



# INTERNATIONAL JOURNAL OF ADVANCE RESEARCH, IDEAS AND INNOVATIONS IN TECHNOLOGY

ISSN: 2454-132X

Impact factor: 4.295

(Volume 4, Issue 3)

Available online at: [www.ijariit.com](http://www.ijariit.com)

## Development and evaluation of gluten-free multi-grain health mix

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### ABSTRACT

*Pseudocereals and some grains were known for special nutritional values along with gluten-free qualities. Amaranth, buckwheat, quinoa, flaxseed, ragi and soybean flours were evaluated for nutritional and sensory evaluation. Most consumers would prefer to improve their diets using food and drink related products rather than using traditional pills or capsules. The industry is looking for a variety of nutritious, convenient and user-friendly products. Although a variety of gluten-free products are available in the market still a small amount of gluten is present in additives such as modified food starch, preservatives, and stabilizers made with wheat moreover these products are very costly. Thus there is a need for the development of nutritious gluten-free products for celiac disease patients which are easy to prepare at home. Hence, the present study was conducted to understand the acceptance of blended healthy gluten-free high protein flour; the aim was to standardize an innovative, preservative free, simple, nutritional blend that is cost-effective. The product designed was multigrain health mix, having a unique blend of amaranth, buckwheat, flaxseed, quinoa, ragi, and soybean. This makes the product gluten-free, protein-rich having high fiber content. The acceptance was studied by conducting a sensory evaluation using the hedonic method based on a 9 point scale. The Evaluation was performed on sensory attributes like Appearance, color, taste, texture, aroma, and overall acceptability of the product. A total of five blends were prepared, the highest nutritional value was recorded in control C3 having a composition of 30g amaranth, 10g buckwheat, 10g flaxseed, 10g quinoa, 20g ragi and 20g soybean. The protein content in the various products ranged from 13.4 to 24.5% and fiber content ranged from 8.3 to 12.4 %. Storage study data revealed 60 days safe storage for health mix.*

**Keywords:** Pseudocereals, Gluten-free, Nutritional evaluation, Sensory evaluation, Celiac disease, Nutritional blend.

### 1. INTRODUCTION

Amaranth, quinoa, and buckwheat have received significant interest because of their gluten-free distinctiveness. Gluten is found in grains such as wheat, barley, and rye. Gluten causes inflammation in the small intestines of people suffering from the celiac disease. A gluten-free diet helps people with celiac disease control their signs and symptoms and prevent complications [1]. Celiac disease also is known as gluten-sensitive enteropathy, is an autoimmune-mediated enteropathy of small intestine characterized by permanent intolerance to specific storage proteins in wheat (gliadin), barley (hordein) and rye (secalin) which are collectively called gluten and affecting individuals with a certain genetic background [2]. It is a life long disease caused by ingestion of gluten. Celiac disease is a disease that leads to malabsorption of nutrients that are not absorbed properly whenever they consume gluten. When people with celiac disease eat foods or use products containing gluten, their immune system responds by damaging or destroying villi—the tiny, fingerlike protrusions lining the small intestine. The function of villi is to allow nutrients from food to be absorbed into the bloodstream by increasing the surface area of the small intestine. Many alternatives to common gluten-containing grains exist, such as oats, buckwheat, quinoa, sorghum, soybean, maize, rice, and besan. These grains are relatively rich in nutrients and thus, incorporation of these gluten-free grains in the gluten-free diet not only add variety but also improve its nutritional quality. Most of the subsequent reports on celiac disease are from northern India (Punjab, Haryana, Delhi, Rajasthan, and Uttar Pradesh) where wheat is the staple cereal in the diet.

Currently, the only scientifically proven treatment for celiac disease is strict lifelong adherence to a gluten-free diet. The dietary changes required by the celiac patients to begin and maintain a strict gluten-free diet are considerable and may have a significant impact on daily life. For instance, staple foods in many areas of the world such as bread and pasta, contain gluten and should,

therefore, be avoided in gluten-free diet. For most people following this diet will stop symptoms and prevent further damage. People with celiac disease can eat a well-balanced diet with a variety of foods. They can use various pseudocereals like amaranth, buckwheat, quinoa and oats, sorghum, maize, rice, pulses and soya instead of wheat flour. Rea and colleagues assessed the nutritional status of 23 Italian children with celiac disease at diagnosis and after one year of adherence to a gluten-free diet, using anthropometry, biochemical analysis, and bone densitometry measurements, it was found that although patients were clearly malnourished at the time of diagnosis, their body mass composition was virtually restored and they experienced a complete recovery after one year of a gluten-free diet [3].

