Assessment of germination time of finger millet for value addition in functional foods

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In the present study, finger millet (*Eleusine coracana*) grains of variety VL Mandua-315 were soaked overnight and germinated $(25^\circ \pm 2^\circ C)$ for varying time intervals ranging from 12 to 96 h. Samples were drawn at every 12 h interval, dried at $50^{\circ} \pm 2^{\circ}$ C, deculmed, powdered and analysed for physicochemical composition, functional properties and mineral content. A significant increase ($P \le 0.05$) in sugars, crude fibre, ascorbic acid, antioxidant activity and water solubility index and decrease ($P \le 0.05$) in starch, protein and ash was found with increase in germination time. Fat, total phenols and tannins reached their minimal value after 48 h of germination followed by a linear increase. Water absorption index, oil absorption index, foam capacity and foam stability decreased with soaking, followed by a linear increase up to 60 h of germination. Amongst all minerals tested, calcium showed a significant increase with increase in germination time. A clustered heat map was used to depict the effect of germination time on the overall properties of finger millet.

Keywords: Clustered heat map, finger millet, functional foods, germination, physico-chemical properties, value addition.

MILLETS have always remained one of the staple foods and serve as a vital crop to ensure food security in semiarid areas¹. Earlier millets were considered as food of the low-income strata². However, recent health claims like low glycaemic index, gluten-free, high amounts of dietary fibre and minerals have placed them in the category of functional foods³. In addition, the climate-friendly attributes of millets, viz. limited or no irrigation requirements, ability to grow on low fertile soil and resistance to insects and pests make them a natural solution to keep drylands productive⁴. Millets can fulfil this dual contrivance and in this connection, finger millet, one of the highly produced millets with high nutrient content, represents tremendous scope in countries like India, China and Pakistan where increase in dry areas has caused reduction in cultivable areas.

India is one of the largest producers of finger millet with an area of 1138.2 thousand ha under its cultivation and production of 1821.9 thousand tonnes in 2015-16 (ref. 5). Finger millet is drought tolerant and grows best in saline soils, i.e. pH 5.0-8.2, which makes it a suitable crop for drylands⁴. It has excellent protein quality, wellbalanced amino acid profile and is considered one of the most nutritious grains⁶. Micronutrients (vitamins and minerals) like calcium, phosphorus, iron, thiamine, riboflavin and nicotinic acid are present in ample amounts⁷. Finger millet is rich in bioactive compounds that contribute to its antioxidant and antimicrobial properties^{3,8}. However, the presence of anti-nutrients like trypsin inhibitors, phytic acid and some of phenolic compounds limits its food value as these compounds interact with essential nutrients like protein and chelate minerals and reduce their absorption⁹. Various processing techniques like popping, roasting, germination and fermentation are known to reduce anti-nutritional factors to a safe limit.

Germination is an important and economical method to reduce the anti-nutritional factors and increase the digestibility of finger millet¹⁰. Germinated grains can be dried to a safer moisture limit and stored for further processing. Chemical composition, functional properties and mineral content of the grains alter with germination. Hence, a study of these changes is important for development of finger millet-based food products with enhanced functional and nutritional quality. In the present study, the effect of soaking and germination on the quality attributes of finger millet was studied at every 12 h interval up to 96 h.

Materials and methods

Finger millet (*Eleusine corcana*) grains of variety VL Manduaa-315 were procured from Vivekananda Parvatiya Krishi Anusandhanshala, Almora, Uttar Pradesh, India. The grains were cleaned by winnowing to separate the residual particles (husks, chaff and unmature seeds). The cleaned grains were collected and dried in a tray drier at $50 \pm 2^{\circ}$ C (Narang Scientific Works Limited) to a final moisture content of $8 \pm 0.5\%$ and stored till further use.

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The moisture content of the samples was determined by oven-drying method at 130°C for 2 h (ref. 11).

