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Development and quality evaluation of flaxseed fortified nutra laddu

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Abstract
The goal of the present investigation was to grow nutra laddu with the incorporation of flaxseed of various proportions. To standardize processing technology and evaluate nutra laddu viz., chemical and fatty acid profile, antioxidant function, tactile, textural and microbial nutra laddu characteristics. The formulations were made using Whole Flaxseed, amaranth, Jaggery in Nutra laddu Preparation. The formulations were made using Whole Flaxseed, amaranth, Jaggery in Nutra laddu Preparation. For C0, C1, C2, C3, C4 and C5 respectively, different formulations were rendered with variation in whole Flaxseed level from 0 to 50 per cent. Using a 9-point hedonic scale, prepared nutra laddu analyzed organoleptic properties in terms of color and appearance, flavor, texture, taste and overall acceptability. Results showed that nutra laddu prepared with a 30 per cent supplement of Whole Flaxseed (C3) obtained the highest score (i.e. 8.0) was superior to the rest of the samples. Nutra laddu prepared with flaxseed has been found to be a rich source of protein and fiber. Therefore, flaxseed with strong nutritional and medicinal value can be well used as a functional ingredient for preparation of nutra laddu.

Keywords: Flaxseed, jaggery, amaranth, honey, nutra laddu, functional food, fatty acid profile, antioxidant activity, textural and microbial, sensory evaluation

Introduction

Flaxseed (*Linum usitatissimum* L) belonging to family *Lineaceae*, popularly known as Alsi, Jawas, Aksebija in Indian languages, Flax seed is a blue flowering crop that produces small, flat seeds ranging in colour from golden yellow to reddish brown. The texture of flaxseed is crisp and chewy possessing a pleasant nutty taste. The spherical fruit capsules contain two seeds in each of five compartments. The seed is flat and oval with a pointed tip. It have smooth glossy surface. It varies in colour dark brown to yellow. Flaxseed is often used to describe flax when consumed by humans while linseed denotes when it is used specifically for industrial Applications. Almost all parts of linseed plant are utilized for various purposes. Seed contains oil which after refining is used for edible purpose (Gutte *et al.*, 2015) [6].

Flaxseed oil is an excellent source of the omega-3 fatty acid linolenic acid with typical levels of 55% in the oil (Oomah, 2001) [13]. Increasing demand for edible oil sources with significant percentages of omega-3 fatty acids is resulting in consumption of flaxseed as a functional food. Analysis of brown Canadian flaxseed conducted by the Canadian Grain Commission showed the average composition of commercial seed was 41per cent, fat 20 per cent, protein 28 per cent, total dietary fiber 7.7 per cent and 3.4 per cent ash (DeClercq, 2012) [4].

Flaxseeds have nutritional characteristics and are rich source of ω-3 fatty acid: α-linolenic acid (ALA), short chain polyunsaturated fatty acids (PUFA), soluble and insoluble fibers, phytoestrogen lignans (secoisolariciresinol diglycoside-SDG), proteins and an array of antioxidants. According to its physicochemical composition, flaxseed is a multicomponent system with bio-active plant substances such as oil, protein, dietary fibre, soluble polysaccharides, lignans, phenolic compounds, vitamins (A, C, F and E) and minerals (P, Mg, K, Na, Fe, Cu, Mn and Zn).

Flaxseed oil is low in saturated fatty acids (9 %), moderate in mono saturated fatty acids (18 %), and rich in polyunsaturated fatty acid (73 %) (Cunnane *et al.*, 1993) [2]. Of all lipids in flaxseed oil, a-linolenic acid is the major fatty acid ranging from 39.00 to 60.42 per cent followed by oleic, linoleic, palmitic and stearic acids, which provides an excellent ω-6:ω-3 fatty acid ratio of approximately 0.3:1 (Pellizzon *et al.*, 2007) [14].

Flaxseed has potential health benefits besides the nutrition, due to mainly 3 reasons: first, due to its high content of ω-3 α-isease, hypertension, atherosclerosis, diabetes, cancer, arthritis, and osteoporosis, autoimmune and neurological disorders.

