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Effect of germination on the physico-functional and nutritional profile of barnyard millet (*Echinochloa frumentacea*)(VL-172)Manasa R^{1*}, Shekhara Naik R¹¹Dept. of Food Science and Nutrition, Yuvaraja's College University Mysuru, Karnataka, India

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ABSTRACT

Background: Barnyard millet is well-known for its abundant nutrients and adaptability, making it a promising candidate for product innovation, especially by exploring new processing methods like germination to boost its nutritional value and effectiveness. It is vital to examine both the physico-functional and nutrient characteristics of barnyard millet in order to fully harness their nutritional advantages in product development. Understanding factors such as grain size and properties like water and oil absorption capacity is crucial for creating inventive, nutritionally enriched foods that can meet a variety of dietary requirements and gain wider market acceptance.

Objectives: This scientific approach fosters the effects of germination on the physico-functional and nutritional properties of Barnyard millet.

Materials and Methods: Barnyard millet was collected and soaked for 12 hours and germinated in different time intervals, then dehydrated at 110°C for 4 hours. Physical properties such as kernel size and volume were analysed. Functional properties, including oil and water absorption capacity, water solubility and nutritional analysis were conducted.

Results and Discussion: The study revealed that as the germination time increased, the length-width ratio increased, accompanied by a slight increase in 1000 kernel weight, volume and decrease in bulk density of germinated millet. Furthermore, functional properties such as oil absorption capacity, water absorption capacity and water solubility index increased with germination time, contributing to improved texture. There was increase in protein and fiber, slight decrease in fat content. The mineral content was increased upon germination.

Conclusion: The obtained results indicated slight variations in the physical and functional and nutritional characteristics of barnyard millet and its flour upon germination.

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1. Introduction

Millets are often hailed as "crops of the future" due to their resilience against a multitude of pests and diseases, coupled with their remarkable adaptability to the challenging conditions prevailing in the arid and semi-arid regions of Asia and Africa.¹ Barnyard millet, also known as Ooda, Oadalu, Sawan, and Sanwa, is a small-sized grain

highly valued for its superior nutritional profile. Cultivated primarily in India, China, Japan, and Korea, it serves both human consumption and fodder purposes, prized particularly for its drought tolerance.^{2,3} Barnyard millet offers a rich array of macronutrients, micronutrients, and nutraceutical properties, including carbohydrates, protein, fats, and fiber, with notable levels of calcium and iron. Its high digestibility and low content of slowly digestible carbohydrates position it as an ideal ingredient for a variety of manufactured food products, including baby food,

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snacks, and dietary foods.^{4,5} Millets contain anti-nutrients that can hinder the absorption of minerals like iron and zinc, but certain processing techniques can enhance nutrient digestibility and bioavailability.⁶

Germination unlocks enzymatic activity in grains, converting cereal sugars to fermentable sugars, offering a transformative solution to underutilized millets in product development, particularly through the utilization of germinated millet flour in value-added products.⁷ Simple traditional processing methods such as soaking and germination can substantially decrease the anti-nutrient levels in cereal grains and enhance the bioavailability of nutrients by boosting phytase activity, which hydrolyses phytic acid. Recent studies confirm that well-structured soaking and germination stages effectively reduce phytates and tannin contents in millet grains.^{8,9}

2. Objectives

1. To evaluate the impact of germination on physical and functional properties of barnyard millet.
2. To estimate the nutrient composition of barnyard millet with varying germination time.

3. Materials and Methods

3.1. Raw materials

The research was conducted in the Department of Food Science and Nutrition at Yuvaraja's College, University of Mysore, Mysuru. Pristine samples of Barnyard millet (*Echinochloa frumentacea*) (VL-172) were procured from the fields of UAS Seeds, V. C Farm, Mandya district, Karnataka.

