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Optimization of the Period of Steeping and Germination for Amaranth Grain

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ABSTRACT

According to Kenya Demographic Health Survey, 7% of children under five years were wasted with 16% of them being underweight probably an indication of poor and inappropriate feeding practices. The children suffer from protein energy malnutrition (PEM) and micro-nutrient deficiencies which may lead to physical, mental and motor development retardation. Children are most at risk of PEM during the introduction of complementary foods usually thin porridge prepared predominantly from cereals and starchy tubers. Such porridge is low in energy and nutrient density, and may be high in anti-nutrients, despite the fact that infants at this stage of rapid development have high requirements of energy and nutrients per unit body weight. There is need therefore to develop appropriate nutrient-dense complementary foods that could be used by low income families. Amaranth grain has high biological value proteins and a better amino acid profile than nearly all cereals. It is also rich in essential fatty acids. However it is not commonly used as a complementary food in Kenya. The main objective was to determine the optimum steeping and germination time for amaranth grain. The grains were steeped and germinated for various time periods. The dry matter loss, proximate composition and some antinutrient levels were determined. Dry matter loss was least in amaranth grain steeped for 5 hours and germinated for 24 hours. At $p<0.05$, there were no significant differences in ash, fat and protein contents with respect to steeping and germination time. The crude fiber content and the invitro protein digestibility varied with different steeping and germination time. The tannin and phytate contents could not be detected after steeping and germination. Based on dry matter loss and reduction in antinutrient levels, steeping amaranth grain for 5 hours and germinating for 24 hours were the optimum processing times.

Key words: Complementary foods Nutrient density Processing Antinutrients Protein Energy Malnutrition**.**

INTRODUCTION

National level estimates show that 35% of children in Kenya underfive years old are stunted, 7% are wasted and 16% are undernourished [1]. The children fail to reach their full potential growth and development, and suffer long term deprivation of energy and nutrients and consequently chronic PEM, often accompanied by micronutrient deficiencies. KDHS (2010) also reported that the most commonly used foods given to breastfeeding children under age 3 include food made from grains (72%), vitamin A rich fruits and vegetables (53%) and other milk (51%). The most commonly used first complementary food for babies in Kenya is porridge [2]. Most families often depend on inadequately processed traditional foods consisting mainly of unsupplemented cereal porridges made from maize, sorghum and millet. These staples may not contain adequate energy and nutrients. These staples are plant based. Plant-based diets are often associated with micronutrient deficits, exacerbated in part by poor micronutrient bioavailability [3]. Therefore the children may develop PEM and micro-nutrient deficiencies.

There is currently, a lot of interest in the amaranth plant, whose leaves are eaten as a vegetable in many parts of Kenya. Amaranth seed contains more protein than other grains such as wheat, maize, rice and sorghum. It contains high levels of minerals especially iron, phosphorus, magnesium, vitamin A and E. It is highly recommended for infants because of its high protein digestibility, absorption and retention by the baby`s body system. Amaranth has satisfactory lysine and tryptophan contents. However it is not commonly used in complementary food preparation. A number of traditional food processing technologies such as germination and lactic acid fermentation have been proposed as a means to improve nutrient density of complementary foods [4]. There is a chance that amaranth grain could produce a nutrient dense complementary food. However there is need to develop processing methods that would enhance the nutrient availability in the amaranth grain. This would enable promotion of its use with recommended processing methods for achievement of maximum nutrient density.

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Objectives

To determine the optimum steeping and germination time for amaranth grain.

MATERIALS AND METHODS

Raw materials

Amaranth grain were purchased from Meru County. The aim was so that the grains could be bought from the same farmers for consistency in the nutritional value.

Determination of dry matter loss

Samples were accurately weighed in to large petri-dishes and distilled water added at a ratio of 2:1. They were steeped for 5, 10, 15, 20 and 24 hours respectively. Then the petri dishes were covered with aluminium foil and kept in the dark at 30^0C. They were removed from the germination chamber at the set times (every 24 hours) and transfered in to the drying oven in the dishes used for germination in order to minimize on leaching losses. They were dried in an oven at 105^0C until constant weight (AOAC method 925.10) [5]. The difference between the dry matter of the unsoaked and the ungerminated samples and that of the steeped and germinated grains was considered as loss in dry matter.

