# Nutritional and Functional Characteristics of Extrusion-Cooked Amaranth Flour

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#### **ABSTRACT**

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Two amaranth grain selections, GUA-17 (Amaranthus cruentus) and CAC-38 (A. caudatus), were processed into flours by extrusion cooking. Two samples of GUA-17 were cooked at 146 and 154° C and one sample of CAC-38 at 154° C using a Brady extruder. The nutritional value of the extruded flours was evaluated by net protein ratio (NPR) assays, and their functional properties were characterized by water absorption index, water retention, amylographic viscosity, damaged starch, and available lysine. The extruded amaranth flours contained slightly more protein and fat than the raw flours, suggesting some contamination with the soybean used to adjust the equipment for extrusion. Furthermore, the extruded flours had higher water absorption, water retention, and damaged starch than the raw samples. Peak Brabender viscosity was reached at 58° C and was higher for raw GUA-17 than for CAC-38, with a peak value of 46° C. Extrusion cooking decreased the viscosity in the extruded flours of both cultivars;

however, peak values for CAC-38 were observed at 25° C and for GUA-17 at 48 and 44° C. Available lysine was not affected by extrusion cooking. The GUA-17 sample extruded at 154° C had a higher NPR (3.59) than the sample processed at 146° C (3.04), and both were higher than the raw sample (2.19). Likewise, CAC-38 processed at 154° C had an NPR of 3.30 as compared to a value of 2.35 for raw. The energy density of a drink based on the GUA-17 extruded flours was partially determined by some hydrolysis of the starch through the addition of 10% germinated amaranth grain. The drum-dried product retained its original protein quality (3.14) with higher total sugar content, higher water retention, and no change in available lysine. A food product containing 45.64% of the extruded and hydrolyzed amaranth flour with 8.72% milk and 45.64% sugar was shown to be acceptable, although some panel members objected to the viscous nature of the drink.

Interest in amaranth grain has increased in recent years because of its high nutritional value as well as some agricultural advantages such as relative high grain yield, resistance to drought, and short production time (Hauptli 1977).

The protein in amaranth grain contains acceptable levels of essential amino acids, particularly lysine, tryptophan, and methionine (Senft 1980), which are found in low concentrations in cereals and leguminous grains of common usage. Therefore, amaranth could be used alone or in combination with cereal grains and with leguminous foods to improve the quality of the protein consumed by the inhabitants of developing countries. Some investigators (Betschart et al 1981, Bressani 1983) reported that cooking increases the nutritional value of amaranth grain. This suggests that crude grain contains some factors that are sensitive to heat and inhibit growth, or that processing increases the availability of proteins or nutrients in the grain.

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Extrusion cooking is often used in the processing of cereal grains and legume foods in order to obtain precooked products, texturized vegetable proteins, and to inhibit antiphysiological factors. Studies on processing amaranth grain by extrusion cooking have not yet been reported; therefore, this study was conducted to determine if extrusion cooking of two varieties of amaranth grain affected their nutritional value and their functional characteristics for food product development, particularly for preschool children.

An instant drink was prepared with the extruded flours that had good organoleptic characteristics and provided a high nutritional value.

### MATERIALS AND METHODS

In the present study, grains of two selections, GUA-17 (Amaranthus cruentus) from Guatemala and CAC-38 (A. caudatus) from Peru, were used. The grain was obtained from a field study during which the yields of the two varieties were compared. The altitude of the adaptation test was 1,300 m above sea level, with an average temperature of 21.9° C. The selection GUA-17 yielded 2,696 kg/ha, whereas selection CAC-38 yielded only 640 kg (Mendoza 1985).

Representative samples of both amaranth species were ground and analyzed for moisture, crude protein, and fat, using the AOAC

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(1970) methods.

The extrusion-cooking process was carried out using the Brady extruder model 2160. Two 40-kg batches of selection GUA-17 and one of CAC-38 were used. The extruder was operated with a constant feeding rate equivalent to 7 kg/min and a cone opening smaller than 1.5 mm in order to avoid the small whole grain from coming out without being processed. The extruder was heated to 166°C using soybean. Once the machine reached a stable temperature, the batches of amaranth grain were fed, without adding water, because of the relatively high levels of oil the amaranth samples contained.