Amaranth (*Amaranthus caudatus*), an ancient grain, contains about thirty percent more protein than cereals such as rice, sorghum and rye [4]. Amaranth flour particularly has an unusually rich source of the essential amino acid, lysine, which is low in other grains [5]. Buckwheat (*Fagopyrum esculentum Moench*), a well-known pseudocereal, possesses good sources of manganese, magnesium and dietary fiber as well as its gluten-free quality. Flaxseed (*Linum usitatissimum*) contains 40 per cent of total dietary fiber, 10 per cent is of soluble while 30 per cent is of insoluble fiber[6]. Quinoa (*Chenopodium quinoa*) is a pseudocereal or pseudo grain rather than a cereal or grain that has been called a superfood because of its remarkable nutritional value. Quinoa seeds contain large amounts of vitamins; protein (13% - 14%) with a good digestibility; a balanced amino acid with high lysine and methionine. Ragi (*Eleusine coracana L.*) is a good source of nutrients especially of calcium, other minerals and fiber [7]. The total carbohydrate content of finger millet has been reported to be in the range of 72 to 79.5 %.Soybean (*Glycine max*) is used for making high protein food for children. It is also used to fortify local foods so as to increase the protein content/quality of such foods. This includes mixing soybean with maize flour, cassava flour, wheat flour etc, to make fufu. There are drink mixes with soybean to boast energy as well as supply protein. Soybean contains a good amount of minerals, salt and vitamins [8].

Keeping above concepts in mind present study was planned with following objectives.

- i) To develop and standardize gluten-free health mix using organic grains.
- ii) To conduct nutritional and organoleptic evaluation of developed supplement.
- iii) To study the shelf-life of the products when packaging is done.

## **2. METHODOLOGIES**

### **2.1 Material**

The various food grains viz., amaranth, buckwheat, flaxseed, quinoa, ragi and soybean were procured from Elworld Agro and Organic Foods private limited.

### **2.2 Processing of grains**

#### **2.2.1 Cleaning**

The various kinds of food grains were taken and cleaned to remove the dirt, stones, chaffs and other foreign particles.

#### **2.2.2 Conditioning for puffing / roasting**

4-5 kg of food grains were taken and moistened with sufficient water for few minutes for gaining of moisture in between 14-15 %.

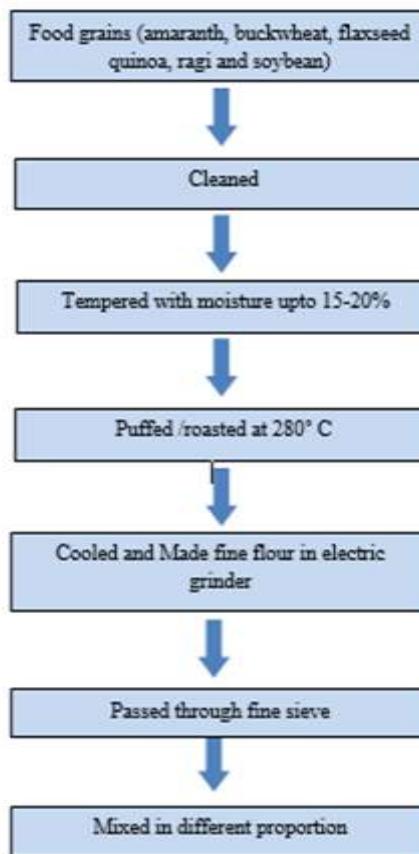
### **2.3. Preparation of health-mix**

#### **2.3.1 Preparation of grain flour**

The moist food grains of buckwheat, quinoa were washed and puffed in an iron pan for 60-90 seconds and cooled for half an hour at room temperature. The grains were ground to make fine powder in electric grinder. The other food grains (amaranth, flaxseed and ragi) were roasted /puffed on an iron pan at 280°C for 2-3 min and processed similarly. The fine powder of these food grains were stored in the air tight plastic containers at ambient conditions until required for formulation of gluten-free multi-grain health mix.