Steeping and germination

Amaranth grain samples were weighed in to a clean gauze. They were steeped for 5 hours, 10 hours, 15 hours, 20 hours and 24 hours. After steeping for the respective time periods, they were germinated for 24, 48 and 72 hours respectively. They were then dried in an incubator at 50^0C and milled in to fine flour (200 mesh).

Determination of chemical composition of amaranth grain

The samples were analysed for protein (Kjeldahl), moisture (air oven), fat(soxlet), crude fibre according AOAC (1995). A nitrogen to protein conversion factor of 6.25 was used. Carbohydrates were determined by difference and the ash content (muffle furnace) AOAC (1995).

Determination of antinutrients in the amaranth grain

Tannin content was determined using the Vanillin-Hydrochloric acid method [6], [7] [8]. Phytates were determined the method by Camire and Clydesdale [9]. Protein digestibility was carried out using Mertz *et al* [10] procedure with slight modification.

RESULTS

Effect of germination on dry matter

For all the treatments of amaranth grain, there was an increase in the loss of dry matter with increase in steeping and germination time as shown by Figure 1. The amaranth grain samples steeped for 5 hours had the lowest loss in dry matter.

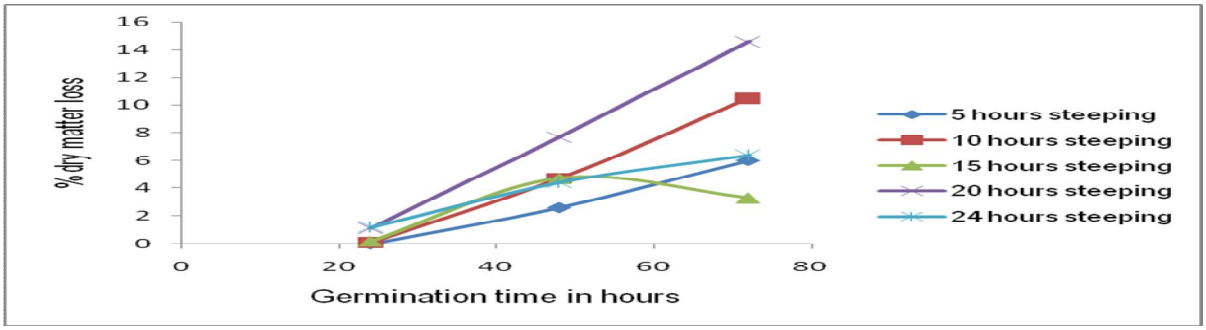

Figure 1: The effect of steeping and germination times on dry matter loss in amaranth grain

Effect of steeping and germination on the chemical composition of amaranth grain

Table 1 : The effect of steeping and germination time on the proximate composition of amaranth grain (dwb[1])