Malting loss

This is a measure of the mass of grain lost in the germination process. It is calculated by subtracting the weight of dried germinated grains from non-germinated grains. The formula for calculating malting loss is

Total malting loss (%) =

 $\frac{\left(\begin{array}{c} \text{Weight of the grains before malting} \\ \text{Weight of grains after malting} \\ \end{array}\right)}{\text{Weight of the grains before malting}} \times 100.$

Nutritional, bioactive and functional properties

Proximate analysis, viz. moisture, ash, fat and crude fibre was done according to the standard methods given by the Association of Official Analytical Chemists¹¹. Sugars were determined following the methods given by the American Association for Cereal Chemists¹². Crude protein content was estimated by micro Kjeldhal method¹² and a factor of 6.25 was used to convert nitrogen into protein. Total phenols were measured by the colorimetric method of Singleton and Rossi¹³. Antioxidant activity was measured according to the method of Brand-Williams et al.¹⁴ using DPPH (2,2-diphenyl-1-picrylhydrazyl) dye. Tannins were determined using Folin Denis reagent according to the method described by Saxena *et al.*¹⁵. Ascorbic acid was determined by colorimetric method¹⁶ and expressed as mg/100 g. Water solubility index (WSI), water absorption index (WAI) and oil absorption index (OAI) were determined using the methods described by Beuchat¹⁷. Foam capacity and foam stability were measured using the method described by Narayana and Narasinga Rao¹⁸

Mineral analysis

Minerals were determined by thermo-electron inductively coupled plasma atomic emission spectrometry (ICP-AES; model iCAP-630). One gram of dry sample was taken in a conical flask to which 10 ml of diacid (nitric acid + perchloric acid (3 : 1)) was added and left overnight. Then the mixture was digested until white fumes were observed¹⁹.

Statistical analysis

The experiments were conducted in triplicate and results represented as mean \pm standard deviation (SD) of three

replicate assays. Data were analysed using SPSS Statistics 24.0 (IBM, NY, USA) to test the variance and compared by Duncan's multiple range test. The means were considered to be statistically significant at P < 0.05. Log values of all parameters were used to generate a heat map.

Results and discussion

Malting loss

The malting loss was non-significant ($P \le 0.05$) in the first 12 h of germination. However, significant ($P \le 0.05$) malting loss was observed on further germination. The variety used in the study, i.e. V L Mandua-315 had a malting loss of 35.39% after 96 h of germination, which was the highest among all the values reported earlier. Kumar²⁰ had reported a malting loss of 30% in a finger millet variety native to Himachal Pradesh. Nirmala *et al.*²¹ reported a malting loss of 32.5% in the Indaf-15 variety of finger millet. During germination, the increased metabolic activities resulted in partial degradation of carbohydrates and thus loss of dry weight²¹. Another reason for the high malting loss in this variety might be the smaller seed size.

Effect of germination on nutritional composition

Soaking had a non-significant ($P \le 0.05$) effect on the sugar content of finger millet. However, germination time had a positive linear relationship with the sugar content. Reducing sugars increased from initial 0.86% to 10.54% and total sugars from 1.70% to 16.10% after 96 h of germination. Starch was negatively correlated with germination time and reduced from initial 62.83% to 41.19% after 96 h of germination. The increase in sugars and decrease in starch might be due to the breakdown of starch into sugars during germination. Seed germination is a complex phenomenon in which due to the activity of hydrolytic enzymes starch is broken down to sugars and other intermediates²². Evans *et al.*²³ had reported an increase in sugars on germination of barley. Similar results were documented by Nirmala et al.21, who reported an increase in reducing sugars and total sugars from 1.44% to 8.36% and 1.5% to 16% respectively, after four days of germination. Malleshi and Desikachar²⁴ reported 3.3% and 5.9% free sugars in finger millet germinated for 48 h and 96 h respectively. A decrease in starch content from 65% to 43% and 65.5% to 53.6% was reported after 96 h of germination^{21,24}. The maximum change in sugars and starch content of finger millet was found after 48 h of germination (Table 1). This might be due to high amylase activity in between 48 and 72 h of germination²¹.