3.2. Method of germination

The barnyard millet grains were sorted and cleaned to remove impurities. The grains were washed and soaked for 12 hours. The water was drained out from grains; it was washed again and tied airtight with the muslin cloth and kept in incubator at 37°C for 12, 24, 36, 48, 60 and 72 hours. Later, dehydrated at 110°C for 4 hours and then milled to produce fine textured powder.⁷

3.3. Physical properties of processed barnyard millet

3.3.1. Length-width ratio(mm)

The average length and width of the randomly picked ten grains were measured in mm with the help of Vernier calipers. The length-width ratio was obtained by dividing the length of a single grain by the corresponding width to determine the size.¹⁰

3.3.2. Sprout length(mm)

The sprout length of the germinated millets was measured using the millimetre scale(mm) at different germination time period.^{11,12}

3.3.3. Kernel grain weight(g and volume(ml))

Thousand kernel weights were measured by selecting 1000 grains randomly from pre-cleaned grains. The selected kernels were weighed on a digital electronic balance. The test was performed in triplicates and the mean value was calculated. The same method was followed for 1000 kernel volume by multiplying each value by 10 to obtain thousand-grain volume and is expressed in ml.¹³

3.3.4. Bulk density(g/ml)

Bulk density was measured using calibrated measuring cylinder of 1000 ml capacity. The cylinder was filled to appropriate height with the clean grains. Bulk density was calculated by taking ratio of the sample weight and volume of the cylinder and is represented as g/ml. Average of 3 replications was taken.¹⁴ $Bulk\ density = \frac{Sample\ weight}{volume}$

3.4. Functional properties

3.4.1. Oil absorption capacity(OAC)

0.5 g of Barnyard millet flour was taken with 5 ml of oil, kept for 30 min at room temperature, and then centrifuged for 25 min at 3000 rpm. Sediments were weighed and Oil absorption capacity was calculated by given formula.¹⁵

$$OBC = \frac{W_2 - W_1}{W_0} \times 100$$

W₀ - weight of the sample

W₁ - weight of centrifuge tube + sample

W₂ - weight of centrifuge tube + sediments

3.4.2. Water absorption capacity(WAC)

1 gram of Barnyard millet flour was taken in centrifuge tube and mix it with 10 ml of distilled water and agitate it 4 times allowing 10 min resting period between each mixing followed by centrifugation at 3000 rpm for 25 min. The supernatant was decanted and tubes were air dried and weighed.¹⁶

$$WAC\ (ml/g) = \frac{Volume\ of\ water\ absorbed \times 100}{Weight\ of\ the\ sample}$$

3.4.3. Water solubility index(WSI)

2.5 g of sample mixed with 30 ml of distilled water and kept at 90°C for 15 minutes in water bath, then cooled to room temperature. Centrifuged at 3000 rpm for 10 min. Supernatant was decanted and weight of the sediments and the weight of dry solid is determined by evaporating the supernatant at 110°C.¹⁶

$$WSI = \frac{Weight\ of\ dissolved\ solids\ in\ supernatant}{Weight\ of\ sample} \times 100$$

3.4.4. Swelling power(SP)

500 mg of the sample was weighed(W1), placed into centrifuge tube and weighed(W2) again. 20 ml of distilled water was added and heated for 30 min in a water bath at 90°C, with occasional stirring; the tubes were cooled and centrifuged at 5000 rpm for 10 min. The supernatant was decanted into a pre-weighed petri plate and dried at 105°C and weighed. The inner side of the centrifuge tube was wiped, dried and weighed (W3).¹⁷ Swelling power was calculated using the following formulae:-

$$SP (g/g) = \frac{W_3 - W_2}{W_1} \times 1$$

3.5. Nutritional analysis

The nutritional analysis was conducted in triplicates using established A.O.A.C. methods.^{18,19} Moisture content was assessed using a hot air oven at 98 to 100°C, protein content by the Micro Kjeldhal method for total nitrogen, ash percentage through high-temperature incineration(600°C) in a muffle furnace, and fat content estimated using the Soxhlet apparatus.²⁰ Additionally, crude fiber content was evaluated employing a crude fiber analyser.²¹ The computation of carbohydrate content involved deducting the sum of moisture, protein, fat, and ash content from 100 per 100g of the sample. The energy values were calculated using the formula: Energy value = [(Protein × 4) + (Carbohydrate × 4) + (Fat × 9)](14). Additionally, the mineral analysis for iron and phosphorus utilized Atomic Absorption Spectrometry (AAS) due to its acknowledged accuracy and precision.[22]

