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NUTRITIONAL AND FUNCTIONAL PROPERTIES OF A COMPLEMENTARY FOOD BASED ON KENYAN AMARANTH GRAIN (*AMARANTHUS CRUENTUS*)

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ABSTRACT

The objective of this study was to determine the nutritional and functional properties of *Amaranthus cruentus* grain grown in Kenya for preparation of a ready-to-eat product that can be recommended as infant complementary food. Amaranth grains were subjected to steeping and steam pre-gelatinization to produce a ready-to-eat nutritious product with improved solubility during reconstitution. The effect of processing on the functional and nutritional properties of amaranth grain was analyzed. Two blends were prepared from raw and processed amaranth grains. Standard procedures of Association of Official Analytical Chemists (AOAC) were used to determine the proximate chemical composition. High Performance Liquid Chromatography (HPLC) was used quantify amino acid, water soluble vitamins, α-tocopherols and phytates, while Atomic Absorption Flame Emission spectrophotometry was used to determine the mineral element composition. Fatty acid composition was determined using Gas Liquid Chromatography (GLC). Tannin composition was determined using vanillin hydrochloric acid method. The overall results indicated that processing amaranth grain did not significantly affect its nutritional and physicochemical properties. Amaranth grain product was rich in protein with 0.5 g/10g of lysine, a limiting amino acid in cereals, and methionine, a limiting amino acid in pulses. The product had good amount 44.4 mg/100g of α-tocopherols important for infant development. The product was also rich in oleic acid (36.3%) and linoleic acid (35.9%) with some amounts of linolenic acid (3.4%) that are important for infant growth. It also had good amounts of minerals of importance such as potassium (324.4 mg/100g), phosphorous (322.8 mg/100g), calcium 189.1 (mg/100g), magnesium (219.5 mg/100g), iron (13.0 mg/100g) and zinc (4.8 mg/100g). Considering amaranth grain product fed to infant three times a day, at a reconstitution of 15% product, the levels of magnesium, manganese and tocopherols were far above the recommended intakes, while protein, phosphorous, iron, zinc, riboflavin and niacin were above the average requirements. Therefore, reconstituting the product with milk would enrich the deficient nutrients, especially for iron and zinc which are crucial nutrients for infants. The processing method is a practical approach aimed at combating the problem of malnutrition among infants and young children in Kenya and other developing countries. The product developed in this study would also be appropriate for use in geriatrics care and also in immuno-compromised individuals. The technique in this study can be easily adopted at both household and village levels to produce high protein-energy weaning food to help enhance the nutritional status of Kenyans.

Key words: Complementary food, Amaranth grain, Nutrition

INTRODUCTION

The major causes of childhood malnutrition in Kenya include low rates of exclusive breastfeeding in the first 6 months of life and limited intake of appropriate foods from 6 to 24 months of age. These factors combined with high rates of infection and poor feeding practices result in high rates of morbidity [1]. Growth faltering is common among young children in developing countries, affecting as many as 33% of children under 5 years of age [2]. The high cost of fortified nutritious proprietary complementary foods is beyond the reach of most Kenyan families; most depend on inadequately processed traditional foods consisting mainly of un-supplemented cereal porridges made from maize, sorghum and millet as weaning foods. Gruels or porridges are a particular problem because they are made from staples with a high content of starch which, when cooked with water, gelatinizes and becomes highly viscous. Considerable amounts of water are commonly added to produce low-viscosity porridge, but this dilutes the nutritional value [3]. There is need to educate families to exploit alternative locally produced foods to produce nutritionally adequate products such as *Amaranthus cruentus* [4].

Amaranth grain has high protein, as well as a high fat content; there is the potential to use it as an energy food. The balance of carbohydrates, fats, and protein, allow amaranth the opportunity to achieve a balanced nutrient uptake with lower amounts of consumption than with other cereals [5]. A number of formulations of nutritious foods have been developed and nutritional qualities of raw and processed amaranth have been reported in literature.

The amaranth seed is becoming a versatile cash crop in Eastern Africa and could provide a good source of income for underprivileged families [5]. It has proved to be a breakthrough in the fight against food insecurity [5]. The challenge remains to incorporate it in the list of Kenyan staple foods. Amaranth grain is 90% digestible and because of its ease of digestion it has traditionally been given to those recovering from illness or fasting period [5]. Development of new products will expand utilization of this raw material for consumption. The nutritional value of amaranth and environmental adaptability creates an excellent potential for the crop to positively impact on thousands of poor farmers who rely on staple crops that are often neither resilient nor nutritious.