Flaxseed oil is believed to bring mental and physical endurance by fighting fatigue and controlling aging process. Flaxseed has also been reported to act as anti-arrhythmic, anti-atherogenic and anti-inflammatory agent in addition to improving vascular function (Gogus and Smith, 2010) [5].

There are two groups of omega fats: omega-3 and omega-6 fatty acids. Linolenic acid, eicosapentaenoic acid (EPA) and docosahexanoic acid (DHA) are three types of omega-3 fatty acids and are nutritionally important. All three fatty acids have been shown to reduce the risk of cardiovascular disease 63. Flax contains a mixture of fatty acids. It is rich in polyunsaturated fatty acids, particularly ALA, the essential omega-3 fatty acid, and linoleic acid (LA), the essential omega-6 fatty acid (Gutte*et al.*, 2015) [6].

The whole flaxseed can be added to bread dough and other baked goods such as muffins, bagels, crackers and waffles. It can also be applied to a variety of multi-grain cereals as well as snack foods to add nutty taste, extra texture and nutritive value. When this seed is sprinkled on top of the baked goods before baking, they also add crunch, taste and eye appeal. Similarly, ground flaxseed can be added in several food products from breads to energy bars (Morris, 2003) [11].

The present study was conducted to incorporate flaxseed in different food products and study the effect of addition on proximate composition, fatty acid profile, sensory and microbial quality of flaxseed incorporated food products such as nutra laddu and nutra cookies. The present research is helps to explore the public knowledge and perceptions of the efficacy, safety and reason to consume flaxseed.

Materials and Methods

Materials

The Raw material like Flaxseed, Jaggery, amaranth, honey, etc. will be procured from the local market of Parbhani. Chemical and reagent will be obtained from laboratory, Department of Food Chemistry and Nutrition, College of Food Technology, VNMKV, Parbhani.

Proximate composition of nutra laddu

Raw materials such as flaxseed and amaranth and nutra laddu were analyzed for proximate composition including moisture, fat, protein, total carbohydrate, crude fiber, ash and mineral composition was carried out as per the methods given by AOAC, 2005**.**

Determination of minerals composition of nutra laddu

Two grams of defatted sample was weighed and heated at 550ºC. Then, the obtained ash were digested with concentrated Hydrochloric acid (HCL) on hot plate. The digested material was then filtered using whatman No. 42 filter paper and the final volume made to 100ml with distilled water that was further used for analysis with respects to iron, calcium, potassium, contents by using methods Ranganna (1986) [15].

Fatty acid composition of nutra laddu

Fatty acid composition of the raw sample was determined using Gas chromatography of FAMES (Fatty Acid Methyl Esters) with Flame Ionization Detector by AOCS Official Method Cd 14c-94 (2003). The oil (10–20 mg) was saponified for 1 hr with 1 ml of methanolic KOH (0.7 N) at 60 ^{0}C, followed by neutralization with 1 ml of methanolic HCl (0.7 N). The resulting free fatty acids were extracted in hexane and evaporated to dryness. The fatty acids were methylated using boron trifluoride (14% in methanol) and 0.2 ml benzene. The FAME was extracted in hexane, washed with water and evaporated to dryness. Fatty acid analysis was performed using a gas–liquid chromatograph (Shimadzu, GC-14B, Shimadzu Corporation, Japan) (Plate 6) fitted with a fused silica capillary column (BP 21: 30 m length, 0.30mm i.e., 0.50 µm film thickness). The GC was equipped with a flame ionization detector, Clarity Lite 420 integrator and at isothermal conditions. The column temperature was set at 220 ^{0}C, the injector temperature at 230 ^{0}C and the detector temperature at 240 ^{0}C. Nitrogen gas was used as the carrier gas with a flow rate of 1 ml/min. Individual fatty acids in the oil were identified by comparison with the retention times of standard fatty acid methyl esters.

Determination of antioxidant activity

Preparation of extract

The extracts were prepared in the usual way, 2 grams of flaxseed was used for antioxidant activity assay. The decoction was prepared by placing the flaxseed in 100 ml boiling distilled water for 3 minutes. The infusion was prepared by steeping the in freshly boiled distilled water for 30 minutes. The extracts were cooled, filtered and the volume was brought up to 100 mL using distilled water (Toda, 2011) [17].