With selection GUA-17, two samples were processed in sequence, one at a discharge temperature between 146 and 154°C and the other at a temperature ranging between 157 and 163°C. The sample of CAC-38 was cooked at a stable temperature of 154°C.

The extruded product leaving the cone during the first minute of extrusion was discarded, because it was assumed that it could contain soybean used to heat the machine. Due to limitations in the amount of raw sample, the actual sample collected for testing for approximately 5 min may not be representative of the process. However, the extruder was set at the temperatures selected when the raw grain was introduced, and after 1 min the product came out of the extruder relatively well expanded. All extruded samples were analyzed for moisture and protein content as indicated above. The three extruded samples were ground in a hammer mill provided with an 80-mesh screen. A batch of raw grain was also ground from each one of the two selections in order to use them as controls.

### **Physicochemical Tests**

The raw samples and extruded flours were evaluated for water absorption index and water retention using the method of Anderson et al (1969), damaged starch by the Farrand method (1964), amylography using the Brabender amylograph model OHG, and available lysine using the method described by Carpenter (1960).

## **Biological Tests**

With each one of the flours, diets were prepared containing 10% protein from the experimental material for net protein ratio (NPR) assays (Bender and Doel 1957). Each diet was supplemented with 4% minerals (Hegsted et al 1941), 1% cod liver oil, 5% cottonseed oil, and 5 ml of a complete vitamin solution per 100 g of diet (Manna and Hauge 1953). Corn starch was used to complete 100%.

The same procedure was used in the preparation of casein diet and one free of nitrogen as required by NPR assays (Bender and Doel 1957).

Each diet was fed to groups of Wistar rats, 21–23 days old. The animals were distributed among the experimental diets by weight so that the average weight per group was the same, and the variation within groups was not more than 1 g. Each group contained eight animals, four males and four females. The animals were placed in individual all-wire screen cages with raised screen bottoms. The diets and water were provided ad libitum during 14 days. The animals and food consumed were weighed every seven days until the experiment ended. Using these data and the nitrogen analysis of the diets, the NPR was calculated (Bender and Doel 1957).

At the end of the NPR assay, true digestibility of the protein of each of the flours was calculated. For this purpose, the animals were kept in the cages and fed with the same diets for five additional days for fecal output collection and control of food intake. The nitrogen of the dry feces was determined using the Kjeldahl method.

True digestibility was calculated by the following formula:

Diet N

where the endogenous nitrogen is the nitrogen contained in the rat's feces when fed the nitrogen-free diet (Pellet and Young 1980).

The NPR and digestibility results were submitted to analysis of variance in order to determine significant statistical differences between the values obtained.

# Starch Hydrolysis Using Germinated Amaranth Grain

Germination. A lot of 200 g of GUA-17 variety was washed several times with distilled water until the washing water was clear; then it was left to soak for 8 hr. The water was drained, and the grains were spread over absorbent towels on metal trays; later they were moistened with distilled water and covered with absorbent towels for germination for 48 hr at 25° C. Although not examined, the germinated grain did not show any microbial contamination. After germination, the grain was dried in an air oven at 60° C for 8 hr and then ground into a flour.

Treatment of extruded amaranth. A total of 1,900 g of extruded amaranth (GUA-17) was suspended in 8 L of water and heated to 80°C stirring continuously. To this suspension, 190 g of germinated pulverized grain was added when the temperature dropped to 50°C; it was mixed and allowed to react for 1 hr. The hydrolyzed sample was then dehydrated with a Reeves laboratory-size double drum-drier, heated with steam at a vapor pressure of 70 psi, a drum speed of 3 rpm, and a distance of 0.22 mm between drums.

The material so obtained was submitted to the following physical and chemical analyses: total sugars using the anthrone method (McCready et al 1950), water absorption index and water retention using the method of Anderson and co-workers (1969), and available lysine by Carpenter's method (1960). The hydrolyzed flour was used to prepare a diet for rats for assay against a reference of a nonhydrolyzed sample and casein. The techniques utilized were the same as described above for the flours obtained by extrusion. The results were statistically analyzed.

### Preparation of an Instant Drink

The ingredients used to prepare an instant drink were the extruded and partially hydrolyzed amaranth flour (45.64%), dry milk powder (8.72%), and sugar (45.64%).