#### **2.3.2 Preparation of full fat soy flour**

Soybean grains were thoroughly cleaned to remove the dust and other foreign materials. The clean grains were tempered with water to 20-25 per cent moisture content and then autoclaved for 15 min in a pressure cooker. They were removed and dried directly in the sun for 3-4 days till the material was completely dried having 6 – 8 per cent moisture content. Soybean was then ground to make fine flour and sieved through 80-100 mesh sieve.

**Figure 1: A flowchart depicting standard preparation method for development of gluten-free health mix**

#### **2.4 Formulation of different kinds of gluten-free multi-grain health mix**

Different kinds of gluten free multi-grain health mix were developed from processed puffed/roasted amaranth, buckwheat, flaxseed, quinoa, ragi and soybean in various combinations mentioned below and they were subjected to sensory and nutritional evaluation by standard procedure.

**Table 1: Formulation of composite flour from different food grains in various combination**

Different proportion	Amaranth (g)	Buckwheat (g)	Flaxseed (g)	Quinoa (g)	Ragi (g)	Soybean (g)
C 0	100	00	00	00	00	00
C 1	50	10	10	10	10	10
C 2	40	10	10	10	20	10
C 3	30	10	10	10	20	20
C 4	25	15	15	15	15	15
C 5	20	10	10	20	20	20

### **3. EVALUATION OF GLUTEN-FREE MULTI-GRAIN HEALTH MIX**

#### **3.1 Sensory evaluation of multi-grain health mix**

The sensory evaluation of multi-grain health mix was carried out on 09 points hedonic scale for various sensory attributes and overall acceptability using untrained panelists according to the method as described by Amerine et al. (1965).

#### **3.2 Proximate evaluation of multi-grain health mix:**

The proximate analysis of various processed food products was carried out to ascertain the level of various biochemical constituents in the foods.

**Estimation of Protein:**

Estimation of total protein content by Microkjeldhal method Nitrogen content was determined using the Kjeldahl apparatus (KelPlus, Pelican Equipment,) and the amount of nitrogen was multiplied by a factor 6.25. Methods described in AOAC 1980 [9].

**Estimation of fat content:**

Estimation of oil is done by soxhlet apparatus. The fat was determined by the procedure (A.O.A.C. 1980) contains usual lipids including waxes pigments, certain gums, and resins. Crude fat was determined using the Soxhlet extractor (Soes Plus, Pelican Equipment) with ether as solvent.

**Estimation of Moisture:**

Moisture content ground nut sample was determined by conventional method i.e., drying in an oven at 100° C for 24 hours.

**Estimation of Ash:**

The sample is ignited at 600° C to burn off all organic material. The inorganic material which does not volatilize at that temperature is called ash. The procedure was described by Ranganna 1986[10].

**Estimation of crude fiber:**

Crude fiber was determined by the method as described in A.O.A.C. (1980).

**Estimation of total Carbohydrates:**

The content of total carbohydrates in the samples was determined by subtracting from 100 the sum of the values moisture, protein, fat, ash, and crude fiber.

**Estimation of Energy:**

The gross food energy was estimated by multiplying the crude protein, crude fat and total carbohydrate by at water factors 4, 9 and 4 respectively [11][12].

**Estimation of Mineral:**

The Iron content was determined calorimetrically as described in "A Manual of Laboratory Techniques" (1983). Calcium was determined by complexometric titration with 0.1M EDTA, by using Erichrome black T indicator and calculated [13]. Phosphorus content in the triacid digest was determined by vanadomolybdate method as described by Koenig and Johnson (1942).

### **3.3 Storage studies on gluten-free multi-grain health mix**

The storage studied on different kinds of gluten-free multi-grain health mix were also made in Polyethylene zip-lock pouches for two months at ambient conditions and subjected to sensory evaluation after storage.

## **4. RESULTS AND DISCUSSION**

The present investigation on "Formulation and Development of organic gluten-free multi-grain health mix" were carried out with the objectives to develop a product with high nutritional quality having good acceptability and storability.