Parameters \rightarrow Treatments \downarrow	Malting loss (%)	Reducing sugars (%)	Total sugars (%)	Starch (%)	Protein (%)	Fat (%)	Ash (%)	Crude fibre (%)
Non-germinated	0.00 ⁱ	$0.86\pm0.06^{\text{g}}$	$1.70\pm0.10^{\rm h}$	62.83 ± 0.65^a	6.04 ± 0.14^{a}	0.57 ± 0.02^{d}	$2.27\pm0.12^{\text{a}}$	3.74 ± 0.16^{g}
Soaked	1.03 ± 0.21^{h}	$0.77\pm0.02^{\rm g}$	$1.77\pm0.10^{\rm h}$	61.52 ± 1.46^{b}	$5.86\pm0.14^{\rm b}$	$0.56\pm0.03^{\text{cd}}$	2.09 ± 0.12^{b}	$3.84\pm0.06^{\rm fg}$
12 h	$1.10\pm0.10^{\rm h}$	$1.32\pm0.05^{\rm f}$	$2.18\pm0.12^{\text{gh}}$	$58.60\pm0.74^{\rm c}$	5.69 ± 0.07^{bc}	$0.50\pm0.02^{\text{de}}$	$2.07\pm0.06^{\text{b}}$	$3.87\pm0.08^{\rm fg}$
24 h	2.49 ± 0.09^{g}	$1.40\pm0.05^{\rm f}$	$2.42\pm0.12^{\text{g}}$	56.94 ± 0.79^{d}	5.65 ± 0.07^{bc}	0.50 ± 0.02^{de}	$2.07\pm0.06^{\rm b}$	4.11 ± 0.17^{de}
36 h	$8.52\pm0.45^{\rm f}$	$3.15\pm0.09^{\text{e}}$	$4.60\pm0.09^{\rm f}$	53.84 ± 0.40^{e}	$5.63\pm0.12^{\rm d}$	0.46 ± 0.03^{e}	$1.93\pm0.03^{\circ}$	4.31 ± 0.10^{d}
48 h	14.37 ± 0.51^{e}	3.31 ± 0.04^{e}	$5.53\pm0.10^{\text{e}}$	$51.87\pm0.45^{\rm f}$	$5.51\pm0.09^{\rm e}$	$0.41\pm0.02^{\rm f}$	1.77 ± 0.05^{d}	4.38 ± 0.28^{cd}
60 h	20.71 ± 0.65^{d}	$5.48\pm0.20^{\text{d}}$	$8.45\pm0.38^{\text{d}}$	49.33 ± 0.59^{g}	$5.10\pm0.13^{\rm f}$	0.53 ± 0.03^{d}	1.71 ± 0.03^{de}	4.65 ± 0.15^{bc}
72 h	$26.03 \pm 0.40^{\circ}$	$7.39\pm0.17^{\rm c}$	$11.95 \pm 0.64^{\circ}$	47.77 ± 0.26^{h}	$4.40\pm0.10^{\text{g}}$	$0.63\pm0.03^{\rm c}$	$1.63\pm0.05^{\rm e}$	4.76 ± 0.23^{b}
84 h	31.53 ± 0.15^{b}	$9.53\pm0.23^{\text{b}}$	$14.43\pm0.58^{\text{b}}$	43.59 ± 0.90^{i}	$3.61\pm0.08^{\rm h}$	$0.76\pm0.04^{\text{b}}$	$1.50\pm0.08^{\rm f}$	4.94 ± 0.31^{ab}
96 h	35.27 ± 0.40^{a}	10.54 ± 0.11^{a}	16.10 ± 0.39^{a}	41.19 ± 0.70^{j}	3.41 ± 0.05^{i}	0.85 ± 0.03^{a}	1.24 ± 0.05^{g}	5.09 ± 0.17^{a}

Table 1. Effect of germination time on physico-chemical parameters of finger millet

Values are means \pm SD. Values with the same superscript in a row are not significantly different at $P \leq 0.05$.

