

NEW RECIPES FOR FOOD BY ADDING FUNCTIONAL INGREDIENTS COMING FROM FOOD SUPPLEMENT PROCESSING

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Abstract

The main aim of this project was to develop new recipes for bakery products, biscuits and cookies by adding functional ingredients coming from food supplement processing such as flax seeds or alfalfa after partial oil extraction.

The project will have a positive impact for both participant companies through designing and marketing of new innovative products for the coordinator Spanish company, and better valorification of plant resources and by-products resulting from food supplement processing for the second one, Romanian company.

Key words: *functional ingredients, by-products, nutritional value, partially defatted flaxseed*

1. INTRODUCTION

Flax is an annual medicinal and industrial plant (approx. 200 varieties) and has been used since ancient times. Flax is native from the Mediterranean regions, Crimea, the Caucasus and Asia minor and today can be found in cultures in Central Europe, Belgium, Netherlands, in Eastern Russia, Egypt, Algeria, East India and North America

From flax – medicinal plant, can be used exclusively to plant seeds, which have a valuable content of bioactive compounds. In phytotherapeutic and dietary goals, the **flax seeds** (*Semen Lini*) are used as such or like **flax oil** (*Oleum Lini*), **flaxseed flour** (*Farina Lini*) or in various mixes along with other traditional herbs.

Flaxseed contains carbohydrates, cellulose, protein, potassium and magnesium salts, oils, E, F vitamins, mucilages, galacturonic acid, rhamnose, galactose, xylose, arabinose, lipids (consisting on polyunsaturated and mono-unsaturated fatty acids - oleic acid) and phenolic compounds (Łukasz Seczyk et al., 2016, Herchi, Arráez-Román et al., 2014; Herchi, Hujaili et al., 2014).

Unsaturated fatty acids constitute about 91% of the total fatty acids – with α -linolenic acid (57% of total fatty acids) and linoleic acid (16% of total fatty acids) predominating (Marpalle et al., 2014).

Although this high linolenic acid content is extremely healthy from the point of view of human nutrition, its instability limits the use of flax seeds in the manufacture of margarine or as edible oil (Zoltán Izsáki and Ildikó Iványi, 2004).

The protein content of linseed is 22-28%, while that of the seedcake remaining after oil extraction is 35-50%. Flax seeds contain essential amino acids, especially lysine.

The amino acids composition of the protein is determined principally by genetic traits, though the protein content and its amino acid composition may also be substantially modified by nutrient supplies used in soil and by the year in various crops.

Many studies have highlighted the numerous health benefits of flaxseed oil for the cardiovascular and skeletal systems (Grielet al., 2007) and for inflammatory conditions such as rheumatoid arthritis, psoriasis and ulcerative colitis (Mantzioris, James, Gibson, & Cleland, 1994).

So, it can reducing the risk of myocardial infarction, in type 2 diabetes, provides significant improvement in attention deficit & hyperactivity disorder in children (Stark, Crawford, & Reifen,

2008 and references therein), lower blood pressure (Paschos, Magkos, Panagiotakos, Volteas, & Zampelas, 2007).

Furthermore, flax seed have great functional properties and therefore many applications in the pharmaceutical sector: for example, benefits such as reduction in the risk of occurrence of breast cancer (Lowcock, Cotterchio, & Boucher, 2013), osteoporosis, diabetes (Wahba & Al-Zahrany, 2013), heart disease and menopausal symptoms (Thompson, 2003).

The objective of the present study was to evaluate the nutritional composition of by-product obtained during manufacture of the flaxseed oil, with the future prospect of using this by-product in the food industry.

2. MATERIALS AND METHODS

2.1. Materials

The partially defatted flaxseed, by-product from getting oil obtained by cold pressing, was kindly supplied by *Hofigal Export – Import SA* (Bucharest, Romania).



Fig 1. Partially defatted flaxseeds flour

2.2. Chemical analysis

- Moisture content was determined at 103 °C (± 2 °C) (2 g test samples) until constant weight was attained (ICC Standard No. 110/1).
- The ash content was determined by incineration at 525 ± 25 °C (ICC No. 104/1).
- Total fat content was determined by extracting 10 g of sample with petroleum ether at 40-65 °C, using a semi-automatic Soxhlet Foss Extraction System 2055 (Foss, Sweden).
- Total nitrogen (N) and crude protein content ($N \cdot 6.25$, conversion factor) was determined by the Macro Kjeldahl Method (Kjeltec System, FOSS, Sweden).
- The carbohydrate content was calculated by difference: $100 - (\text{ash content} + \text{protein content} + \text{fat content} + \text{moisture content})$. All experiments were performed in triplicate.

