



MILLET GRAINS

## THE EFFECTS OF GERMINATION PERIODS ON PROXIMATE, MINERALS AND ANTINUTRIENT PROFILE OF PEARL MILLET (*Pennisetum glaucum*) AND GRAIN AMARANTH (*Amaranth cruentus*) FLOURS

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Amaranth plant with grain

### INTRODUCTION & AIM

Cereal-based foods serve as fundamental components of the diet for both adults and young children, particularly in the preparation of infant complementary food in rural and impoverished urban communities across Africa (Ramashia et al., 2021). They serve as the primary source of carbohydrates and proteins, along with other crucial nutritional components within rural and impoverished urban demographics (Balakrishnan & Schneider, 2022).

Grain amaranth has garnered significant interest as a pseudo-cereal renowned for its exceptional nutritional properties compared to other primary cereal crops (Fabiola et al., 2019). Cereal continues to serve as a primary source of raw material for infant gruel and porridge in African nations, including Nigeria, Ghana, and Uganda to mention few (Temba et al., 2016). Of the various millet varieties, pearl millet is predominantly cultivated in Africa and Asia, leading to a heightened research focus in this domain (Sharma et al., 2021).

Amaranth grains are rich in essential amino acids, exhibiting a more balanced amino acid composition in comparison to the majority of cereals. Combining amaranth grains with other protein nutrient deficient cereals provides a potential avenue for creating a protein source with a balanced amino acid profile. Furthermore, amaranth is a noteworthy reservoir of essential minerals, including calcium, magnesium, and iron (Akin-Idowu et al., 2017).

Although cereals are known to contain array of macronutrients, micronutrients, and phytochemicals good for health, however, it is important to note that they also have anti-nutritional factors that have the potential to limit the bioavailability of these crucial nutrients within the body (Samtiya et al., 2020). Germination represents an economically viable processing technique suitable for household applications and has been proposed as an effective method for mitigating anti-nutritional factors such as phytates, oxalate, tannins, and trypsin inhibitors. This study aims to evaluate the influence of varying germination periods as a pretreatment to enhance the nutritional composition of millet and amaranth grains processed into flours. The findings are anticipated to offer valuable insights for rural nursing mothers contemplating the utilization of germination as a pretreatment method for the preparation of baby gruel using amaranth and millets.

### METHOD

#### ❖ Raw material collection

The raw materials were obtained as follows: Pearl millet (*Pennisetum glaucum*) was acquired from the Bodija market in Ibadan, while grain amaranth seeds (*Amaranthus cruentus*) were sourced from the field at the National Horticultural Research Institute (NIHORT) in Ibadan, Nigeria

#### ❖ Preparation of germinated millet and grain amaranth flours

Germinated millet and amaranth grain flours were prepared according to the methodology outlined by Okoth et al. (2011) and Mohammed et al. (2023). The grains underwent sorting, thorough washing, and immersion in water at a grain-to-water ratio of 1:3 (w/v) for 24 hours at a constant room temperature of  $28 \pm 0.1^\circ\text{C}$ . Subsequently, the grains were enveloped in a clean white cloth and subjected to varying germination periods of 24, 48, and 72 hours. Following germination, the grains were subjected to drying at temperatures ranging from  $45\text{--}50^\circ\text{C}$  and were subsequently milled using a Marlexcella electric grinder, model 750. The resultant flour was finely powdered through a 0.25mm-diameter sieve and then stored in an airtight container.



1-Sorted grains



2-Wrapped grains for germination



3-Sprouting of the grains



4-Drying of the germinated grain

Fig 1-Simple illustration of germination processes

#### ❖ Proximate Determination

The samples were analyzed for moisture, crude protein, ash, fat, and crude fiber using AOAC (2010). Carbohydrate content was calculated as  $\% \text{Carbohydrate} = 100 - \% (\text{Moisture} + \text{Crude Protein} + \text{Fat} + \text{Ash} + \text{Crude Fiber})$ . Energy content was determined by multiplying the percentage values of protein, fat, and carbohydrate by the Atwater

#### ❖ Mineral Analysis

Weigh approximately 2g of the samples and heat them at  $550^\circ\text{C}$  in a muffle furnace (Model: KejianKinsgeoKJ-3020). Then, dissolve the ashes with 100 ml of 1M HCl. The dissolved ash should be analyzed for zinc, iron, calcium, potassium, sodium, and magnesium contents using an atomic absorption spectrophotometer (Model: Buck Scientific ACCUSYS 211).