As in most other developing countries, the high cost of fortified nutritious proprietary complementary foods is always, if not prohibitive, beyond the reach of most Kenyan families, which often depend on inadequately processed traditional foods consisting mainly of un-supplemented cereal porridges made from maize, sorghum and millet. Due to the fact that amaranth grain has high protein, as well as a high fat content, there is the potential to use it as an energy-rich food. The balance of carbohydrates, fats, and protein, allow amaranth the opportunity to achieve a balanced nutrient uptake with lower amounts of consumption than with other cereals [5]. The amaranth grain is also high in minerals such as calcium, potassium, phosphorus, as well as dietary fiber, than cereal grains [6, 7]. Amaranth grains contain about 15% of protein and 60% of starch. Its amino acid profile makes it an attractive protein source and, because of its high lysine content, it tends to be viewed as a protein that, if consumed along with other cereals, can provide a "balanced" protein source [8, 9]. The amaranth seeds can be subjected to several treatments such as popping, toasting, or grinding to be consumed as suspensions with water or milk or to be included in bread, tortillas, cookies, or other preparations [10]. Tocols have been characterized in amaranth seeds and include gamma and delta tocotrienols, the unsaturated forms of vitamin E—all with antioxidant activity—and are under scrutiny as hypocholesterolemic and antitumor agents [11]

This report provides information about the processing technique and its effect on nutritional and physicochemical characteristics of amaranth grain product.

Research questions

1. Can amaranth grain be used to fight protein malnutrition? What is the amino acid composition of the common local variety?
2. Can the processing technique be adapted for household use?
3. Is amaranth product adequately nutritious as complementary food?

Hypothesis of the study

Processing amaranth grain has no effect on its physicochemical and nutritional properties.

Research Objectives

Overall objective

To develop amaranth grain complementary food product and evaluate its physicochemical and nutritional properties.

Specific Objectives:

a) To formulate amaranth grain complementary food.
b) To identify the appropriate processing method of the amaranth grain product through sensory characteristics.
c) To assess the effect of processing method of amaranth grain on the physicochemical and nutritional properties of amaranth grain product.
d) To determine the antinutrient content and physicochemical properties in the amaranth grain product.

MATERIALS AND METHODS

Research design

The research involved processing raw amaranth grain into flour for analysis of nutritional and functional properties. The treatment structure involved ungelatinized and pregelatinized amaranth grain flour referred to as the control and the product respectively. Analysis of nutritional and physicochemical properties was done in the laboratory. For each treatment, duplicate samples were each analyzed three samples.

Amaranth grain

Grains of *Amaranthus cruentus* variety were purchased from farmers in Baringo and Nakuru Districts, where the grains were ready for harvest during the period of research and could be easily sampled from the farm during harvesting. The grains were selected randomly, carefully sorted, dried in an oven to moisture content of 5%, and then stored within 48 hours at 15^{o}C before analysis.

Formulation of amaranth grain product

Two samples of ready-to-eat amaranth grain flour product was prepared from amaranth grains by the traditional wet-milling process described by Adeyemi et al. [12]. The test sample of pregelatinized amaranth flour was prepared from amaranth grains. The grains were weighed into a container and steeped overnight by soaking in warm water at temperature of 70°C (held for 10 minutes) to allow swelling of grains, degradation of soluble carbohydrates and removal of some pigments, micro-organisms and bitter substances from the grains.

After steeping, the water was decanted and the grains spread on four perforated trays, allowed to drain for 15 minutes, then steamed at atmospheric pressure for 10, 20, 30 and 40 minutes. The steamed grains were dried at 70°C in a cabinet dryer to a moisture content of less than 5%, cooled, conditioned, milled with an attrition mill and sieved to obtain flour fraction of <500 µm [13]. Steaming caused softening and cooking of the grains. The grains were dried to acquire desired moisture content and dry milled to attain a fine powder in ready-to-eat form, which was used for sensory evaluation.

The control sample of ungelatinized amaranth grain flour was also produced from the raw amaranth grains; clean grains were milled with an attrition mill to obtain flour fraction of <500 µm. The test and control samples of amaranth grain flour were packed in sealed plastic bags and stored at temperature below 15^{o}C for further analysis.