Treatment of amaranth grain	% Moisture	% Ash	% Fat	% Protein	%Carbohydrate	% Crude Fibre
Ungerminated	9.0 ± 0.2	2.7 ± 0	8 ± 0.6	15.4 ± 0.9	64.9	4.3±0.1
Steeping 5 h germ 24 h	5.8 ± 0.6	2.0 ± 0	8.6 ± 0.4	16.6 ± 0.2	67	3.9 ± 0.3
Steeping 5 h germ 48 h	6.3 ± 0.2	2.8 ± 0	8.6 ± 0.5	18.9 ± 0.0	63.4	3.7 ± 0.9
Steeping 5 h germ 72 h	5.0 ± 0.6	3.1 ± 0	9.1 ± 0.7	13.6 ± 0.2	69.3	3.4 ± 0.6
Steeping 10 h germ 24 h	6.8 ± 0.0	2.0 ± 0.1	7.3± 0.3	16.4 ± 0.0	67.5	5 ± 0.6
Steeping 10 h germ 48 h	6.6 ± 0.0	1.8 ± 0	7.3± 0.3	12.5 ± 0.5	71.8	4.4 ± 0.5
Steeping 10 h germ 72 h	6.3 ± 0.1	1.3 ± 0	8.3± 0	13.4 ± 0.2	70.7	4.0 ± 0.5
Steeping 15 h germ 24 h	6.5± 0.0	2.7 ± 0.1	11± 0.5	20.3 ± 0.1	59.5	6 ± 1.3
Steeping 15 h germ 48 h	6.6 ± 0.3	2.2 ± 0.2	8.7±0.4	14.1 ± 0.9	68.4	4.2 ± 0.7
Steeping 15 h germ 72 h	7.7 ± 0.1	2.7 ± 0	8.4±0.5	16.8 ± 0.8	64.4	8.5 ± 0.4
Steeping 20 h germ 24 h	7.2 ± 0.2	2.2 ± 0.1	8 ± 0.4	14.3 ± 0.9	68.3	5.5 ± 0.2
Steeping 20 h germ 48 h	6.6 ± 0.2	1.8 ± 0.2	8.2 ± 0.2	15.5 ± 0.3	67.9	6.2 ± 0.5
Steeping 20 h germ 72 h	5.5 ± 0.3	2.5 ± 0	7.4± 0.3	15.1 ± 0.9	69.5	6.7 ± 0.5
Steeping 24 h germ 24 h	4.5 ± 0.6	2.0 ± 0	9.3± 0.3	10.9 ± 0.9	73.3	2.5 ± 0.9
Steeping 24 h germ 48 h	5.9 ± 0.2	3.3 ± 0	8.1± 0	10.9 ± 0.9	71.8	5.4 ± 0.9
Steeping 24 h germ 72 h	5.9 ± 0.1	2.6 ± 0	8.4± 0.4	20.0 ± 0.9	63.1	5.4 ± 0.4

dwb[1] : Dry weight basis Germ: germination h: hours

Table 2: The effect of steeping and germination on some antinutrients in amaranth grain (dwb[1])

Treatment of amaranth grain	Tannins (% Catechin equivalents CE)	Phytate (mg per 100 g)	Invitro protein digestibility (IVPD) (%)
Ungerminated	0.8	7.9	77.6
Steeping 5 h germ 24 h	ND	ND	93.2
Steeping 5 h germ 48 h	ND	ND	93.2
Steeping 5 h germ 72 h	ND	ND	93.1
Steeping 10 h germ 24 h	ND	ND	99.2
Steeping 10 h germ 48 h	ND	ND	93.4
Steeping 10 h germ 72 h	ND	ND	97.4
Steeping 15 h germ 24 h	ND	ND	87.8
Steeping 15 h germ 48 h	ND	ND	94.9
Steeping 15 h germ 72 h	ND	ND	97.2
Steeping 20 h germ 24 h	ND	ND	97.2
Steeping 20 h germ 48 h	ND	ND	96.7
Steeping 20 h germ 72 h	ND	ND	94.7
Steeping 24 h germ 24 h	ND	ND	89.4
Steeping 24 h germ 48 h	ND	ND	81.5
Steeping 24 h germ 72 h	ND	ND	97.8

dwb[1] Dry weight basis
ND Not detected

DISCUSSION

Effect of steeping and germination on the dry matter of amaranth grain

The greater the germination time, the more the grains sprouted therefore increasing consumption of nutrients and increasing the dry matter loss. At $p<0.05$ level of significance there were interactions between steeping and germination time for the amaranth grain. Long germination periods resulted in significant losses in dry matter through respiration which is undesireable [4]. Wijngaard *et al [12]* reported that increase in steeping time increased malting losses. He further reported that steeping losses are mainly due to three factors: displacement of dust, dissolving of materials from the grain by leaching and metabolic activity of the grain, releasing CO2 and small amounts of ethanol. Malting losses result from leaching of compounds from grain during steeping, respiration of the grain and fermentative processes and the removal of rootlets [12]

Effect of steeping and germination on the proximate composition of amaranth grain

The ash content is almost similar to the amount got by other researchers. The ash content of amaranth cruentus was reported as 3.2% on dry weight basis [12], [13]. Ruiz and Bressani [14] reported that the ash content in ungerminated amaranth grain is 3.0%. Ruiz and Bressani [14] reported that there were no changes in ash content with respect to germination time.The ungerminated amaranth grain had a fat content of 8% dry weight basis. At $p<0.05$, there were no significant differences in fat content with regard to steeping and germination time. Ruiz and Bressani [14], reported that the fat content in ungerminated amaranth crentus is 7.1% on dry weight basis. According to Ruiz and Bressani [14] there was no significant change in fat content extracted using ether during germination.