#### ❖ Phytate

The phytate content was determined following the method described by (wuozor,2019).

#### ❖ Oxalate content

Oxalate content was determined as described by (Chinma and Igyor 2007).

#### ❖ Tannin content

Tannin content was determined according to method described by name (Ijarotimi, 2012).

#### ❖ Trypsin inhibitor determination

Trypsin inhibitor unit was determined using the method described by (Panta, 2017).

## RESULTS & DISCUSSION

Table 1: proximate composition of millet and amaranth flours as influenced by different germination periods

Samples	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Fibre(%)	CHO (%)	Energy(Kcal/100g)
<b>Millet (X1)</b>							
Control	9.44±0.06 <sup>d</sup>	7.71±0.02 <sup>d</sup>	7.67±0.02 <sup>a</sup>	1.11±0.01 <sup>abc</sup>	2.26±0.01 <sup>b</sup>	71.49±0.01 <sup>d</sup>	366.69±0.01 <sup>d</sup>
24hrs germination	8.30±0.01 <sup>c</sup>	8.61±0.01 <sup>a</sup>	5.72±0.01 <sup>bcd</sup>	1.17±0.01 <sup>ab</sup>	3.07±0.01 <sup>a</sup>	73.79±0.01 <sup>b</sup>	385.02±0.01 <sup>a</sup>
48hrs germination	8.81±0.01 <sup>b</sup>	8.54±0.15 <sup>ab</sup>	5.76±0.15 <sup>b</sup>	1.19±0.10 <sup>a</sup>	3.03±0.0 <sup>c</sup>	74.38±0.02 <sup>a</sup>	382.42±0.10 <sup>b</sup>
72hrs germination	8.10±0.01 <sup>a</sup>	8.07±0.01 <sup>c</sup>	5.73±0.01 <sup>bc</sup>	1.19±0.01 <sup>a</sup>	3.08±0.01 <sup>a</sup>	73.40±0.01 <sup>bc</sup>	379.33±0.10 <sup>c</sup>
<b>Amaranth (X2)</b>							
Control	8.41±0.01 <sup>a</sup>	16.08±0.12 <sup>c</sup>	5.75±0.01 <sup>d</sup>	2.19±0.01 <sup>d</sup>	4.37±0.01 <sup>d</sup>	63.28±0.10 <sup>b</sup>	371.16±0.01 <sup>d</sup>
24hrs germination	7.20±0.01 <sup>d</sup>	17.37±0.01 <sup>a</sup>	6.75±0.05 <sup>c</sup>	2.65±0.01 <sup>ab</sup>	4.62±0.01 <sup>c</sup>	62.46±0.01 <sup>d</sup>	379.61±0.01 <sup>a</sup>
48hrs germination	7.51±0.01 <sup>c</sup>	16.73±0.01 <sup>b</sup>	7.64±0.01 <sup>b</sup>	2.47±0.01 <sup>c</sup>	4.68±0.02 <sup>b</sup>	63.18±0.11 <sup>c</sup>	375.03±0.01 <sup>b</sup>
72hrs germination	7.70±0.04 <sup>b</sup>	16.05±0.56 <sup>cd</sup>	8.13±0.20 <sup>a</sup>	2.69±0.02 <sup>a</sup>	4.97±0.01 <sup>a</sup>	63.61±0.11 <sup>a</sup>	374.69±0.01 <sup>bc</sup>

Note: Values are means ± standard deviation of duplicate determinations. Values with the same superscripts in the same column are not significantly different at p < 0.05.