3.6. Statistical analysis

The physical and the functional characteristics of germinated barnyard millet was performed in triplicates and the mean values were computed by applying Holm sidak method of statistical analysis where, the obtained experimental values are mean ± SD(n=3) *p value < 0.05.²²

4. Results and Discussion

4.1. Physical properties of germinated barnyard millet

Barnyard millets were germinated for 12, 24, 36, 48, 60 and 72 hours. Among all variations the germination was seen maximum at 48 hours with 12 hours of soaking. After 60 hours, the germinated grains became slimy in appearance with foul smell which was inappropriate for the study; hence the germination was terminated at 72 hours.

4.2. Length - width ratio(mm and sprout length (mm))

The physical parameters of germinated barnyard millet flour are summarized in(Table 1); showing length-width ratio ranging from 1.03 to 1.21 mm and sprout lengths varying between 1.5 to 15 mm. There was an increase in the

length and width of the millet grains as the germination time increased, this may due to the swelling of starch granules during soaking as water migrates to grains during soaking and leads to irreversible swelling.²³ One of the reasons behind increase in length might be the adherence of dried epicotyl and hypocotyl to grain after drying as their complete removal is not possible. Similar observations were reported by Nout and Davies (1982) in finger millet, Singh and Bains(1983) in wheat, Pawar and Pawar (1997) in foxtail millet, Nirmala et al. (2000) in finger millet and Suhasini et al. (2004) in wheat varieties.^{24–27}

4.3. Kernel weight (TKW and 1000 kernel volume (TKV))

Notably, the germinated barnyard millet demonstrated a slight increase in TKW (2.65 to 2.76 g) and TKV(2.30 to 2.40 ml); might be due to increase in moisture content of the samples.

Similar results were obtained by Hadimani and Malleshi (1995) in pearl millet, Singh and Goswami(1996) in cumin seeds, Kumari and Srivastava (2000) in finger millet, Shashi, B.K(2005) in finger millet and Balasubramanian and Viswanathan(2010) in minor millets.^{28–31}

4.4. Bulk density(g/ml)

The bulk density of the germinated grains went on decreasing(0.68 to 0.40 g/ml) with the increase in germination time; this was due to the breakdown of complex form of nutrients like protein and starch into simpler units which reduces the grain weight and volume due to germination.³² Hence, there was a simultaneous drop of bulk density was found with elevating duration of germination. Density is dependent on surface properties like surface area and sample volume and an increase in these parameters resulted in decrease in density.

Similar results were reported by Nefale & Mashau,(2018) in finger millet and Onwurafor et al.(2020) in mung bean, whereas, Onimawo and Asugo (2004) in pigeon pea, found increased bulk density by 8.7% after germination of melon seeds.^{33–35}

4.5. Functional properties of germinated barnyard millet flour

4.5.1. Oil absorption capacity(OAC)

The Functional properties of germinated barnyard millet flour are summarized in(Table 2);

The Oil Absorption Capacity(OAC) of germinated flour samples exhibited a notable increase with prolonged germination time, ranging from 1.14 to 1.52 ml/g. This increase in OAC is attributed to the rise in lipophilic content during grain germination, potentially linked to alterations in protein quality and the process of protein dissociation

Table 1: Physical properties of germinated barnyard millet

No. of hours of germination	Length-width Ratio (mm)	Sprout length(mm)	1000 kernel weight(g)	1000 kernel volume (ml)	Bulk density (g/ml)
0	1.03±0.04	00.00	2.65±0.04	2.30±0.05	0.68±0.07*
12	1.05±0.02	00.00	2.66±0.10	2.31±0.02	0.65±0.01
24	1.08±0.07	1.50±0.04	2.68±0.02	2.32±0.08	0.59±0.04
36	1.10±0.01	5.50±0.08	2.70±0.07	2.35±0.04	0.52±0.05
48	1.16±0.04	9.00±0.01	2.73±0.02	2.37±0.01	0.44±0.09
60	1.19±0.07	10.50±0.03	2.75±0.01	2.39±0.02	0.42±0.06
72	1.21±0.08*	15.0±0.05*	2.76±0.02*	2.40±0.03*	0.40±0.02