2.3. Fatty acids content analysis

Using H-NMR spectral technique was determined the fatty acids composition, especially the concentrations of short-chain saturated fatty acids (C_4 - C_8), di-unsaturated fatty acids, mono-unsaturated fatty acids and long-chain saturated fatty acids ($>C_8$). H-NMR spectra were recorded on a Bruker Ascend 400 MHz spectrometer, operating at 9.4 Tesla corresponding to the resonance frequency of 400.13 MHz for the ^1H nucleus. Samples were analyzed in 5 mm NMR tubes (Wilmad 507). The NMR samples were prepared by dissolving 0.2 mL oil in 0.8 mL $CDCl_3$. The chemical shifts are reported in ppm, using the TMS as internal standard.

2.4. Crude fiber content analysis

Crude fibers include cellulose, hemicellulose, and lignin. The crude fiber content of the samples was determined using a Fibretherm-Gerhardt apparatus.

The method for determination of crude fibers begins with treating the sample with an acid detergent solution (20 g *N*-cetyl-*N,N,N*-trimethylammonium bromide dissolved in 1 L sulfuric acid 0,5 M). In this solution, cellulose and lignin from the analyzed material are insoluble, unlike all other components. Using special FibreBags, the dilution and filtration steps are simplified. The most important aspects of this method of analysis of fibers are adherence to strict boiling times and to weighing procedures.

After treatment with the acid detergent solution, the insoluble residue is dried, weighed and then calcinated. The acid detergent fiber (ADF) content represents the insoluble part of the sample that is left after boiling in acid detergent solution from which the ash obtained upon calcination is subtracted:

$$\%ADF = \frac{((\chi - \alpha) - (\delta - \xi)) * 100}{\beta}$$

$$Blank\ value\ (\xi) = \gamma - \psi$$

where:

α = mass of FibreBag (g)

β = sample mass (g)

χ = mass of crucible and dried FibreBag, after digestion (g)

δ = mass of crucible and ash (g)

ξ = blank value of empty FibreBag (g)

γ = mass of crucible and ash of the empty FibreBag (g)

Ψ = mass of crucible (g)



Fig 2. Equipment for crude fiber determination

2.5. Mineral content analysis

Mineral content was determined using an atomic absorption spectrophotometer (ContrAA 700; Analytikjena). Total ash was determined by incineration at 550°C, in an oven. Analysis was performed using an external standard (Merck, multi element standard solution) and calibration curves for all minerals were obtained using 6 different concentrations. Dried samples were digested in concentrated HCl.

2.6. Amino acid content analysis

For the analysis of amino acid content, samples were hydrolyzed at 100–120 °C in 6N hydrochloric acid for 22–24 hours under vacuum. After evaporation to dryness of hydrochloric acid, the dry residue was diluted using 4 mM stock solution of Norleucine.

The chemical score of alfalfa concentrate flour was calculated according to FAO/WHO (1985) as follows:

$$\text{Chemical Score} = \frac{\text{mg/g of essential amino acid in test protein}}{\text{mg/g of essential amino acid reference protein}} \times 100$$



Fig 3. Equipment for amino-acid content determination

2.7. Statistical analysis

All analyses were performed in triplicate and the mean values with the standard deviations were reported. Microsoft Excel 2003 Program was employed for statistical analysis of the data with the level of significance set at 95%. Analysis of variance (ANOVA) followed by Tukey's test was used to assess statistical differences between samples. Differences were considered significant for a value of $p < 0.05$.

3. RESULTS AND DISCUSSION

The chemical composition of alfalfa concentrate flour is shown in table 1 and shows that this by-product is a rich source of protein and crude fiber, as their contents are 34.24% and 21.38%, respectively.

Table 1. Chemical composition of partially defatted flaxseeds flour (g/100 g, based on dry weight)

Constituents	Partially defatted flaxseeds flour *
Total protein (N x 6.25)	32.4 ± 0.28
Ash	5.93 ± 0.12
Crude fibers	12.75 ± 0.32
Total lipids	15.59 ± 0.12
Total carbohydrates	46.08 ± 0.05

* Results given as: M ± SD (mean ± standard deviation) of triplicate trials.