### RESULTS AND DISCUSSION

The moisture, protein, and fat contents of the two amaranth selections and those of the processed samples are presented in Table I.

As shown, the first extruded product leaving the extruder had protein values higher than the raw sample or the other samples collected, a fact that demonstrates contamination of the materials by soybeans. The slightly higher amounts of protein in the other samples mainly results from an increase in the percentage of dry matter, as there was a decrease in moisture during the extrusion process. Accepting that the additional protein in the extruded flours used for the study is caused by soybean contamination, this would represent 0.2–0.6% soybean protein by calculation, which is really an insignificant amount. The fat content was also somewhat higher in the extruded flour samples than in the raw sample at equal moisture levels representing between 0.8 and 8.2% soybean oil contamination. These higher levels were not expected to interfere with the protein quality assay of the extruded materials.

TABLE I
Percentage of Moisture, Protein, and Fat
of the Raw and Processed Amaranth Samples

Variety/	Moisture	Protein	Fat
Sample	(%)	(%)	(%)
GUA-17			
Raw grains flour	10.3	16.2	6.6
Discarded sample	5.6	19.0	10.2
Extruded sample 1	5.8	17.2	7.1
Extruded sample 2	5.2	17.8	8.6
CAC-38			
Raw grains flour	10.6	17.0	5.9
Discarded sample	5.9	20.7	10.7
Extruded sample	5.4	18.1	8.0

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#### **Physicochemical Test**

The amylographic curves for the raw grain and extruded flours are shown in Figures 1 and 2.

The amylographic curve for raw CAC-38 showed maximum viscosity at 47.5° C. The curve for raw GUA-17 gave a temperature of maximum viscosity at 58° C, which was higher than for the CAC-38 cultivar.

The curves for the extruded flour of both CAC-38 and GUA-17 gave maximum viscosity temperatures, but much smaller than the respective raw flour. Comparing the two GUA-17 extruded flours, the highest temperature of processing in the second extruded sample (Ext-2) induced a lower viscosity.

The behavior of the CAC-38 extruded flour was different from the behavior of the GUA-17 flour in that the maximum point was reached at the beginning, and then the curve dropped.

Table II shows the results for the functional characteristics of the flours. The water absorption index increased significantly in both varieties due to extrusion. With GUA-17, the value increased from 0.96 to 3.51 and 3.31 for Ext-1 and Ext-2 flour, respectively. With CAC-38 the increase was from 1.41 to 2.45. These differences may result from differences in starch granule size, probably smaller for GUA-17 (A. cruentus), which is a smaller grain than CAC-38 (A. caudatus), and from the ratio of amylose to amylopectin, the latter being more abundant in amaranth starch.

The water retention percentage also increased with the extrusion process. This is because when the grain of starch is damaged, the chains of amylose and amylopectin are free to form more hydrogen bonds with water molecules.

The percentage of damaged starch increased considerably from the raw to processed samples. In GUA-17, the raw sample had a small percentage of damaged starch of 8.4, probably produced by milling. With this variety, the higher process temperature increased the percentage of damage in the starch. The highest percentage of damaged starch (81.15%) was observed in the cooked sample of CAC-38. These values are in accordance with the results of the amylographic curves that indicated that a greater degree of damage

in the starch was obtained with higher processing temperatures.

High absorption indexes, retention of water, and damaged starch in the flours indicate that they are very soluble in water.

#### **Nutritional Evaluation**

Table III presents the results obtained from the biological test. It can clearly be observed that the NPR value of the extruded samples for both selections is higher than that of the raw grain. The analyses of variance demonstrated that there are significative differences between the results of the raw and extruded samples (P < 0.05). According to the Tukey statistical test, the values of NPR for GUA-17 Ext-1 and Ext-2 belong to statistically different groups.

The above confirms the results of other authors (Betschart et al 1981, Bressani 1983) in that a controlled thermic process improves the nutritional value of amaranth grain. This suggests the presence in the grain of some antiphysiological substances that depress the growth of animals fed with raw flour. During the process, the heat deactivates these substances, increasing the nutritional value of the product. It has not yet been established which are the substances that cause this phenomenon. It is of interest to point out the significant increase in food intake of the processed as compared to the raw samples. This increased food intake may be caused by an increased palatability resulting as a consequence of the inactivation of antiphysiological factors as with food legumes. However, it could also result from an improvement in protein quality through an increase in amino acid availability. Obviously, a higher food intake will increase protein intake, resulting in improved growth of animals, and thus, in efficiency of protein utilization. An improved carbohydrate utilization may also be responsible, as other studies with sorghum have shown increased protein quality (MacLean Jr. et al 1981, 1983).