[Values are mean ± SD (n=3) *p value < 0.05 (holm sidak method)]

and denaturation. These mechanisms may expose the polar amino acids of millet protein, increasing hydrophobicity and ultimately enhancing OAC in processed flours.^{36,37}

Similar results were indicated by Kumar et al. (2021) in the germination of finger millet. Nefale and Mashau(2018) also reported oil absorption capacity of 163% for un-germinated finger millet flours, and 178% for 72-h germinated finger millet flours.^{33,36}

4.5.2. Water absorption capacity(WAC)

WAC also increased from 1.28 to 1.50 ml/g. This increase in WAC can be attributed to the increase in damaged starch and surface area. Damaged starch is more hygroscopic than native starch and hence absorbs more water.³⁸ WAC represents the volume occupied by the starch after swelling in excess water, which maintains the integrity of starch in aqueous dispersion. Siddiqua et al. reported that higher water absorption capacity helps to improve the softness, bulkiness, and consistency of products.³⁹

Similar results were reported by Adeniyi and Obatolu(2014) in the germination of Amaranthus grains. These results are in line with the report of Kumar et al. (2021) in finger millet and Ocheme and Chinma (2008) in pearl millet flour.^{36,40,41}

4.5.3. Water solubility index(WSI)

WSI values increased from 2.59 to 6.23 %, likely resulting from enzymatic action on millet granules, leading to the production of lower molecular weight compounds and heightened hygroscopicity, indicative of increased solubility and digestibility in germinated millets.

A similar observation was conducted by Kumar et al.(2021) who reported that water solubility index increased significantly with increasing germination time. WSI increased due to the starch hydrolysis and increased sugar level during germination.³⁶

4.5.4. Swelling power(SP)

The swelling Power of barnyard millet decreased slightly, ranging from 5.31 to 4.23 g/g, possibly due to prolonged germination. This reduction may stem from the breakdown of nutrient reserves such as starch and protein into soluble

sugars by internal enzymes, supporting sprout growth during germination.

The decrease in swelling power of millet flour is in line with the report of Nazni P, Devi SR (2016) in barnyard and foxtail millet, Nefale and Mashau (2018) in in finger millet. On the other hand, the findings of Ocheme and Chinma(2008) in pearl millet flour found that swelling power increased as germination time increased because fat content dropped during germination, reducing the swelling power of the flour by forming a complex with starch as shown by Horstmann et al.(2017).^{33,38,41,42}

4.6. Nutritional Composition of Germinated Barnyard Millet Flour

The nutritional composition of germinated barnyard millet flour is summarized in(Table 3);

- 1. Moisture:** The moisture content of grains was $\leq 10\%$ recommended as a safe limit, for extended preservation of flours. The moisture content slightly increased at 24 hour germination, and then decreased as germination time increases. It may be due to hydration of millet seeds during soaking and germination. The result obtained in this study is in line with the results obtained by Aserse Yenasew & Kelebessa Urga (2022) in finger millet varieties; Banusha and Vasantharuba (2013) where the moisture content of finger millet increased during malting. Onwurafor et al.(2020) reported similar observation during malting of Mungbean grain. Obadina et al.(2017) also reported the increment of the moisture content of pearl millet with the increase in germination periods. The mean moisture content increased significantly after germination same as in case of sorghum reported by Warle BM (2015).^{34,43–46}
- 2. Fat:** The fat content decreased as the germination time increases from 4.5 to 2.0 g/100g. The significant reduction in fat content could be due to the increased activity of lipase enzymes and severed as an energy source during germination. The fat content decrease in germinated flour might increase the shelf life by decreasing rancidity, which is most likely due to enzymes released in the flour. Similar observation

Table 2: Functional properties of germinated barnyard millet flour (Values are mean \pm SD (n=3))