### **4.1 Sensory analysis of organic gluten-free multi-grain health mix**

On the basis of sensory analysis of the 6 blends of health mix developed from processed flours of amaranth, buckwheat, flaxseed, quinoa, ragi and soybean in the different combinations were evaluated for various sensory attributes (appearance, colour , taste, texture, flavour and overall acceptability) on 9 points of hedonic scale as represented in Table 2. The health mix was presented to a panel of 25 untrained panelists. The obtained results showed that the values for various sensory attributes ranged from 6.3 to 8.5. The highest value was recorded in C3 (30g amaranth,10g buckwheat, 10g flaxseed, 10g quinoa ,20g ragi and 20g soybean) having an average score of 8.2 and the lowest value was recorded in C5 (20g amaranth,10g buckwheat ,10g flaxseed, 20g quinoa ,20g ragi and 20g soybean) having an average score of 6.8. Thus, the product developed in the ratio mentioned in control C3 was considered the best from sensory points of view.

**Table 2: Sensory evaluation of gluten-free health mix formulated in different proportions**

Treatment	Appearance	color	Texture	Taste	Flavor	Overall acceptability	Average mean
C 0	8.5	7.5	7.5	8.0	8.0	7.5	7.8
C 1	7.2	7.0	7.1	7.0	7.2	7.0	7.0
C 2	7.0	7.6	7.5	7.0	7.2	7.5	7.3
C 3	8.0	8.0	8.1	8.5	8.5	8.5	8.2
C 4	7.2	7.2	7.0	6.5	7.3	7.0	7.0
C 5	6.7	6.3	7.1	7.5	6.5	6.7	6.8

**4.2 Proximate composition of organic gluten-free multi-grain health mix**

The results of proximate analysis of organic gluten-free health mix made from amaranth, buckwheat, flaxseed, quinoa, ragi and soybean in the different ratio are given in tables below. Different kinds of gluten-free multi-grain health mix made from processed food grains made in the different ratio were subjected to nutritional evaluation for various nutrients viz., protein, fat, carbohydrates, ash, crude fiber, total energy, minerals, calcium, phosphorus and iron. The obtained results have been represented in Table 3. The obtained data revealed that protein content in the various products ranged from 13.4 to 24.5%, fat from 4.5 to 14.5%, total carbohydrates from 56.5 to 67.5% total ash from 2.5 to 3.6 %, crude fiber from 8.3 to 12.4 % and total energy from 336.5 to 412 kcal per 100g. The highest amount of protein was recorded in C3 and lowest in C0. The highest value of fat was recorded in C4 and lowest in C0. The highest value of total carbohydrate was recorded in C4 and lowest in C3. The highest value of ash content was recorded in C1 and lowest in C0. The highest value of crude fiber was recorded in C3 and lowest in C2. The highest value of energy was recorded in C3 and lowest in C0.

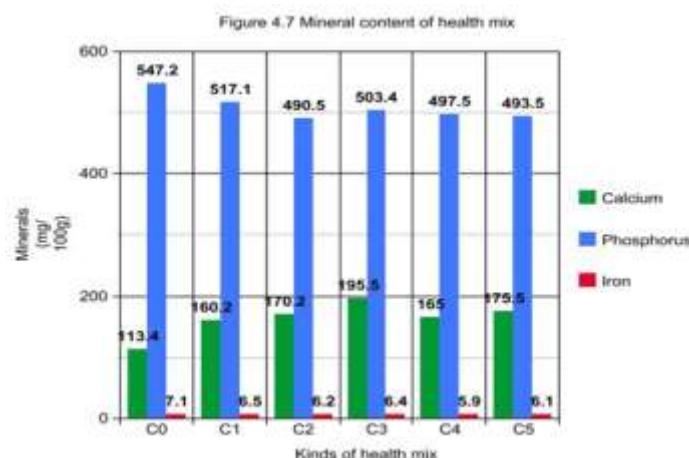
**Figure 2: Different formulated gluten-free health mix****Table 3: Nutrient composition of gluten-free health mix formulated in different proportions**