The protein content was reduced to 3.41% from initial 6.04% after 96 h of germination. These results are in agreement with other reported studies^{25,26}. The decrease in protein content during germination might be due to the outpacing of protein synthesis by proteolysis, and hence, an increase in free amino acid content (lysine, tryptophan, methionine and cysteine) and migration of seed nitrogenous matter to the growing embryo²⁷. However, some researchers reported increase in protein content on germination²⁸ and considered synthesis of enzyme proteins, imbibition and degradation of other constituents as the major reasons for increase in protein content.

The crude fat content decreased from 0.57% to 0.41% after 48 h of germination followed by a gradual increase and was found to be 0.85% in samples germinated for 96 h. A decrease in crude fat content of finger millet was reported by Banusha and Vasantharuba²⁵ after 36 h of germination. Lee et al.²⁹ reported a 10% decrease in crude fat of buckwheat after seven days of germination. Megat Rusydi et al.²⁷ found significant decrease in crude fat of 72 h germinated soybean, peanut and rice (white, black and brown). A decrease of 30-38% fat content was reported for three cultivars of sorghum germinated for 72 h (ref. 30). The decrease in fat with increase in germination might be due to oxidation of fat to fatty acids and water, and use as a carbon source^{27,31}. On the contrary, Obizoba and Atii³² reported 100% increase in fat content of sorghum germinated for 96 h. Elkhier and Hamid³³ reported an increase of 78% and 42% of fat in two sorghum cultivars. The increase in fat content on prolonged germination might be due to the replacement of sugars as energy source, depletion of carbohydrates and hence increase in the percentage of fat present in the grains.

Germination above 24 h resulted in a significant increase in crude fibre. The crude fibre increased from initial 3.74% to 5.09% after 96 h of germination. A similar increase of crude fibre was reported by Malleshi and Klopfenstein³⁴ in 96 h germinated finger millet. The increase in crude fibre content might be due to rapid loss of starchy components. Parts of roots and shoots that

remain adhered to millet grains after deculming can also increase the crude fibre content.

Germination had a negative effect on ash content, which reduced from initial 2.27% to 1.24% post 96 h of germination. The lower ash content of germinated grains might be due to the use of some of minerals in seeds sprouting metabolism³⁵. The loss of the bran layer due to abrasion in the process of removal of roots and shoots of germinated grains might also be responsible for the loss of minerals³⁶. Other studies have reported similar results^{24,27,34}.

Effect of germination on bioactive compounds

Table 2 depicts the effect of germination time on bioactive compounds such as total phenols, ascorbic acid, tannins and antioxidant activity. Polyphenols play an important role in maintaining body functions and health⁴. The major polyphenols in cereals are phenolic acids and tannins with a small amount of flavonoids³. In this study, it was found that the total phenol content in nongerminated finger millet grains was 1.50%, which decreased to 0.49% after 24 h of germination. However, prolonged germination time increased the phenol content and it was found to be 1.13% after 96 h of germination. A similar trend was observed in tannins and after an initial decrease, a gradual increase in tannin content was found with increase in germination time (Table 2). Decrease in phenols and tannins on germination can be attributed to leaching of polyphenols in soaking water and increased enzymatic action during germination¹⁰. However, an increase in polyphenols on prolonged germination might be due to the increase in the total percentage of seed coat in later stages of germination because of the loss of carbohydrates. The phenols and tannins are generally concentrated in the seed coat (pericarp and aleurone layer) of the grains³⁷. The ascorbic acid content of finger millet increased during germination from 9.76 to 17.40 mg/100 g after 96 h of germination. The increase in ascorbic acid might be due to vegetative growth which is