Texture profile analysis (TPA)

Stable Micro System *TAXT2plus* Texture Analyzer was used for texture profile analysis (TPA) of energy bar. The test was configured so that the hardness calculated at the time of the test by determining the load and displacement at predetermined points on the TPA curve. Hardness (H) was the maximum load expressed in kg applied to the samples during the first compression. Textural determinations were made by using three point bendrig probes for bend test. The bars were bended to determine structural characteristics present inside or on the surface. Samples for bend test were placed centrally on heavy duty platform under three point bendrig probes. Both the load cell and probe were calibrated before test. Hardness measurement of bars by bending involved plotting force (g) versus time (sec). The maximum force (g) was used as an index of hardness for the bend test (Rehman *et al.*, 2012).

Microbial examination of product

The microbial examination of samples was carried out as per the method cited in Indian Standard Institute (ISI) (1969). The results obtained for each count was recorded as colony forming unit per ml of sample i.e. cfu/ml.

Table 1: Formulation of Nutra laddu with different levels of flaxseed

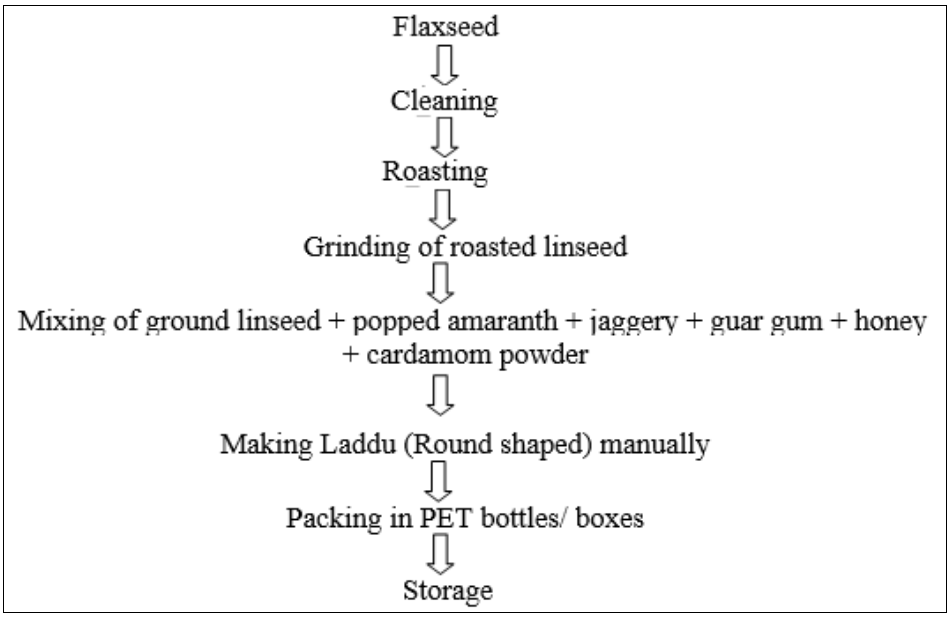
Ingredients	C_0	C_1	C_2	C_3	C_4	C_5
Linseed	00	10	20	30	40	50
Popped amaranth seed	50	40	30	20	10	00
Jaggery	30	30	30	30	30	30
Honey	20	20	20	20	20	20
Guar gum	0.2	0.2	0.2	0.2	0.2	0.2
Cardamom powder	0.1	0.1	0.1	0.1	0.1	0.1

Control = 50% Amaranth
C_1 =40% Amaranth + 10% Linseed
C_2 = 30% Amaranth + 20% Linseed
C_3 = 20% Amaranth + 30% linseed
C_4 = 10% Amaranth + 40% linseed
C_5 = 00% Amaranth + 50% linseed

Preparation of nutra laddu

Linseed was cleaned to remove impurities. The cleaned linseeds were then roasted slightly in a pan at low flame till it become crispy and dark brown in colour. The roasted linseeds were then grinded into powder using mixer grinder. Other ingredients such as jaggary, popped amaranth, guar gum, cardamom powder and honey were mixed uniformly with linseed powder. Finally, the round shaped laddu was made manually and packaged.

Flow sheet for preparation of nutra laddu



Results and Discussion

Proximate composition of ingredients of nutra laddu

The proximate composition of raw materials is an important aspect to be considered before its value addition in products. It will enhance the nutritional value of the prepared products. Hence, the proximate composition of jaggery, amaranth honey was calculated and stated in table 2.