The GUA-17 Ext-2 processed sample at a higher temperature had an NPR of 3.59, higher than 3.04 for GUA-17 Ext-1. This could indicate that at a higher temperature, the quantity of inactivated antiphysiological substances is higher, or that nutrients become more easily available.

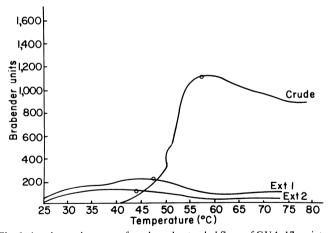


Fig. 1. Amylograph curves of crude and extruded flour of GUA-17 variety.

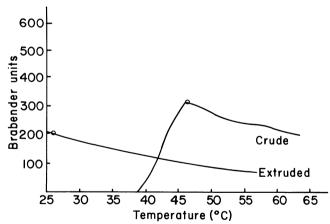


Fig. 2. Amylograph curves of crude and extruded flour of CAC-38 variety.

TABLE II
Results Obtained with the Extruded Samples of Amaranth in the Biological Test, Using Rats

		Food	Protein	Net		Lysine
Variety/ Sample	Increase in Av. Wt	Consumed (g)	Consumed (g)	Protein Ratio	Digestibility (%)	Availability (g/g N)
GUA-17						
Raw	$29 \pm 4.5$	$164 \pm 5.8$	16.2	$2.19 \pm 0.34$	$78.0 \pm 2.9$	0.39
Extruded 1 <sup>a</sup>	$60 \pm 4.0$	$220 \pm 7.1$	22.0	$3.04 \pm 0.35$	$79.0 \pm 3.1$	0.39
Extruded 2 <sup>b</sup>	$65 \pm 3.2$	$199 \pm 6.9$	19.9	$3.59 \pm 0.41$	$75.0 \pm 3.7$	0.35
CAC-38						
Raw	$21 \pm 3.7$	$123 \pm 5.0$	11.7	$2.35 \pm 0.38$	$76.0 \pm 2.8$	0.38
Extruded	$53 \pm 3.1$	$179 \pm 7.4$	18.1	$3.30 \pm 0.30$	$78.0 \pm 3.0$	0.36
Casein	$58 \pm 3.2$	$182 \pm 7.0$	20.4	$3.16 \pm 0.34$	$93.0 \pm 3.3$	

<sup>&</sup>lt;sup>a</sup>Sample processed at 146-154°C.

<sup>&</sup>lt;sup>b</sup>Sample processed at 157–163°C.

TABLE III
Functional Characteristics of Raw and Extruded Flours

Variety/ Flour	Water Absorption Index	Water Retention (ml%)	Damaged Starch (%)	
GUA-17				
Raw	0.96	1,230	8.4	
Extruded 1	3.51	1,500	59.9	
Extruded 2	3.31	1,600	70.5	
CAC-38				
Raw	1.41	1,200	0	
Extruded	2.45	1,600	81.1	

The NPR values of GUA-17 Ext-2 (3.59) and of CAC-38 (3.30) surpass the 3.16 value of casein. This confirms the higher nutritional value obtained in processing amaranth grain by a process similar to extrusion. It is unlikely for such higher protein quality values to be caused by the very small amounts of soybean protein that probably contaminated the samples, because results of other studies (Bressani et al 1984) indicate that 10% soybean addition did not significantly improve the protein quality of amaranth.

The values obtained for true digestibility are also shown in Table III; they vary between 75 and 79%. It was established that there are no significant differences between digestibility values (P < 0.05); therefore, it seems that the extrusion process does not have an effect on the digestibility of amaranth protein.

With respect to available lysine, GUA-17 Ext-2 processed at the highest temperature gave an available lysine value of  $0.35\,\mathrm{g/g}$  of N, which may represent a small loss not reflected in NPR probably because it still is enough to meet the requirements of this amino acid in growing rats. In CAC-38 the processing temperature did not produce significant changes in available lysine (0.38 versus 0.36  $\mathrm{g/g}$  of N).