Parameters \rightarrow Treatments \downarrow	Total phenols (% as gallic acid equivalent)	Ascorbic acid (mg/100 g)	Tannins (mg/100 g as tannic acid)	Antioxidant activity (%)
Non-germinated	1.50 ± 0.05^{a}	9.76 ± 0.24^{e}	173.7 ± 11.5^{a}	72.22 ± 1.93^{e}
Soaked	0.80 ± 0.15^{d}	$8.40\pm0.13^{\rm f}$	133.0 ± 8.5^{b}	$67.77 \pm 1.92^{\rm f}$
12 h	0.67 ± 0.005^{de}	7.48 ± 0.12^{g}	121.1 ± 2.8^{b}	57.77 ± 1.92^{g}
24 h	0.49 ± 0.06^{g}	9.99 ± 0.20^{e}	90.0 ± 5.6^{cd}	74.44 ± 1.92^{de}
36 h	$0.57 \pm 0.08^{\rm ef}$	11.67 ± 0.22^{d}	66.3 ± 5.1^{f}	77.77 ± 1.92^{cd}
48 h	0.65 ± 0.05^{de}	12.01 ± 0.45^{d}	$64.3 \pm 9.7^{\rm f}$	78.88 ± 1.92^{bc}
60 h	0.83 ± 0.10^{cd}	$16.53 \pm 0.31^{\circ}$	$75.2 \pm 8.3^{\rm ef}$	82.22 ± 1.93^{ab}
72 h	$1.00 \pm 0.03^{\rm bc}$	16.91 ± 0.30^{bc}	$78.5 \pm 5.93^{\rm ef}$	82.22 ± 1.93^{ab}
84 h	1.06 ± 0.13^{b}	17.21 ± 0.29^{b}	85.2 ± 10.00^{cd}	83.33 ± 3.34^a
96 h	1.13 ± 0.10^{b}	$17.40\pm0.17^{\text{a}}$	$94.66 \pm 12.00^{\circ}$	84.44 ± 1.92^a

Table 2. Effect of germination time on bioactive compounds of finger millet

Values are means \pm SD. Values with the same superscript in a row are not significantly different at $P \leq 0.05$.

Table 3. Effect of germination time on the functional properties of finger millet

Parameters \rightarrow Treatments \downarrow	WSI (%)	WAI (ml/g)	OAI (ml/g)	Foam capacity (%)	Foam stability (%)
Non-germinated	$3.64\pm0.04^{\rm f}$	$1.33\pm0.03^{\text{cd}}$	$2.22\pm0.02^{\rm g}$	$12.33\pm0.58^{\text{b}}$	51.00 ± 1.00^{cd}
Soaked	$3.36\pm0.04^{\text{g}}$	1.27 ± 0.05^{de}	$2.12\pm0.03^{\rm h}$	$11.67 \pm 0.58^{\circ}$	49.33 ± 1.15^{d}
12 h	3.39 ± 0.10^{g}	1.28 ± 0.04^{de}	$2.26\pm0.04^{\rm f}$	13.00 ± 1.00^{b}	70.33 ± 0.58^{a}
24 h	$3.65\pm0.06^{\rm f}$	1.30 ± 0.02^{de}	2.36 ± 0.02^{cd}	15.33 ± 0.78^a	72.00 ± 1.00^{a}
36 h	6.05 ± 0.28^{b}	$1.31 \pm 0.03^{\circ}$	$2.39\pm0.03^{\rm c}$	15.33 ± 0.58^a	60.67 ± 1.15^{b}
48 h	6.43 ± 0.06^{d}	1.33 ± 0.03^{bcd}	$2.45\pm0.04^{\text{b}}$	$11.67 \pm 0.58^{\circ}$	50.67 ± 1.15^{cd}
60 h	$8.47 \pm 0.06^{\circ}$	1.45 ± 0.04^{a}	2.52 ± 0.02^{a}	$11.33 \pm 0.58^{\circ}$	42.00 ± 2.00^{e}
72 h	$8.56 \pm 0.04^{\circ}$	1.39 ± 0.03^{b}	2.35 ± 0.01^{de}	$9.67\pm0.58^{\rm d}$	40.67 ± 1.15^{e}
84 h	14.76 ± 0.06^{b}	$1.37 \pm 0.01^{\rm bc}$	2.37 ± 0.02^{cd}	9.00 ± 1.00^{d}	51.33 ± 1.15^{cd}
96 h	15.73 ± 0.09^a	$1.32\pm0.01^{\text{cde}}$	2.31 ± 0.02^{e}	7.33 ± 0.58^{e}	$53.33\pm3.06^{\circ}$

WAI, Water absorption index; WSI, Water solubility index and OAI, Oil absorption index. Values are means \pm SD. Values with the same superscript in a row are not significantly different at $P \le 0.05$.

known to enhance ascorbic acid content. An increase in ascorbic acid has also been reported by Malleshi and Desikachar²⁵ during germination of finger millet. Antioxidant activity also get enhanced with germination time. This may be attributed to increase in ascorbic acid content during germination.