Table 2: Proximate composition of raw materials of nutra laddu

Parameters (%)	Flaxseed	Jaggery	Amaranth seeds	Honey
Moisture	4.82 ± 0.27	4.2±0.03	2.28±0.06	25.36±0.40
Ash	3.08 ± 0.05	1.39±0.02	15.7±0.6	0.60±0.12
Protein	24.48 ± 2.15	0.51±0.03	8.24±0.4	1.01±0.03
Fat	31.82 ± 0.54	1.90±0.02	3.54±0.03	0.23±0.03
Carbohydrate	25.64± 0.01	91.50±0.3	66.29±0.55	67.57±0.40
Dietary fiber	8.97±0.19	0.50±0.03	1.81±0.2	5.23±0.03

*Each value represents the average of three determinations

The results presented in Table 2 demonstrates that data proximate composition of flaxseed. The composition of flaxseed can vary with genetics, growing environment, seed processing and method of analysis (Daun *et al*., 2003) [3]. From the results of proximate of flaxseed it was reported that moisture content 4.82 per cent whereas crude fat content was recorded 31.82 per cent. The average value of total carbohydrates and crude fibre of flaxseed was found to be 25.64 and 8.97 per cent respectively. The proximate composition particularly with respect to crude protein and ash content was reported to be 24.48 per cent and 3.08 per cent respectively. These results were in close agreements with the findings of Morris (2007) [12], Morris (2003) [11] and Kajla (2015) [9].

The protein was observed for jaggery (0.51%), amaranth seed (8.24%) and honey (1.01%) and fat content was observed for flaxseed (38.45%), jaggery (1.90%), amaranth seed (3.54%) and honey (0.23%).The amount of protein present in flaxseed indicates that the crop can form a part of human diet. The carbohydrate content in flaxseed was found to be for flaxseed (27.73%), jaggery (91.50%), amaranth seed (66.29%) and honey (67.57%). This showed that flaxseed is good source of energy. The dietary fiber content was found to contain for flaxseed (4.32%), jaggery (0.50%), amaranth seed (1.81%) and honey (5.23%). The obtained results were found comparable with the findings of Morris *et al*. (2007) [12].

Proximate composition of flaxseed incorporated nutra laddu

Data pertaining to various proximate composition of flaxseed nutra laddu were determined for moisture, fat, protein, carbohydrates, ash and crude fiber and results obtained are depicted in Table 3.

Table 3: Proximate composition of flaxseed incorporated nutra laddu

Sr .No.	Treatments	Proximate composition (g/100 g)					
		Moisture	Crude Protein	Fat	Ash	Crude fiber	Carbohydrate
1.	Control	3.05	9.02	23.60	1.10	1.21	60.02
2.	C_1	3.20	8.70	23.40	1.28	1.48	59.96
3.	C_2	3.24	8.37	23.00	1.37	1.65	59.37
4.	C_3	3.30	8.09	22.50	1.48	1.85	60.68
5.	C_4	3.10	8.66	23.50	1.21	1.38	61.15
6.	C_5	3.18	8.25	23.00	1.33	1.49	61.55

* Each value is average of three determinations

Physical properties of prepared flaxseed incorporated laddu were analyzed and obtained values are statistically analyzed and presented in Table 3. It is observed from table 18 that the highest moisture content was found to be in C_3 sample up to 3.30 per cent. Control sample was found to have the highest amount of crude protein and fat content. Ash and crude fiber content was found highest in sample C_3 as 1.48 per cent and 1.85 per cent respectively. Carbohydrate content was found to be highest in C_3 sample than other samples These results are closely related with the results recorded by Mohammed. (2015) [10]

Mineral content of flaxseed incorporated nutra laddu

Mineral composition analysis is important to assess the nutritional value of flaxseed nutra laddu. The mineral composition of flaxseed nutra laddu were investigated which includes calcium, phosphorus, sodium, potassium and iron. The results reported for these minerals are depicted in table 4.