The partially defatted flaxseed flour can be a rich vegetal source of carbohydrates with potential probiotic function, useful for new recipes of functional food, dietary supplements and nutraceuticals.

Table 2. Fatty acids content of partially defatted flax seeds flour (g/100 g)

Constitutes	Partially defatted flaxseeds flour
Short-chain saturated fatty acids	13,80
Mono-unsaturated fatty acids	22,05
Di-unsaturated fatty acids	13,54
Poli-unsaturated fatty acids	50,61

In fig. 4 is presented the fatty acids profile of partially defatted flaxseeds.

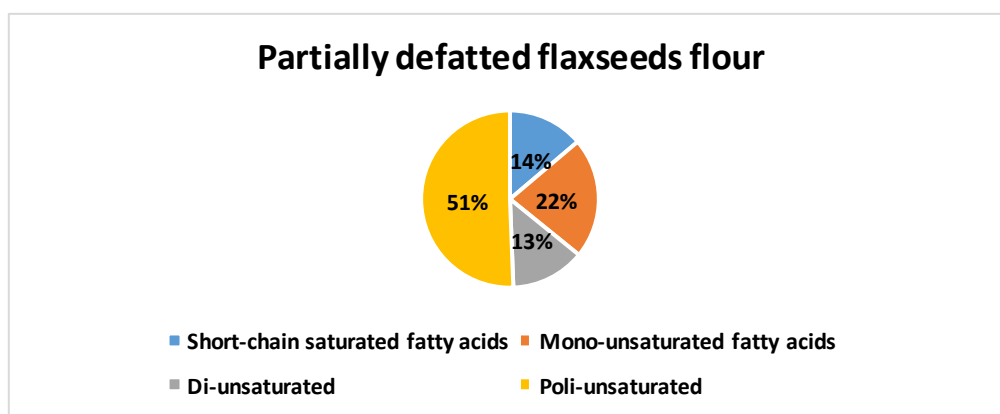


Fig. 4. Fatty acids profile of partially defatted flaxseeds

The study of the food potential of partially defatted flax seeds are not yet available in the scientific literature. Taking into account that consumers are more and more aware about the food quality, especially from the nutritive point of view, the new food resources rich in bioactive compounds are necessary to be found. In this respect, flax seeds meet the expectations of such consumers.

In the present study, the contents of six biologically essential mineral elements were analyzed: potassium (K), calcium (Ca), magnesium (Mg), iron (Fe), sodium (Na) and manganese (Mg), as well as an additional one essential trace element: zinc (Zn).

Table 3. Mineral contents of partially defatted flaxseeds flour (mg /100 g)

Constituents	Partially defatted flax seeds flour (mg/100g)	RDI (FDA 2011)
Potassium	600 ± 1.85	4700
Calcium	80 ± 0.29	1000
Magnesium	200 ± 0,90	400
Iron	95 ± 22.15	18
Sodium	90 ± 1.8	2400
Manganese	3.7 ± 1.91	400
Zinc	5 ± 1.95	15

From the performed analyses, it can be observed that the partially defatted flaxseed flour represents a material with important mineral contents, 100 g of this material providing the daily intake for some of these elements according to The Reference Daily Intake (RDI) of macronutrients and micronutrients recommended by the FDA (2011). Specifically, 100 g of this by-product contain more than the necessary daily intake of iron and significant amounts of magnesium, potassium, iron, manganese and zinc.

The amino acid composition of partially defatted flaxseed flour is given in table 4. The results show that partially defatted flaxseed flour contains high amounts of all essential amino acids, particularly leucine (7.1%), valine (5.96%), and phenylalanine (5.63%). However, partially defatted flaxseed flour protein is a fairly low source of Lysine (2.22%).

Essential amino acids are very important for human health as they cannot be biosynthesized, but serve as building blocks of certain proteins with major roles in the human body (Zho 2007).

Table 4. Amino acids composition of partially defatted flaxseed flour (as g amino acid/100 g protein).