Effect of germination on functional properties

Functional properties of grains are of great significance in product development. These properties get changed during germination and hence their study is crucial for product development. WAI and OAI are important parameters in pasta and macaroni³⁷. WSI is important for the development of cereal-based drinks. The changes in functional properties are mainly due to changes in composition and structure of carbohydrates and proteins³⁸. Germination has also been reported to decrease the amylose content of starch³⁹ and reduce the proteins of high molecular weight⁴⁰. Table 3 presents the changes in functional properties of finger millet during 96 h of germination. WSI increased significantly with increase in germination time and was found to be 15.73% after 96 h of germination. The increase in WSI might be due to increase in sugar content (Table 1) and breakdown of starch

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into smaller granules. WAI and OAI increased up to 60 h of germination followed by a linear decrease. This increase in WAI and OAI can be attributed to the increase in damaged starch and surface area. Damaged starch is more hygroscopic than native starch and hence absorbs more water⁴¹. Similar results were reported by Adeniyi and Obatolu⁴² in the germination of Amaranthus grains. The decrease in WAI and OAI after 60 h of germination might be due to the breakdown of starch to sugars and reduction in starch content.

Foam capacity and foam stability depend upon several factors like protein concentration, salts, sugars, lipids, temperature and pH^{43} . Foam capacity increased up to 36 h of germination followed by a significant decrease. A similar trend was observed in foam stability and with increase in germination time the stability of foam also increased, which might be due to the increased concentration of sugars and salts. A decrease in protein content might be responsible for loss of foaming properties (Table 1).

Effect of germination on mineral content

The mineral content in non-germinated samples was within the range reported earlier⁴. All minerals, except