Sensory Evaluation

Sensory characteristics of pregelatinized amaranth grain product were determined by a panel of ten that consisted of mothers selected from the staff and graduate students of the Department of Food Science and Technology, Jomo Kenyatta University of Agriculture and Technology. All the panelists were conversant with the factors governing the quality of the products and infant feeding requirements. Each panelist evaluated the following characteristics: a) Color, b) Flavor, c) Texture, and d) Overall acceptability. The samples were prepared by use of clean drinking water boiled for 5 minutes and allowed to cool. The gruels prepared for this purpose were made with a standardized recipe using 15 g of the mix per 100 ml of water to obtain gruel with pouring consistency. This test was used to rate samples on the extent of starch gelatinization, using the Hedonic scale [14]. The average score of each sample was then calculated. To ascertain uniformity of judgments among the total score assigned by each of the panel members, the scores were added for the various individual characteristics and the best samples selected on the basis of overall acceptability and least steaming time.

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Chemical analyses

Standard procedures of AOAC were used to determine the moisture content, crude fat, crude protein (N x 5.85), crude fibre, ash and Nitrogen free extract (NFE) [15]. Fatty acid composition was determined using a gas chromatography system (GC-9A Shimadzu Co., Tokyo, Japan). Minerals were determined by Atomic Absorption Flame Emission Spectrophotometer (Shimadzu Corp., Tokyo Japan, Model AA 6200), using the respective cathode lamps. Phosphorus was determined with the vanadomolybdate colorimetric method [16]. α-Tocopherol was analyzed using Perkin-Elmer (PE) series 400 liquid chromatograph fitted with a UV detector using the method by Zahar and Smith [17]. Ascorbic acid, thiamin riboflavin and niacin were determined as described in the method by Ekinci and Kadakal [18] using HPLC (Shimadzu, Japan) PE series 400 liquid chromatograph fitted with a photo-diode array detector, a C_{18} stainless steel column (ODS 250 mm x 4.0 mm) at 35^{o}C oven temperature. The amino acid profile was determined with reversed-phase HPLC (Model LC-10AS, Shimadzu Corp., Kyoto, Japan) PE series 400 liquid chromatograph fitted with a binary pump delivery system, column thermostat and a fluorescence detector [19].

Antinutrient factors were determined as phytates by HPLC combining the column/mobile phase conditions [20] and tannins determined using the Vanillin-Hydrochloric acid method [21,22].

Data Analysis

Data were subjected to analysis of variance and treatment means that differed significantly (P<0.05) were separated by the least significant difference (LSD) using SAS program (SAS Institute Inc., Cary NC, USA 2002-2003).

RESULTS

The mean sensory evaluation scores for color, flavor, texture and overall acceptability of samples subjected to different heat treatment are presented in Table 1. Raw amaranth grain samples (A) were rated lowest in all characteristics, and were not significantly different ($p \geq 0.05$) in colour and flavor from the product steamed for 10 minutes (S^2). The color, flavour, texture and overall acceptability of amaranth grain samples steamed for 20, 30 and 40 minutes were not significantly different ($p \geq 0.05$) from each other but showed significant improvement ($p \leq 0.05$) compared to the 10 minutes steaming and the control, and were considered as ready to eat. It was therefore concluded that amaranth grain samples steamed for a minimum of 20 minutes could be used to make a ready to eat complementary product. Considering the economic implication of steaming, 20 minutes was taken as the benchmark for further work in this project, with the product obtained referred to as processed amaranth grain flour.

Average chemical composition of amaranth grain is shown in Table 2. Processing amaranth grain did not have a significant effect ($p \geq 0.05$) on its proximate composition; protein, ash, fibre and fat, except moisture reduction after drying, thus raising the nitrogen free extract. Amaranth grain is considered to have a unique composition of protein, carbohydrates, and lipids; thus it can provide a balanced nutrient uptake with lower amounts of consumption when compared to cereals [23,24]. In previous research, amaranth grains were found to contain about 15% of protein and 60% of starch with good amino acid profile that makes it a balanced protein source [25].

The amaranth grain oil fatty acids composition is presented in Table 3. The predominant acids in the oil were linoleic, oleic and palmitic. Total unsaturated acids ranged from 72.20% to 76.05% and saturated fatty acids 22.28% to 25.05% for the processed and raw amaranth grain, respectively. Linolenic acid was present at a low concentration.