No. of hours of germination	OAC(ml / g)	WAC(ml / g)	WSI(%)	SP (g/g)
0	1.28 \pm 0.01	1.28 \pm 0.05	2.61 \pm 0.02	5.31 \pm 0.05*
12	1.14 \pm 0.03	1.29 \pm 0.01	2.59 \pm 0.01	5.20 \pm 0.08
24	1.16 \pm 0.02	1.32 \pm 0.04	3.81 \pm 0.03	5.01 \pm 0.05
36	1.19 \pm 0.01	1.35 \pm 0.07	4.02 \pm 0.02	4.82 \pm 0.07
48	1.22 \pm 0.01	1.42 \pm 0.02	5.92 \pm 0.06	4.51 \pm 0.01
60	1.49 \pm 0.06	1.48 \pm 0.01	6.02 \pm 0.05	4.40 \pm 0.04
72	1.52 \pm 0.03*	1.50 \pm 0.01*	6.23 \pm 0.03*	4.23 \pm 0.05

*p value < 0.05 (holm sidak method)(OAC: oil absorption capacity, WAC: water absorption capacity, WSI: water solubility index, SP- swelling power)

Table 3: Nutritional composition of germinated barnyard millet flour (g/100g) (Values are mean \pm SD(n=3))

No. of hours of germination	Moisture	Fat	Protein	Ash	Fiber	Carb
0	10.0 \pm 0.12	4.5 \pm 0.24	10.5 \pm 0.14	2.3 \pm 0.08	10.5 \pm 0.12	72 \pm 0.09
12	10.1 \pm 0.11	4.1 \pm 0.18	10.6 \pm 0.18	2.3 \pm 0.12	10.7 \pm 0.15	70 \pm 0.11
24	10.5 \pm 0.23	3.8 \pm 0.13	10.8 \pm 0.17	2.2 \pm 0.15	11.5 \pm 0.03	69 \pm 0.15
36	10.2 \pm 0.15	3.1 \pm 0.11	11.0 \pm 0.11	2.1 \pm 0.20	12.1 \pm 0.09	69 \pm 0.20
48	9.8 \pm 0.11	2.5 \pm 0.23	11.2 \pm 0.22	2.0 \pm 0.16	12.5 \pm 0.13	70 \pm 0.09
60	9.3 \pm 0.25	2.2 \pm 0.18	11.5 \pm 0.09	1.9 \pm 0.09	12.8 \pm 0.19	71 \pm 0.15
72	9.0 \pm 0.30	2.0 \pm 0.14	11.9 \pm 0.10	1.8 \pm 0.15	13.0 \pm 0.08	71 \pm 0.11

*p value < 0.05 (holm sidak method)

Table 4: Mineral analysis of germinated barnyard millet flour (mg/100g)

No. of hours of germination	Iron	Phosphorous	Calcium	Potassium
0	7.59 \pm 0.06	210 \pm 0.03	24.2 \pm 0.24	215 \pm 0.08
12	9.40 \pm 0.09	219 \pm 0.18	30.4 \pm 0.26	230 \pm 0.09
24	11.9 \pm 0.12	233 \pm 0.20	39.1 \pm 0.19	253 \pm 0.11
36	13.3 \pm 0.18	245 \pm 0.15	52.3 \pm 0.12	278 \pm 0.16
48	14.0 \pm 0.07	252 \pm 0.09	60.2 \pm 0.21	296 \pm 0.09
60	14.7 \pm 0.10	255 \pm 0.11	67.4 \pm 0.18	302 \pm 0.10
72	15.2 \pm 0.06	258 \pm 0.12	70.3 \pm 0.20	305 \pm 0.11

[Values are mean \pm SD(n=3) *p value < 0.05(holm sidak method)]

was reported by Owhero et al.(2019) in finger millet and pearl millet flours. Moreover, the fat content decreased significantly(P < 0.05) in pearl millet flour as indicated by Ocheme and Chinma(2008). And by Aserse Yenasew & Kelebessa Urga (2022) in finger millet.^{41,43,47}