The protein content in germinated millet flour was found to be 8.61% after 24 hours, exhibiting a significant increase ( $p < 0.05$ ) compared to values recorded at 48 hours (8.54%), 72 hours (8.07%), and in ungerminated millet (7.71%). Furthermore, the highest protein content of 17.37% was observed in germinated grain amaranth after 24 hours of germination, representing a significant increase compared to other time periods and ungerminated amaranth (Table 1).

Some documented studies reported an increase in crude protein levels in germinated grains, including oats, waxy wheat, and barley (Youssef et al., 2013). Bozena and Dariusz (2012) observed a higher protein concentration (7.40%) after 24 hours of germination compared to 48 hours (6.83%) during the germination of bean seeds. Fabiola et al. (2019) in study of physicochemical and nutritional changes in two amaranth species (*Amaranthus quitensis* and *Amaranthus caudatus*), noted that a short germination period of 24 hours appeared sufficient to induce significant alterations in the physicochemical and nutritional properties of amaranth, as the protein concentration substantially increased after 24 hours compared to ungerminated seeds.

Table 2. Mineral composition of millet and amaranth grain flours as influenced by germination period

Samples	Fe(mg/100g)	Ca(mg/100g)	Zn(mg/100g)	Mg(mg/100g)	K(mg/100g)	Na(mg/100g)
<b>Millet</b>						
Control (RM)	3.31±1.10 <sup>d</sup>	7.32±0.02 <sup>c</sup>	3.32±0.02 <sup>c</sup>	9.41±0.01 <sup>d</sup>	251.73±0.63 <sup>d</sup>	30.01±0.01 <sup>d</sup>
24hrs germination	4.77±0.01 <sup>c</sup>	9.40±0.03 <sup>ab</sup>	4.92±0.02 <sup>b</sup>	12.07±0.01 <sup>a</sup>	257.50±0.51 <sup>c</sup>	33.01±0.02 <sup>c</sup>
48hrs germination	4.90±0.02 <sup>ab</sup>	9.49±0.01 <sup>b</sup>	4.94±0.01 <sup>b</sup>	11.29±0.00 <sup>c</sup>	268.83±0.55 <sup>b</sup>	33.66±0.11 <sup>b</sup>
72hrs germination	4.94±0.01 <sup>a</sup>	10.82±0.01 <sup>a</sup>	5.60±0.08 <sup>a</sup>	12.03±0.01 <sup>b</sup>	282.73±0.55 <sup>a</sup>	43.54±0.11 <sup>a</sup>
<b>Amaranth</b>						
Control	3.50±0.12 <sup>d</sup>	33.14±0.31 <sup>d</sup>	3.17±0.01 <sup>d</sup>	90.01±0.01 <sup>d</sup>	250.83±0.60 <sup>d</sup>	25.01±0.01 <sup>d</sup>
24hrs germination	4.10±0.01 <sup>c</sup>	38.74±0.01 <sup>b</sup>	4.92±0.02 <sup>b</sup>	94.16±0.02 <sup>b</sup>	271.43±0.60 <sup>b</sup>	30.20±0.01 <sup>c</sup>
48hrs germination	5.86±0.01 <sup>ab</sup>	38.59±0.01 <sup>bc</sup>	4.81±0.01 <sup>bc</sup>	94.30±0.01 <sup>b</sup>	264.30±1.92 <sup>b</sup>	30.81±0.02 <sup>b</sup>
72hrs germination	5.89±0.01 <sup>a</sup>	40.80±0.01 <sup>a</sup>	5.75±0.01 <sup>a</sup>	95.81±0.01 <sup>a</sup>	291.36±0.50 <sup>a</sup>	37.70±0.01 <sup>a</sup>

During the 24-hour germination period, the iron (Fe) concentration in millet flour was 4.77 mg/100 g, which was lower than the concentrations observed at 48 hours (4.90 mg/100 g) and 72 hours (4.94 mg/100 g) of germination. However, it was significantly higher than the concentration in ungerminated millet flour, which was 3.31 mg/100 g.