Table 4: Mineral content of flaxseed incorporated nutra laddu

Minerals	Control Sample	Samlpe C_3
Potassium (mg/100g)	325.8	406.3
Calcium (mg/100g)	126.2	166.4
Phosphorous (mg/100g)	296.3	380.4
Magnesium (mg/100g)	210.8	237.4
Sodium (mg/100g)	11.20	14.45
Iron (mg/100g)	1.21	2.46

* Each value is average of three determinations

From data depicted in table 4 it is stated that phosphorus content sample C_3 was highest than control sample. The phosphorus content of sample C_3 and control sample was found as 380.4 and 296.3 However calcium content of sample C_3 and control sample gave reading of 166.4 and 126.2. The magnesium content of sample C_3 was seen as 237.4 and in control sample 210.8 was seen. The iron content in control sample and sample C_3 was found to be 2.46 and 1.21 respectively.

Antioxidant activity of flaxseed incorporated nutra laddu

The nutra laddu prepared by incorporating flaxseed were determined for antioxidant activity with respects to hydrogen peroxide and nitric oxide scavenging activity and results are depicted in table 5.

Table 5: Antioxidant activity of flaxseed incorporated nutra laddu

Samples	Antioxidant activity (%)	
	Hydrogen peroxide (H_2O_2)	Nitric oxide
Nutra Laddu	28.88	17.43

It is evident from data presented in above table that flaxseed nutra laddu exhibited strong scavenging activity with hydrogen peroxide and nitric oxide. Flaxseed nutra laddu showed 28.88 percent of hydrogen peroxides scavenging activity whereas, 17.43 percent nitric oxide scavenging activity.

Fatty acid profile of flaxseed incorporated nutra laddu

Fatty acid profile with respects to saturated fat, polyunsaturated fat and monounsaturated fats of essential oil extracted from flaxseed nutra laddu such as flaxseed nutra laddu were determined by using gas chromatography mass spectroscopy. Results obtained for fatty acid profile are illustrated in table 6

Table 6: Fatty acid profile of flaxseed incorporated nutra laddu

Fatty acid	Flaxseed nutra laddu Average value (mg/100g)
Saturated fatty acid (SFA)	9.2 ± 0.2
Mono unsaturated fatty acid (MUFA)	16.8 ± 0.3
Poly unsaturated fatty acid (PUFA)	61.4 ± 0.6
Alpha Linolenic acid	54.38 ± 0.5
Linoleic acid	7.28 ± 0.2
Oleic acid	19.87 ± 0.3
Stearic acid	5.84 ± 0.02
Palmitic acid	6.16 ± 0.02

*Each value is average of three determinations

Data presented in table 6 revealed that the saturated fatty acids in flaxseed nutra laddu were found to be 9.2g/100g respectively. Moreover, the polyunsaturated fats were analyzed which indicated that Flaxseed nutra laddu contained high amount of unsaturated fats (61.4 g/100g). Results shown for monounsaturated fats reported that flaxseed nutra laddu found to have high monounsaturated fats (16.8 g/100g) respectively.

Sensory Evaluation of nutra laddu

The fore treatment (C_1, C_2, C_3 C_4 and C_5) and control were made differently with different flaxseed, amaranth and jaggery to achieve a highly acceptable product. The organoleptic evaluation of nutra laddu carried out by a 10 semi trained panel member based on 9 point hedonic scale and the score were given by evaluating the sensory attributes for nutra laddu such color, appreance, texture, taste and overall acceptability which was compared with control sample and expressed table 7.

Table 7: Sensory characteristics of flaxseed incorporated nutra laddu

Sr. No.	Sample	Sensory attributes					
		Appearance	Colour	Taste	Flavor	Texture	Overall acceptability
1.	Control	7.9	8.0	8.1	8.1	8.0	8.0
2.	C_1	7.8	8.0	8.2	8.1	8.1	8.0
3.	C_2	8.0	7.8	7.9	7.8	8.0	7.9
4.	C_3	8.1	7.6	7.2	7.8	7.6	7.8
5.	C_4	7.4	7.1	7.0	6.6	7.0	7.0
6.	C_5	6.8	6.6	6.2	6.0	6.2	6.5

* Each value is average of three determinations

It is revealed from table 7 that among all treated samples, contol and C_1 sample were found to be good with score of 8.0 for overall acceptability than others samples. The samples C_2 and C_3 were found to be good in appearance, colour, taste, flavour and texture compared to C_4 and C_5 samples.

