room temperature and for 20 months at ambient temperatures under warehouse conditions [22,23].

### **Composition of Flaxseed:**

The components of this herb are summarized in Table 1. Also, the content of vitamins, minerals, and active compounds in flaxseed is summarized in Table 2. The protein content in flaxseed has been reported as 10–30% in some studies [24,25,26]. In terms of protein distribution, the highest amount of protein is in the cotyledons of the flaxseed (between 50 and 70%), and about 30% is in the coat and endosperm [27]. The amino acid profile of flaxseed protein is comparable with that of soy. As shown in Table 1, higher amounts of the some amino specially arginine, valine, glycine, leucine, valine, and serine have been found in flaxseed [28]. In terms of essential amino acid index of flaxseed, it has been reported that the score for this index for flaxseed is 69, and this score is near the score for canola and soy, that is, 75 and 79,

respectively [29]. Also, according to the Food and Agriculture Organization (FAO) reports, flaxseed score based on the limiting amino acid is 82; however, this score for soybean is 67 [30]. It has also been shown in some studies that 34.3% of the amino acids in flaxseed are essential amino acids, and the ratio of lysine to arginine, which is an indicator of cholesterolemic and atherogenic effects of a protein, is about 0.22–0.37, which reveals that flaxseed is less atherogenic than canola and soybean [31].

Components (%)	Whole Flaxseed	Component	g/100g of Flaxseeds	Component	g/100g of Flaxseeds
Moisture	7.13	A-linolenic acid	22.8	Arginine	9.2
Nitrogen	4.01	Linoleic acid	5.9	Glycine	5.8
Fat	41	Oleic acid	7.3	Leucine	5.8
Protein	20	Searic acid	1.3	Valine	4.6
Total dietary fiber	28	Palmitic acid	1.2	Serine	4.8
Ash	3.4	Glutamic acid	19.6	Soluble fibres	4.3-8.6
		Aspartic acid	9.3	Insoluble fibres	12.8-17.1

**Table 1: The most important flaxseed components[32]**

Components	g/100g of Flaxseeds	Components	g/100g of Flaxseeds	Phenolic Compounds mg/100g of flaxseed	
Calcium	236	Δ-tocopherol	10	Secoisolariciresinol	165
Magnesium	431	Vitamin C	0.5	Laricinesol	1.7
Phosphorus	622	Vitamin B1	0.5	Pinoresinol	0.8
Potassium	831	Vitamin B2	0.2	Total Flavonoids	35-70
Sodium	27	Vitamin B3	1.2		
Zinc	4	Vitamin B6	0.6		
Iron	5	Pantothenic acid	0.6		
Manganese	3	<b>Phenolic Compounds mg/g flaxseed powder</b>			
γ-tocopherol	522	Ferulic acid	10.9		
α-tocopherol	7	Chlorogenic acid	7.5		
		Gallic acid	2.8		

**Table 2: The content of vitamins, minerals, and active compounds in flaxseed[32]**

The Canadian Grain Commission examined brown Canadian flaxseed, finding that it typically included 41% fat, 20% protein, 28% total dietary fiber, 7.7% moisture, and 3.4% ash [33]. As reported, flaxseeds are rich in oil (40–46% by seed weight) with a healthy fatty acid profile. In terms of type of fatty acids, flaxseed contains about 73% polyunsaturated fatty acids (PUFAs) and 10% saturated fatty acids. Also, in terms of the type of PUFAs, the evaluation results on different samples of flaxseed show the presence of about 52–57% α-linolenic acid (ALA) and an ω-3/ω-6 ratio of 1:0.3 [35]. Considering that higher amounts

of ALA make the oil prone to oxidation and Linola seeds have about 5% less ALA compared with the brown flaxseed, therefore, this type of flaxseed is more resistant to oxidation than brown flaxseed and for applications such as frying and shortenings are more suitable [34].

As shown in Table 2, in terms of minerals and vitamins content, flaxseed is one of the appropriate sources of calcium, phosphorus, potassium, magnesium, and  $\gamma$ -tocopherol [33]. In addition, flaxseed is a good source of some active compounds such as phenolic compounds. It has been reported that some of the bioactive phenolic compounds are ferulic acid, chlorogenic acid, and gallic acid [36]. Cyanogenic glycosides (CG), phytates, lignans, and antipyridoxin factors are some of flaxseed's minor constituents that have been identified. Flaxseed has amounts of the lignan secoisolariciresinoldiglucoside (SDG) that are 75–800 times higher than those of any other documented plants or vegetables [35]. Flaxseed is one of the best dietary functional foods in terms of supply lignan, and the amount of lignan in it varies between 0.9 and 3% depending on various factors [34]. Based on the evidence in some experimental studies, the lignans in flaxseed have anti-estrogenic effects and may have a preventive role in the pathological process of some hormone-dependent cancers [37]. The most predominant lignan in flaxseed is SDG. Enterolactone and enterodiol, which have antiestrogenic properties and structural similarities to estrogen and can bind to cell receptors to inhibit cell proliferation, are formed from SDG [37]. Numerous items made from flaxseed are being sold in the market and promoted for their benefits as functional foods and nutraceuticals. Whole seeds, ground or milled flaxseed (flour), oil extracted from flaxseed (by pressing; cold-pressed or not), flax meal, the coat of the seed, a portion of the seed that has been removed, flaxseed hull, cyclic peptides (orbitides) from flaxseed oil, and lignan extracts from the flaxseed hull are among them [38]. Some of the experimental studies evaluated the digestibility of proteins in the flaxseed, and in some studies, the digestibility percent of this plant has been reported as 81.4–85.8% [39,40], and this is near the soybean digestibility percent (digestibility percent = 84–85%) [41]. Also, the flaxseed biological value (BV) ranges from 66.4% to 77.4% [42]. The matrix elements, particularly mucilage and oil, and the earlier preparation of the seeds have an impact on the flaxseed protein digestibility [43]. It has been reported in some experimental studies that elimination of flaxseed mucilage improved protein digestibility to 50% compared with the 12.6% for full-fat and mucilage containing ground flaxseed [44]. Also, it has been reported that some of the other processing methods such as boiling, heat treatment can improve flaxseed protein digestibility [45]. One of the concerns related to flaxseed is allergy to the its proteins. However, to date, the results of several studies have reported rare cases of allergy to the proteins contained in flaxseed. Some researchers believe that this allergy observed in some cases is not due to flaxseed proteins, but the reason is the contamination in some seeds [46].