Nutrients (g/100 g)	Different kinds of health mix					
	C0	C1	C2	C3	C4	C5
Moisture	7.5	6.2	5.9	6.1	6.2	6.3
Protein	13.4	16.2	15.5	24.5	18	20.5
Fat	4.5	10.7	9.9	10.5	14.5	11.5
Total Carbohydrates	60.6	58.6	59.3	55	67.2	56.5
Total Ash	2.5	3.6	3.2	2.9	3.4	2.6
Crude Fiber	8.5	8.6	8.3	12.4	10.5	8.9
Total energy (kcal)	336.5	395	388	412	420.2	401

**Table 4: Mineral composition of gluten-free health mix formulated in different proportions**

Minerals (mg/100 g)	Different kinds of health mix					
	C0	C1	C2	C3	C4	C5
Calcium	113.4	160.2	170.2	195.5	165	175.5
Phosphorus	547.2	517.1	490.5	503.4	497.5	493.5
Iron	7.1	6.5	6.2	6.4	5.9	6.1

The quantity of important minerals viz., calcium phosphorus and iron were represented in the Table 4. The obtained results showed that calcium was found to vary in the range of 113.4 to 195.5 mg/100g product with a highest value is C3 and lowest in C0. The phosphorus content in the products were found to vary in the range of 490.5 to 547.2 mg/100g product with the highest value in C0 and lowest in C2. The iron content in various kinds of health mix ranged from 5.9 to 7.1 mg/100g product with the highest value in C0 and lowest in C4.



#### 4.3 STORAGE STUDIES OF ORGANIC GLUTEN-FREE HEALTH MIX

The various kinds of health mix developed from processed flours of various food gains were stored in polyethylene zip-lock pouches for the period of 2 months at ambient conditions. The result of sensory evaluation is represented in table 5. The obtained results on sensory quality characteristic of the products revealed that the values of various sensory attributes were found to vary in the range of 7.0 to 8.8 in the samples stored for two month.

The result showed that there were no variations in the sensory values of the products stored for 2 months. Thus, it was concluded that the products could be well stored in polyethylene zip-lock pouches for the period of 2 months without any deteriorations of the food quality.

**Table 5: Sensory evaluation of different kinds of health mix stored in polyethylene zip-lock pouches for the period of two months**

Kinds of Health-mix	Storage Period (months)	Sensory attribute			
		Taste	Flavor	Overall acceptability	average value
C1	0	8.4	8.2	8.1	8.2
	1	8.1	8.0	8.0	8.0
	2	8.0	7.9	7.8	7.9
C2	0	8.2	8.3	7.9	8.1
	1	8.0	8.3	7.8	8.0
	2	7.9	8.0	7.5	7.8
C3	0	8.4	8.2	8.6	8.4
	1	8.3	8.0	8.4	8.2
	2	8.0	8.0	8.4	8.1
C4	0	7.8	7.5	7.6	7.6
	1	7.7	7.2	7.1	7.3
	2	7.7	7.0	7.0	7.2
C5	0	8.0	8.2	8.3	8.1
	1	7.9	8.3	8.2	8.1
	2	7.9	8.2	7.9	8.0

## 5. CONCLUSION

Thus, on the basis of above observations, it was concluded that various kinds of health mix could be developed depending on the functionality of nutrients and particular types of nutritional requirements for the body. Availability and variety of gluten-free products was limited and prices were high. Cluster analysis found similarities in the nutritional content of gluten-free and conventional food products, suggesting that although raw materials different than wheat were being employed, the composition patterns are the same. Certain advantages in the composition of gluten-free products were observed, regarding mainly calories, protein and dietary fiber values .By including cereals such as soybean, ragi or pseudocereals like amaranth, buckwheat, quinoa etc in place of wheat their diet can be made nutritious because these grains have been proven to be a rich source of proteins, B-complex vitamins, minerals like iron and zinc. Gluten-free foods are special products produced for individuals with celiac disease. Nutritional quality of gluten-free products may be higher. Control C3 contains higher amount of protein, dietary fiber, calcium and other nutrients. The nutrient-rich health drink is suitable for celiac patient as well as people in metro cities and urban areas who find less time to do exercises and stay fit. Hence to keep them healthy, their food habits need to be changed. This value-added health mix using different grains will be gift for them.

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