3. **Protein:** The protein content increases with germination time from 10.5 to 11.9 g/100g due to the activity of the protease, which degrades peptides into amino acids.⁴⁸ The result of the current research study is in line with the research finding of Aserse Yenasew & Kelebessa Urga (2022) in finger millet; Swami et al.(2013) who reported that the germination period of 8 to 24 h increased the protein content of finger millet flour from 14.7 to 17%; However, Ashwani Kumar et al.(2021) reported that the protein content of finger millet flour decreased from 6.04 % to 3.41% at 96 h germination.^{49,50}

4. **Ash:** The ash content decreased from 2.3 to 1.8 g/100g due to the removal of shoots, roots and bran layers, and also, some minerals in seeds might be used for sprouting metabolism.³⁶ Another reason for the reduction of ash content during germination might be the leaching of minerals during steeping and washing. Similar results were stated by Kumar et al.(2021) who reported the ash content of finger millet flour, which decreased from 2.27% to 1.24% in non-germinated and 96 h germination period, respectively; Malleshi, N. G. (1986) in finger millet; Megat Rusydi, M. R(2011) in legumes; Hama, F., Icard-Vernière, (2011) in pearl millet and white sorghum.^{36,51–53}

5. **Fiber :** The fiber content increased from 10.5 to 13.0 g/100g, this is due to the synthesis of structural compounds like cellulose and hemicellulose and the breakdown of starch during germination.^{41,45} Similar observations were reported by Banusha and Vasantharuba (2013), Ocheme and Chinma(2008),

Obadina et al. (2017), Auta et al. (2014) and Agbor Asuk et al.(2020) in finger millet, pearl millet, pearl millet, pearl millet and sorghum, respectively.^{41,44,45,54,55}

6. **Carbohydrate:** There is no significant difference in the carbohydrate content; slight decrease in carbohydrate during germination was due to the use of carbohydrates by the sprouts and could be due to rise and reduction of other food components such as moisture, fat, protein, ash and crude fiber during germination.⁵⁶ The result of the current study especially at 24 h germination is similar to the suggestion of Obadina et al. (2017) in pearl millet flour. This result is not in line with the report of Ocheme and Chinma(2008) in pearl millet flour at 48 h germination and also Owhero et al.(2019) in finger millet, but similar results have been reported in pearl millet at 3 days of germination. In contrary, Derbew and Moges (2017) also showed that the carbohydrate content of sorghum flour decreased at 48 h and 72 h of germination.^{41,45,47,56}

4.7. Mineral Analysis of Germinated Barnyard Millet Flour

The mineral analysis of germinated barnyard millet flour is summarized in (Table 4).

The increase in minerals may be due to reduction in anti-nutritional factors present in the millet after germination. Among all millets mineral profile was improved significantly after the germination. The results were in accordance with Nakarani et al.(2021) in finger millet; Nithyashree (2019) in small millets; Shonisani et al.(2019) in finger millet; and Kehong et al. in foxtail millet (2018).^{57–60}

5. Conclusion

Barnyard millet emerges as a highly nutritious functional food, boasting essential nutrients, antioxidants, and plant-based protein essential for a balanced diet. A study investigating the impact of germination on barnyard millet revealed significant insights into its physical and functional characteristics. Notably, germination influenced various physical properties, including length-width ratio, 1000 kernel weight, and volume, with outcomes varying according to processing duration. Germination time led to a slight increase in 1000 kernel weight, 1000 kernel volume, length-width ratio and sprout length, whereas there was decrease in bulk density. Furthermore, functional properties were affected, with increase in oil absorption capacity, water absorption capacity and water solubility index and decrease in swelling power enhancing food texture with prolonged germination. Germination also has an impact on proximate composition; there was decrease in moisture, fat and ash

content, no significant decrease in carbohydrate content, whereas there was increase in protein and fiber content and all the mineral contents like calcium, iron, phosphorous, and potassium were also increased.

In conclusion, the study underscores the nutritional significance of barnyard millet as a functional food rich in essential nutrients. Germination exerted notable effects on both physical and functional properties, highlighting the dynamic nature of the grain during processing. These findings suggest that germination not only alters the nutritional profile but also impacts the textural and functional attributes of barnyard millet, making it a versatile and valuable ingredient in food formulations.

6. Source of Funding

None.

7. Conflict of Interest

None.


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