Mineral content, tannin and phytates are presented in Table 4. The processing method did not have a significant effect ($p \geq 0.05$) on amaranth grain mineral composition. Amaranth grain product was rich in potassium, phosphorus, calcium and magnesium important for the growth of healthy bones. Processing amaranth grain led to a significant decrease ($p \leq 0.05$) in total tannins and not on phytates level. Phytic acid contents were found to be in the range reported for other amaranth species and do not seem to be unacceptably high [26].

Vitamin contents for the amaranth grain flour are presented in Table 5. Water soluble vitamins, ascorbic acid, pyridoxine, thiamine and riboflavin, were assessed. There was a significant ($p \leq 0.05$) reduction during processing on the concentration of the water soluble vitamins analysed, which is likely to affect the nutritional value of the product.

The amino acids composition obtained in amaranth grain flour samples prior to and after processing are shown in Table 6. The highest amino acid was glutamic acid followed by aspartic acid and threonine. The essential amino acids of major importance in the amaranth grain product were histidine, threonine, valine, methionine, isoleucine, leucine, phenylalanine and lysine.

DISCUSSION

From this study, it can be concluded that steeping and 20 minutes steaming pregelatinization of amaranth grain do not significantly ($p \geq 0.05$) affect its nutritional composition, except for the water soluble vitamins especially ascorbic acid which reduced with processing, due to effects of oxygen and light. Steeping is important because it improves the cleaning operation, to obtain a higher quality product. During steeping, certain physical and biochemical changes occur, such as swelling of grains, degradation of soluble carbohydrates and removal of some pigments, micro-organisms and bitter substances from the grains [27].

Amaranth grain product was rich in protein 16.7%, with high levels of lysine, a nutritionally critical amino acid, ranging from 0.7 to 0.8% of the total protein content [28]. Other essential amino acids identified were: histidine, threonine, valine, lecine, isoleucine and phenylalanine. The limiting amino acid is usually reported to be

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leucine although some reports indicate that threonine actually may be the amino acid which is more biologically limiting than leucine [29, 30]. A comparison was done for the amaranth grain product with the recommended dietary intake for infants as shown in table 7.

Grain amaranth has higher protein (12 to 18%) than cereal grains and has a significantly higher lysine content, which makes it particularly attractive for use as a blending food to increase the biological value of processed foods [31]. Furthermore, the presence of high levels of unsaturated fatty acids (oleic and linoleic) plus the high protein content makes it a nutritionally balanced grain. Amino acid content of complementary food is a particular relevant issue in infant feeding due to Protein-Energy Malnutrition (PEM), which is a major contributory factor to the high infant mortality rate in most developing countries [32].

The product had good amount of tocopherol that is important for infant growth and development. Niacin concentration was similar to 0.9 mg/100g sample, as reported for *A. cruentus* by other researchers [31,32, 33], while thiamine, riboflavin and pyridoxine were higher and ascorbic acid was lower. Niacin is important for proper blood circulation and the healthy functioning of the nervous system. Niacin helps to maintain the normal functions of the gastro-intestinal tract and is essential for the proper metabolism of proteins and carbohydrates. Niacin also helps to maintain a healthy skin and dilates the blood capillary system. Thiamin (vitamin B1), a water-soluble vitamin, is needed by infants to help the body release energy from carbohydrates during metabolism and plays a vital role in the normal functioning of the nervous system [33]. The loss of ascorbic acid during processing was quite high, which may be due to the effects of oxygen and light.

The product was also rich in fat (7.0%), ash (2.6%) and fibre (3.1%). The predominant fatty acids were oleic (36.3%) and linoleic (35.9%), with some amounts of linolenic acid (3.4%) that are important for infant growth. Amaranth oil has been found to reduce the amount of cholesterol in blood serum, and has been recommended as a functional food product for the prevention and treatment of cardiovascular diseases [34]. The inclusion of amaranth oil in the diet contributes to an increase in the concentration of polyunsaturated fatty acids and effective natural antioxidant supplement capable of protecting cellular membranes against oxidative damage [33].

The product contained good amounts of minerals of importance and poses no negative effect on the bioavailability of the essential nutrients. Phytates constitute 10-20 g kg^{-1} of many cereals and oilseeds, and amounts as high as 60 g kg^{-1} has been found in certain plant foods [34].