The results obtained from the NPR assay and available lysine indicate that the extrusion process with the temperatures used in this study did not have any adverse effect on the quality and nutritional value of the amaranth protein. This confirms results by a number of authors (Smith 1960, Harper and Jansen 1985) that the extrusion process is a method that permits obtaining products with higher nutritional value, because it has the property of inactivating the toxicity of the substances contained in the food without affecting its protein quality.

### Starch Hydrolysis of GUA-17 Extruded Flour

Preliminary tests of solubility and suspension of the extruded flours in water demonstrated that with concentrations above 10%, products with high viscosity and low flow properties were obtained. These are not acceptable for the preparation of an instant drink fed in nursing bottles to small children, although they are quite acceptable for spoon feeding. This high viscosity and low flow rate are probably caused by the high amylopectin content found in amaranth starch (Becker 1981).

The above problem suggested the need to hydrolyze the starch to obtain a more fluid product in concentrations higher than 10%. This was done using amylases of germinated amaranth grains, as described in Materials and Methods.

Some changes taking place in the extruded GUA-17 flour no. 1 upon partial hydrolysis with germinated amaranth grain are shown in Table IV.

The total sugar values increased by partial hydrolysis from  $7.15 \, \text{mg}/100 \, \text{g}$  in the extruded flour to  $19.59 \, \text{mg}/100 \, \text{g}$  in the hydrolyzed flour. The above confirms that part of the starch was hydrolyzed to sugars such as glucose and maltose. The water absorption did not change; however, water retention values increased with the hydrolysis process, which may be due to the liberation of sugars.

As shown in Table IV, the hydrolysis and subsequent drying of the product did not significantly affect the level of available lysine, which decreased slightly from 0.39 to 0.36 g/g of N.

Table IV also presents the NPR value obtained for the hydrolyzed flour. It had a value of 3.14, as compared to a value of 3.04 for the extruded flour without hydrolysis. Statistical analysis

TABLE IV
Physical, Chemical, and Nutritional Characteristics of the Extruded
and Hydrolyzed GUA-17 Flours

Flour	Total Sugars (mg%)	Water Absorption Index	Water Retention (g%)	Available Lysine (g/g N)	Net Protein Ratio
Extruded	7.15	4.02	1,500	0.39	$3.04 \pm 0.35$
Hydrolyzed	19.59	4.08	1,815	0.36	$3.14 \pm 0.34$
Casein	•••	•••		•••	$3.18 \pm 0.51$

indicated that there is no significant difference (P < 0.05) between the two NPR values.

The biological test of NPR and the analysis of available lysine indicate that the hydrolysis processing of the starch and its drying did not affect the nutritional value of the flours previously reached by the extrusion process.

On the basis of previous studies, which indicated that 10% milk powder increased the quality of the protein of processed amaranth (Bressani et al 1984), a drink was prepared with extruded-hydrolyzed flour, skim milk, and sugar. This drink was offered to a 14-member panel who evaluated it for taste, consistency, and color. The results indicated the beverage to be highly acceptable for taste and color, although some panel members objected to its consistency, described as being somewhat viscous. In future studies this aspect will be considered by increasing the hydrolysis of the starch.

The drink could be used in the diets of preschool children with nutritional benefits because of its high nutritional value and high protein content. It is possible to improve the nutritional value of the drink if some vitamins and minerals such as vitamin A, niacin, thiamine, riboflavin, iron, phosphorus, and calcium are also added.

### **CONCLUSIONS**

The results of the present study indicate that extrusion-cooking of amaranth grain increased its protein quality, confirming previous results on the effect of thermal processing on amaranth nutritive value. At present, it is impossible to indicate the reasons for the effects observed. It is possible that amaranth grain contains heat-labile growth inhibitors, or that heat processing increases nutrient availability.

The extrusion process induced the classical changes in the physicochemical characteristics of starches reported for cereal grains with the same process.

The solubility characteristics of the extruded flours can be increased by hydrolysis of the damaged starch using germinated amaranth grain as has been reported for cereal grains. A high-quality beverage made from extruded, partially hydrolyzed amaranth flour, skim milk, and sugar was found acceptable by a panel but requires further development.

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