These results demonstrate that the longer the germination period, the higher the concentration of minerals. A similar improvement in mineral concentration was also observed in germinated amaranth flour compared to ungerminated.

Iron and zinc are trace minerals that are essential for infants' normal growth and development, especially during their first 6 months of life.

Iron deficiency can lead to anemia, which is common in early childhood and late infancy (Fewtrell et al., 2017). Zinc deficiency in malnourished children can result in stunted growth and increased susceptibility to infections, such as diarrhea and pneumonia (Cormack et al., 2019).

Table 3: Anti-nutritional factors of millet and amaranth grains as influenced by different germination period

Samples	Tannin(g/100g)	Oxalate(g/100g)	Phytate(g/100g)	Trypsin.I(TTU/g)
<b>Millet</b>				
Control (RM)	0.850±0.00 <sup>a</sup>	0.620±0.12 <sup>a</sup>	0.836±0.01 <sup>a</sup>	0.570±0.00 <sup>a</sup>
24hrs germination	0.416±0.05 <sup>b</sup>	0.253±0.01 <sup>b</sup>	0.326±0.05 <sup>b</sup>	0.363±0.01 <sup>b</sup>
48hrs germination	0.333±0.11 <sup>c</sup>	0.251±0.01 <sup>bc</sup>	0.230±0.00 <sup>c</sup>	0.341±0.00 <sup>c</sup>
72hrs germination	0.323±0.05 <sup>cd</sup>	0.213±0.01 <sup>d</sup>	0.173±0.11 <sup>d</sup>	0.340±0.01 <sup>cd</sup>
<b>Amaranth</b>				
Control(RA)	0.633±0.12 <sup>a</sup>	0.520±0.52 <sup>a</sup>	0.613±0.81 <sup>a</sup>	0.656±0.85 <sup>a</sup>
24hrs germination	0.273±0.05 <sup>b</sup>	0.223±0.21 <sup>b</sup>	0.286±0.18 <sup>b</sup>	0.513±0.46 <sup>b</sup>
48hrs germination	0.163±0.05 <sup>c</sup>	0.213±0.22 <sup>c</sup>	0.226±0.21 <sup>c</sup>	0.460±0.59 <sup>b</sup>
72hrs germination	0.154±0.33 <sup>cd</sup>	0.196±0.19 <sup>d</sup>	0.210±0.23 <sup>cd</sup>	0.446±0.44 <sup>d</sup>

various degree of reduction in concentration of the antinutritional factors was observed in germinated millet and amaranth flours as compared to un-germinated.

The concentration of tannin and phytate in millet flour significantly ( $p < 0.05$ ) reduced from 0.850 g/100 g and 0.836 g/100 g to 0.416 g/100 g and 0.326 g/100 g, which represent 48.9% and 61.0% reduction respectively after 24 h germination period. A significant reduction in antinutrient concentration was observed also in amaranth flour as influenced by periods of germination

Anti-nutritional factors are secondary metabolites that affect the nutritional value of foods (Abera et al., 2023). However, their concentration can be reduced using traditional food processing techniques such as germination (Senlik & Alkan, 2023).

Germination lower anti-nutritional factors and consequently increase the bioavailability of essential nutrients such as minerals and protein.

Thus, germination process could be traditionally engaged in the treatment of cereals to reduce their antinutrients as observed in this study.

## CONCLUSION

Cereals are integral to human diets due to their rich content of vital nutrients such as carbohydrates, protein, and minerals. However, their nutritional value can be compromised by antinutritional factors. This study indicates that the presence of antinutritional factors in pearl millet and grain amaranth seeds can be reduced through germination pre-treatment, consequently enhancing their overall nutritional contents. Notably, a 24-hour germination period has been identified as giving the best in protein content, a critical factor in infant nutrition and development

## FUTURE WORK / REFERENCES

### Future work

Study should extend to comparing the effect of other processing techniques such as fermentation period on nutritional profile of millet and amaranth flours.

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