	Non-germinated	Soaked	12 h	24 h	36 h	48 h	60 h	72 h	84 h	96 h
Macro minerals Calcium Phosphorus Potassium Magnesium Sulphur Sodium	$\begin{array}{c} 193.5 \pm 0.9^{c} \\ 219.7 \pm 0.6^{f} \\ 520.3 \pm 1.12^{h} \\ 102.90 \pm 0.69^{e} \\ 103.44 \pm 0.68^{g} \\ 38.94 \pm 0.53^{g} \end{array}$	$\begin{array}{c} 175.5 \pm 3.9^{a} \\ 214.6 \pm 1.1^{e} \\ 340.0 \pm 0.66^{8} \\ 96.33 \pm 0.55^{ab} \\ 87.40 \pm 0.76^{e} \\ 25.64 \pm 0.44^{f} \end{array}$	$\begin{array}{c} 175.9\pm2.5^{a}\\ 214.2\pm1.7^{c}\\ 340.4\pm0.74^{b}\\ 96.49\pm2.32^{ab}\\ 88.23\pm0.65^{c}\\ 24.76\pm0.79^{f}\end{array}$	$\begin{array}{c} 183.9\pm0.32^{b}\\ 213.6\pm0.7^{e}\\ 339.4\pm1.24^{8}\\ 96.6\pm0.95^{ab}\\ 90.02\pm0.36^{de}\\ 20.77\pm0.37^{e}\end{array}$	$\begin{array}{c} 191.9\pm0.83^{\circ}\\ 213.5\pm0.9^{\circ}\\ 328.9\pm0.87^{f}\\ 95.50\pm0.77^{a}\\ 89.72\pm0.34^{d}\\ 20.65\pm0.94^{de} \end{array}$	$\begin{array}{c} 195.1 \pm 1.8^{\circ} \\ 213.54 \pm 1.2^{\circ} \\ 317.7 \pm 0.28^{\circ} \\ 98.13 \pm 0.51^{bc} \\ 91.01 \pm 0.42^{\circ} \\ 20.57 \pm 0.79^{de} \end{array}$	216.9 ± 0.5^{d} 206.9 ± 1.2^{d} 254.8 ± 1.73^{d} 99.78 ± 0.62^{ed} 94.71 ± 0.97^{f} 19.56 ± 0.79^{d}	$\begin{array}{c} 226.4 \pm 1.6^{\circ} \\ 195.4 \pm 0.74^{\circ} \\ 242.7 \pm 0.53^{\circ} \\ 100.33 \pm 0.4^{d} \\ 82.53 \pm 0.5^{b} \\ 9.07 \pm 0.17^{\circ} \end{array}$	$\begin{array}{c} 227.8\pm0.7^{\rm e}\\ 186.2\pm3.23^{\rm b}\\ 214.01\pm1.04^{\rm b}\\ 109.91\pm0.44^{\rm f}\\ 76.43\pm0.59^{\rm a}\\ 7.18\pm0.84^{\rm b} \end{array}$	244.9 ± 1.7^{f} 157.7 ± 0.99^{a} 171.4 ± 1.05^{a} 111.37 ± 0.89^{g} 76.67 ± 0.34^{a} 4.66 ± 0.29^{a}
Manganese Micro minerals Iron Zinc Copper	$10.19 \pm 0.05^{\circ}$ $5.56 \pm 0.39^{\circ}$ $2.10 \pm 0.08e$ 0.40 ± 0.01^{d}	$\begin{array}{l} 9.25 \pm 0.04^{a} \\ 5.49 \pm 0.52^{b} \\ 2.10 \pm 0.06^{e} \\ 0.36 \pm 0.03^{be} \end{array}$	$\begin{array}{l} 9.40 \pm 0.09^{b} \\ 5.36 \pm 1.50^{b} \\ 2.09 \pm 0.05^{c} \\ 0.38 \pm 0.01^{cd} \end{array}$	$\begin{array}{l} 10.21 \pm 0.09^{c} \\ 2.88 \pm 0.39^{a} \\ 2.0 \pm 0.04^{d} \\ 0.38 \pm 0.0^{bc} \end{array}$	$\begin{array}{c} 10.42 \pm 0.07^{d} \\ 2.40 \pm 0.35^{a} \\ 1.99 \pm 0.05^{d} \\ 0.37 \pm 0.01^{bc} \end{array}$	10.68 ± 0.03^{e} 2.69 ± 0.53^{a} 1.96 ± 0.02^{ed} 0.37 ± 0.02^{bb}	10.92 ± 0.11^{f} 2.66 ± 0.56^{a} 1.90 ± 0.05^{bc} 0.35 ± 0.02^{b}	$\begin{array}{c} 11.81 \pm 0.07^{g} \\ 2.44 \pm 0.18^{a} \\ 1.83 \pm 0.02^{ab} \\ 0.31 \pm 0.01^{a} \end{array}$	12.17 ± 0.10^{h} 2.29 ± 0.23^{a} 1.81 ± 0.02^{a} 0.32 ± 0.03^{a}	12.21 ± 0.04^{h} 2.02 ± 0.05^{a} 1.75 ± 0.02^{a} 0.30 ± 0.01^{a}
Values are means	Values are means \pm SD. Values with the same superscript in a row are not significantly different at ($P \leq 0.05$)	the same supersor	ipt in a row are no	t significantly diff	erent at $(P \leq 0.05)$	-				

Table 4. Effect of germination on the mineral content of finger millet (mg/100 g)