Considering amaranth grain product fed to infant three times a day, at a reconstitution of 15% product, the levels of magnesium, manganese and α-tocopherol will be above the recommended intakes, while protein, phosphorous, iron, zinc, riboflavin and niacin will be above the average requirements. Therefore reconstituting the product with milk would enrich the deficient nutrients and improve the complementary product, especially for iron and zinc which are crucial nutrients for infants.

The processing technique used in this study can be easily adopted at both household and village levels to produce high protein-energy weaning food to help enhance the nutritional status of children in Kenya.

CONCLUSION AND RECOMMENDATION

The study revealed that ready to eat complementary foods can be prepared from amaranth grain using locally available resources and technology. Steeping and steam pregelatinization of amaranth grain produced a ready nutritious product with improved solubility during reconstitution, suitable for infant feeding.

The processing parameters and formulations developed through this study successfully produced a high protein-energy food with acceptable physicochemical and sensory characteristics. The findings can therefore be used to promote amaranth grain production in resource poor areas of Kenya, which can increase the food output and combat infant malnutrition as a complementary food.
Further study is required for improved processing method to preserve nutrients especially the water soluble vitamins.

Table 1: Mean sensory scores for amaranth grain flour[1]

Samples	Texture	Colour	Flavour	Overall acceptability
A	4.4±0.4^c	6.0±0.3^b	2.6±0.3^b	5.6±0.4^c
S^2	5.9±0.4^b	6.7±0.4^b	2.3±0.4^b	7.0±0.3^b
S^3	8.8±0.1^a	8.0±0.3^a	6.8±0.3^a	8.7±0.2^a
S^4	8.9±0.1^a	8.0±0.3^a	6.7±0.4^a	8.8±0.1^a
S^5	8.9±0.1^a	7.9±0.3^a	6.8±0.4^a	8.8±0.1^a

[1]Each score is a mean ± standard error. In each column means followed by the same letter are not significantly different (P≤0.05).
A is the control with 100% raw amaranth grain flour
S^2 is amaranth grain steamed for 10 minutes
S^3 is amaranth grain steamed for 20 minutes
S^4 is amaranth grain steamed for 30 minutes
S^5 is amaranth grain steamed for 40 minutes.

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Table 2: Chemical composition of amaranth grain flour (grams/100 g) [1]

	Control[3]	Product[4]
Moisture	10.2±0.1[a]	2.4±0.5[b]
Protein (db)[2]	17.2±0.4[a]	16.7±0.4[a]
Fat (db)	7.0±0.1[a]	7.0±0.2[a]
Ash (db)	2.7±0.1[a]	2.6±0.1[a]
Fibre (db)	3.8±0.1[a]	3.1±0.3[a]
Nitrogen free extract(db)	59.2±0.3[b]	68.3±0.4[a]

[1]Means of two samples analysed in triplicate ± standard error. In each row means followed by the same letter are not significantly different ($P \leq 0.05$)
[2]db is dry basis
[3]Raw amaranth grain flour
[4]Processed amaranth grain flour steamed for 20 minutes

Table 3: Fatty acid composition of amaranth grain oil (%) [1]

	Control[2]	Product[3]
Lauric	0.5±0.0[b]	0.6±0.0[a]
Myristic	0.1±0.0[b]	0.2±0.0[a]
Palmitic	20.7±0.2[b]	22.3±0.1[a]
Stearic	1.5±0.0[b]	2.0±0.0[a]
Oleic	38.2±0.2[a]	36.3±0.0[b]
Linoleic	37.8±0.2[a]	35.9±0.0[b]
Linolenic	1.7±0.2[b]	3.4±0.0[a]
% Saturated fatty acids*	22.8	25.1
% Unsaturated fatty acids*	76.1	72.2

[1]Means of two samples analysed in triplicate ± standard error. In each row means followed by the same letter are not significantly different (P≤0.05)
[2]Raw amaranth grain flour
[3]Processed amaranth grain flour
*Calculated

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Table 4: Mineral and antinutrient composition of amaranth grain flour on dry matter basis (mg/100 g)[1]