## **PHYTOCHEMICAL SCREENING METHODS:**

### **Extraction Method :**

#### **Oil Extraction:**

To obtain oil by solvent extraction method flaxseed powder was extracted with polar solvent (iso propanol) and non polar solvent (hexane) using Soxhlet apparatus (Borocil) for 5 hours at 70 degree Celsius temperature and the remaining solvent was removed by rotary evaporator (BUCHI)[47].

#### **Phytochemical screening:**

Flaxseed extract was tested for the presence of active phytochemicals such as Triterpenoids, Steroids, Glycosides, Saponins, Alkaloids, Flavonoids, Tannins, Proteins, Free amino acids, Carbohydrates, Phenolic compounds and Vitamin C. Following standard procedures were used[47].

**Test For Steroids And Triterpenoids:**

**Liebermann Burchard Test** – The sample extract was mixed with few drops of acetic anhydride, after that the mixture was boiled and cooled. Then concentrated sulphuric acid was added to the test tube and the formation of a brown ring at the junction of two layers was observed. Green coloration of the upper layer and the formation of deep red color in the lower layer confirm the presence of steroids and triterpenoids respectively[47].

**Test For Glycosides :**

**Keller Killiani Test** – The sample was treated with few drops of glacial acetic acid and Ferric chloride solution. Then concentrated sulphuric acid was added to the mixture, and two layers formation was observed. Lower reddish brown layer and upper acetic acid layer which turns bluish green indicated the presence of glycosides[47].

**Bromine Water Test** – The sample was dissolved in bromine water and the formation of yellow precipitate confirmed presence of glycosides [47].

**Test For Saponins :**

**Foam Test** – Sample was mixed with water and shaken and the formation of froth, which is stable for 15 minutes, confirmed the presence of saponins.

**Test For Alkaloids:**

**Hager's Test** – Few drops of Hager's reagent (saturated picric acid solution) was added to the sample. Formation of yellow precipitate indicated the presence of alkaloids.

**Dragendorff's Test** - By adding 1 mL of Dragendorff's reagent to 2 mL of extract, an orange red precipitate was formed, indicating the presence of alkaloids.

**Mayer's Test** - Few drops of Mayer's reagent were added to 1 mL of extract. A yellowish or white precipitate was formed, indicating the presence of alkaloids[48].

**Tests For Flavonoids :**

**Alkaline Reagent Test** - Two to three drops of sodium hydroxide were added to 2 mL of extract. Initially, a deep yellow colour appeared but it gradually became colourless by adding few drops of dilute HCL, indicating that flavonoids were present.

**Shinod's Test** - Ten drops of dilute HCL and a piece of magnesium were added to 1 mL of extract, the resulting deep pink colour indicating the presence of flavonoids[48].

**Test For Tannins:**

**Gelatin Test** – Sample solution was treated with gelatin solution, appearance of white precipitate indicated the presence of tannins.

**Lead Tetra Acetic Acid Test** - One milliliter of lead tetra acetate solution was treated with 0.5 mL of extract, precipitate formation indicating the presence of phenolic compounds and tannins[48].

**Test For Proteins :**

**Biuret Test** - Two drops of 3% copper sulphate and few drops of 10% sodium hydroxide were added to 1 mL of extract, violet or red colour formation indicating that proteins are present.

**Ninhydrin Test** - Two drops of 0.2% freshly prepared ninhydrin solution added to 1 mL of extract. Production of purple colour shows the presence of proteins[48].

**Test For Carbohydrates :**

**Molish Test** - Few drops of alcoholic a-naphthol solution were added to 2 mL of extract. Later, few drops of concentrated H<sub>2</sub>SO<sub>4</sub> were added along the walls of test tube. At the junction of two liquids, a violet colour ring appeared, indicating that carbohydrates were present.

**Benedict's Test** - To 5 mL of Benedict's reagent, 8-10 drops extract were added, then heated for five minutes; the resulting dark red precipitate indicated the presence of carbohydrates.

**Fehling's Test** - To 2 mL of extract, an equal volume of Fehling's (A & B) solution was added and heated for five minutes, the resulting red/dark red precipitate indicating the presence of carbohydrates[48].

**Test For Starch :**

**Iodine Test** - Two milliliters of iodine solution with potassium iodine were added to 2 mL of test extract, and the appearance of a blue colour indicated that presence of starch.

## RESULT AND DISCUSSION:

Many polar and non polar solvents are used to extract oil from flaxseed. Here iso propanol was used as polar solvent and hexane was used as non polar solvent. 35% flaxseed oil was extracted by polar solvent (iso propanol) and 40% flaxseed oil was extracted by non polar solvent (hexane). Table 1 shows the oil yield percentage by polar and non polar solvent extraction method.

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