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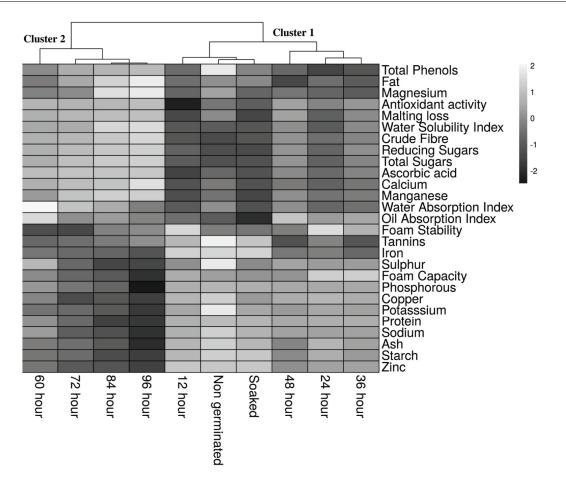


Figure 1. Clustered heat map depicting the effect of germination time on overall properties of finger millet.

zinc, showed significant loss upon soaking (Table 4). This might be due to the leaching of minerals in soaking water. The calcium content of non-germinated finger millet grains reduced after soaking followed by a linear increase with increase in germination time. These results are comparable with those of Suma and Urooj²⁶, who reported an increase in calcium content of pearl millet on germination for 72 h. The increase in calcium might be due to loss of organic dry matter from the grains during germination and hence increase in the percentage of calcium in the grains. Phosphorus showed a linear decrease with increase in germination time and a loss of 28% phosphorus was noticed after 96 h of germination. These results are in contradiction to Azeke et al.⁴⁴, who reported 6% increase in phosphorus content of three days germinated sesame seeds and 19.7% increase in phosphorus content of ten days germinated millets. Plants require phosphorus for complex energy transmission and early root formation⁴⁵, which might be the reason for the decrease in phosphorus content. A decreasing trend was found for potassium, sodium and sulphur. This decrease might be due to solubility and leaching of these minerals in soaking water. Magnesium and manganese decreased with soaking, but a linear increase in their contents was

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found with increase in the germination time. Luo *et al.*⁴⁶ reported decrease in manganese content on 48 h germinated cereals and legumes.

Microminerals like iron, zinc and copper are considered essential elements in human nutrition. Germination had a negative effect on iron, zinc and copper and there was 63.7%, 16.7% and 25% reduction in these trace elements after 96 h of germination. These findings are in agreement with those Luo and Xie⁴⁷, who reported a decrease of 38.2–38.9% iron and 24.5–29.2% of zinc on 72 h germination of faba beans. However, these results are in contrast with other studies^{26,46} which reported an increase in these trace minerals on germination.

Clustered heat map

A cluster heat map was generated using log values to show the effect of germination on the overall properties of finger millet (Figure 1). Grey scale was used for visualization of the heat map. The darker the intensity of the white colour, lesser is the content of the corresponding parameter/nutritional content, whereas darker the intensity of black colour, higher is the content. It is clear from the

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grayscale variation that minerals, like zinc, sodium, potassium, copper, phosphorus and sulphur showed more or less similar trends across variation in germination times. Hierarchical tree cluster across germination times was divided into two groups. Cluster one comprised non-germinated, soaked, 12–48 h germinated finger millet grains. The second cluster consisted of grains germinated for 60–96 h. It is evident from the heat map that the most noticeable changes took place between 12 and 24, 48 and 60, 84 and 96 h of germination.

Conclusion

Germination time had a significant effect on the physicochemical characteristics, functional attributes as well as mineral content. With increase in germination time there was an increase in sugars and crude fibre, while starch, proteins, fat and mineral content decreased. The total phenols and tannins contents decreased with germination; however, on prolonged germination (above 48 h) increase in total phenols and tannins was observed. Germination also increased ascorbic acid content, antioxidant activity and functional properties like WSI, WAI, OAI, foaming capacity and foam stability. Calcium content increased with germination while there was a decrease in other minerals, like phosphorus, potassium, magnesium, sulphur, sodium, manganese, iron, zinc and copper. On the basis of the studied parameters, a germination time above 48 h is recommended for the preparation of malted drinks that usually require higher levels of sugars and water solubility. A prolonged germination above 72 h resulted in high malting loss and increase in anti-nutrients. Hence, prolonged germination of finger millet grains should be avoided.

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