	Control[2]	Product[3]
Calcium	190.7±2.9[a]	189.1±1.7[a]
Copper	0.6±0.0[a]	0.6±0.1[a]
Manganese	6.3±0.54[a]	5.9±0.7[a]
Magnesium	220.4±2.7[a]	219.5±0.7[a]
Iron	13.9±1.5[a]	13.0±0.8[a]
Zinc	5.2±0.2[a]	4.8±0.3[a]
Potassium	326.8±0.5[a]	324.4±3.9[a]
Sodium	8.1±1.5[a]	8.0±1.5[a]
Phosphorous	323.2±0.4[a]	322.8±1.0[a]
Phytates	0.3±0.0[a]	0.2±0.0[a]
Tannins	0.1±0.0[a]	0.1±0.0[b]

[1]Means of two samples analysed in triplicate ± standard error. In each row means followed by the same letter are not significantly different ($P \leq 0.05$)
[2]Raw amaranth grain flour
[3]Processed amaranth grain flour

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Table 5: Vitamin composition of amaranth grain flour on dry matter basis (mg/100 g)[1]

Sample	Ascorbic acid	Pyridoxine	Niacin	Thiamin	Riboflavin	Tocopherol
Control[2]	1.1±0.2[a]	0.6±0.2 [a]	1.4±0.1 [a]	0.6±0.1 [a]	0.5±0.1 [a]	46.7±1.5 [a]
Product[3]	0.6±0.2 [b]	0.4±0.1 [b]	0.9±0.2[b]	0.2±0.0 [b]	0.5±0.1 [a]	44.4±1.6 [b]

[1]Means of two samples analysed in triplicate ± standard error. In each column means followed by the same letter are not significantly different (P≤0.05)
[2]Raw amaranth grain flour
[3]Processed amaranth grain flour
LSD[4] is least significant difference

Table 6: Amino acids contents in amaranth grain (g/100g sample)[1]

Amino Acid	Control[2]	Product[3]
Lysine	0.5±0.1 [a]	0.5±0.0 [a]
Threonine	1.4±0.0 [a]	1.4±0.1 [a]
Valine	0.7±0.0 [a]	0.7±0.0 [a]
Cysteine	0.9±0.0 [a]	0.8±0.0 [a]
Methionine	0.6±0.0 [a]	0.5±0.0 [a]
Isoleucine	0.6±0.0 [a]	0.6±0.0 [a]
Leucine	0.9±0.0 [a]	0.7±0.0 [b]
Phenylalanine	0.4±0.0 [a]	0.4±0.0 [a]
Arginine	0.6±0.0 [a]	0.5±0.0 [a]
Alanine	0.5±0.0 [a]	0.6±0.1 [a]
Histidine	0.6±0.0 [a]	0.7±0.0 [a]
Aspartic acid	1.8±0.1[a]	1.6±0.2 [a]
Glutamic acid	7.1±0.2 [a]	5.8±1.1 [a]

[1]Means of two samples analysed in triplicate ± standard error. In each row means followed by the same letter are not significantly different ($P \leq 0.05$)
[2]Raw amaranth grain flour
[3]Processed amaranth grain flour

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Table 7: Recommended intakes of nutrient for normal infants in comparison with amaranth grain product

Nutrient	Recommended intake per day. 7-12 months[c]	Amaranth grain product composition per 100 g	Amaranth grain product at a reconstitution of 15%
Energy (kcal)[a]	850	402.4	60.4
Protein (g) [a]	14	16.7	2.5
Calcium (mg)[b]	270	189.1	28.4
Phosphorous (mg)[b]	275	322.6	48.4
Magnesium (mg)[b]	75	219.5	32.9
Sodium (mg) [a]	200	8.0	1.2
Potassium (mg) [a]	700	324.4	48.7
Iron (mg)	11	13.0	2.0
Zinc (mg)	3	4.8	0.7
Copper (µg)[b]	220	0.6	0.1
Manganese (µg)[b]	0.6	5.9	0.9
Vitamin E (mg α-tocopherol equivalents)b	6	44.4	6.7
Thiamine (mg)[b]	0.3	0.2	0.1
Riboflavin (mg)[b]	0.4	0.5	0.1
Niacin (mg NE)b	4	0.9	0.1
Pyridoxine (mg)[b]	0.3	0.4	0.1

[a]Recommend Dietary Allowance (for energy and protein) and minimum requirement (for sodium and potassium)
[b]Adequate intake (such as mean intake of normal breast-fed infants)
[c]Source: Barbara and Robert, 2001 [35].

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