The crude protein content in ungerminated amaranth grain was 15.4%. After 5 hours of steeping the crude protein content increased to 16.6%. At $p< 0.05$ there were no significant differences in protein content with respect to steeping and germination times. Ruiz and Bressani [14] reported that the protein content in ungerminated amaranthus crentus as 14.6%. They also reported that there was no significant change in protein content with increase in germination time. Mbithi-mwikya *et al* [4] reported that there was a slight but significant increase in protein content of fingermillet at each sampling time, from 6.1% in ungerminated seeds to 7.9% during the 96 hours of germination. Mbithi-mwikya[4], attributed the increases in protein content to be due to dry matter loss particularly through carbohydrates through respiration causing an apparent increase in other nutrients such as proteins. Khalil *et al* [15], also reported a slight increase in protein content after germination of soybean and lupin seeds for 72 hours. Ungerminated amaranth grain had a crude fibre content of 4.3%. The crude fiber content of amaranth grain varied with different steeping and germination time as shown by Table 2. At $p<0.05$, there were significant differences in the crude fibre content of the amaranth grain samples. Ruiz and Bressani [14] reported that the crude fibre content of amaranth crentus is 2.2% and that there was no significant change in crude fibre content of amaranth grain with change in germination time.

Effect of steeping and germination on some antinutrients in amaranth grain

Ungerminated amaranth grain had a tannin content of 0.8% CE. When amaranth grain was germinated tannin content could not be detected. It must have been reduced to very low levels. Whittaker and Ologunde [16], reported that the tannin content in raw amaranth grain is 0.22 mg CE/ 100g. Hemalatha *et al* [17] reported that germination significantly

reduced the tannin content in some food grains such as green gram, chickpea and fingermillet.

In ungerminated amaranth grain the phytate content was found to be 7.9 mg per 100g. When the amaranth grain was germinated, their phytate content could not be detected. This could mean that the phytates were all broken down from the inosital hexaphosphate form. Whittaker and Ologunde [16] reported that phytate content in raw amaranth cereal is 7.92 mg/g. Ruiz and Bressani [14] reported the phytic acid content in amaranth crentus grain as 0.29%. However with increase in germination time the phytic acid content decreased and after 72 hours of germination the phytic acid content could not be detected. Archana [18] reported that malting with 72 hours of germination was most effective in reducing the antinutrient levels of pearl millet grains. Because germination is mainly a catabolic process that supplies important nutrients to the growing plant through hydrolysis of reserve nutrients, reduction in phytic acid was expected [18]. Since phytic acid may be one of the factors responsible for reducing mineral bioavailability its reduction during germination may enhance the nutritional quality with respect to mineral bioavailability of amaranth grain.

Ungerminated amaranth grain had an IVPD of 77.6% on dry weight basis. After steeping for 5 hours and germinating for 24 hours the IVPD increased by 15.6%. The IVPD varied with various steeping and germination times as shown by Table 2. Mbithi-mwikya[4] reported that IVPD increased from 33.9% in the ungerminated seeds of fingermillet to 55.4% at 96 hours, an increase of 64%. They suggested that partial solubilization and some proteolysis which usually occurs during germination could have caused this. Germination of ingredients increased nutrient density and invitro protein digestibility [20]. Chauman and Kumar [19] reported that percent protein digestibility (in vitro) of pearl millet grain improved following germination at all temperatures. They reported that the improvement in protein digestibility during germination may be attributed to modification and degradation of storage proteins of the grain. Sprouting causes mobilisation of proteins with the help of activated proteases, leading to the formation of polypeptides, oligopeptides and amino acids. Furthermore hydrolytic reduction of phytates during germination may also partly account for the improved protein digestibility of millet sprouts because phytates are known to inhibit proteases [19].

Conclusion

There was change in the proximate composition of the amaranth grain with processing. However loss in dry matter will affect the total nutrients. Therefore the lesser time for steeping and germination the better. The antinutrient level are generally lower than other grains and are reduced to levels they can not be determined even with minimum processing periods.

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