Microbial parameters of flaxseed incorporated nutra laddu

Prepared Flaxseed nutra laddu was selected on the basis of sensory quality and analyzed for microbial load at interval of 30 days up to 90 days stored at room temperature. The microbial study of sample was done for total plate count and yeast and mold count. The results obtained on microbial examination from present investigation are presented in the table 8.

Table 8: Microbial parameters of flaxseed incorporated nutra laddu

Treatments	Total Plate Count (cfu/g) x 10^1	Yeast and Mold Count (cfu/g) x 10^1
Control	1.10	0.92
C_1	1.69	1.49
C_2	1.71	1.58
C_3	1.61	1.42
C_4	1.64	1.60
C_5	1.37	1.48

* Each value is average of three determinations

It is evident from above table that TPC observed in Flaxseed nutra laddu sample was nil and yeast and mold count was found to be zero on the day of production and similar results were obtained up to 45 days of storage (at room temperature).
The data pertaining to the microbial analysis in terms of Total plate count (TPC) as well as Yeast and Mold count (YMC) of laddu incorporated with flaxseed at ambient temperature for the storage period of 48 hours is presented in table 8.
It is evident from Table 8 that Total Plate Count (TPC) and Yeast and Mold Count (YMC) of laddu incorporated with flaxseed was found to be increased with the incorporation flaxseed, while it was observed that TPC and YMC were less in control laddu samples.

Textural quality parameters of flaxseed incorporated nutra laddu

A texture analysis is primarily concerned with measurement of the mechanical properties of a product. Texture analyzer performs this test by applying controlled force to the product and recording its response in the form of force, deformation and time. Hardness is the force necessary to attain a given deformation of the material or it is the force required to bite through the sample (Itagi *et al.*, 2013) [7].
Texture of flaxseed nutra laddu was measured using *TA XT2* texture analyser (stable micro system) within 24 hours after preparation. The results shows the hardness of nutra laddu are presented in table 9

Table 9: Textural quality parameters of flaxseed incorporated nutra laddu

Sample Nutra laddu	Hardness (kg)
C_0	0.872
C_1	0.896
C_2	0.923
C_3	1.212
C_4	1.456
C_5	1.721

* Each value is average of three determinations

The result from table 9 revealed that the incorporation of flaxseed significantly (P <0.05) increased the hardness of nutra laddu (0.872 to 1.721) as compared to control laddu without addition of flaxseed. As the flaseed was rich in fiber, the hardness of nutra laddu significantly influenced by addition of flaxxeed since the refined amaranth flour contained less fiber as compared to flaxseed. Similar findings by Lee and Beuchat (1991) reported that more strength was needed to break the laddu incorporated with legumes flour.

Conclusion

The movement towards enriching new functional food commodity is a novel and exciting approach to food enrichment. Nutra laddu supplementing with various amounts of flaxseed. So it can be concluded that the C3 30 percent Flaxseed sample found superior with acceptable sensorial consistency in the light of scientific evidence from the present investigation. Usage of different amounts of Flaxseed will also boost nutritional consistency of nutra laddu. The formulation was performed using Whole Flaxseed, amaranth, and Jaggery in Nutra laddu Preparation. For C0, C1, C2, C3, C4 and T5 respectively, different formulations were rendered with variation in the entire Flaxseed range from 0 to 50 per cent. Using a 9-point hedonic scale, prepared nutra laddu assessed organoleptic properties in terms of color and appearance, flavor, texture, taste and overall acceptability. The results showed that 30 percent Whole Flaxseed (C3) supplemented laddu nutra prepared with 30 percent secured highest score (i.e. 8.0) was superior to the rest of samples. Nutra laddu prepared with flaxseed has been found to be a rich source of protein and fiber. Thus, flaxseed can be well used as a functional ingredient for nutra laddu preparation with strong nutritional and medicinal value.The method of nutra laddu preparation can be techno-economically feasible, justifying the appropriateness of flax seed as a rich source of protein and energy. Nutra laddu is eaten happily by children and communities of all ages. Supplementing this product with Jaggery with different levels of flaxseed will enrich their snacks.

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