Type	Amino acids	Partially defatted flaxseed flour
Indispensable amino acids	Methionine	2.18
	Leucine	7.1
	Valine	5.96
	Lysine	2.22
	Phenylalanine	5.63
	Isoleucine	4.64
	Threonine	4.12
	Histidine	1.99
Dispensable amino acids	Glutamic ac.	22.2
	Glycine	6.91
	Serine	3.27
	Proline	4.31
	Aspartic ac.	9.47
	Alanine	5.30
	Tyrosine	2.65
	Arginine	10.03
	Cysteine	1.80

The method is based on record each amino acid from the analyzed protein and report it to a standard protein (Whitaker & Tannenbaum, 1977).

Figure 5 shows the amino acid contents of partially defatted flaxseed flour in comparison with those of the standard protein set by FAO/WHO/UNU (1985).

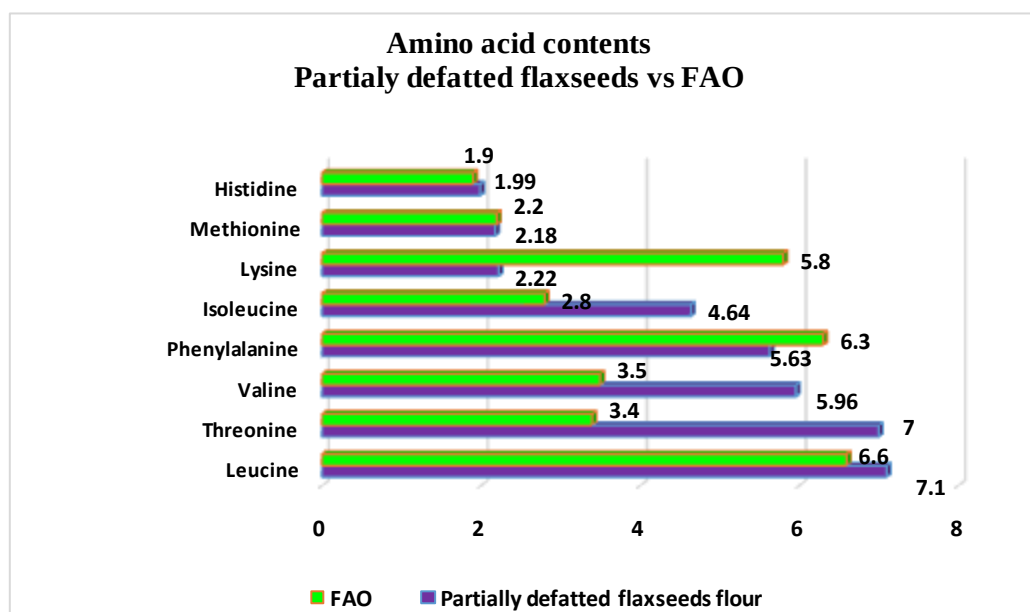


Fig 5. The percentages of amino acids in the partially defatted flaxseed flour protein in comparison with the percentages of amino acids in the standard protein.

Table 5 presents the chemical scores of selected amino acids from the partially defatted flaxseed flour calculated as defined in section 2.6.

Table 5. Chemical scores of amino acids in the partially defatted flaxseed flour

	FAO/WHO/UNU	Partially defatted flaxseed flour	% Amino Acid Scores
Leucine	6.6	7.1	107.58
Threonine	3.4	4.12	121.18
Valine	3.5	5.96	170.29
Phenylalanine	6.3	5.63	89.37
Isoleucine	2.8	4.64	165.71
Lysine	5.8	2.22	38.28
Methionine	2.2	2.18	99.09
Histidine	1.9	1.99	104.74

Can be observed a direct relation of the obtained chemical results with the protein quality, from the biological point of view (Hegsted 1974).

Results recorded into table no. 5 indicates that the defatted flaxseed proteins are a rich vegetal source of valine (chemical score of 170.29), isoleucine (chemical score of 165.71) and threonine (chemical score of 121.18). Also, average similar chemical score was obtained for leucine and histidine (107.58, respectively 104.74). For lysineis was detected the lower level of the chemical score.

4. CONCLUSIONS

The aim of this study was to evaluate the functional potential of partially defatted flaxseed flour for use in preparation of foods rich in valuable biocompounds.

The chemical characterization performed in this study proved that the partially defatted flax seeds flour is a valuable ingredient, in terms of nutritional components (protein, unsaturated fatty acids, minerals, indispensable amino acids) as well as a "source of fiber", according the Nutritional Claims approved by EFSA.

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