International Journal of Food Science + Technology

International Journal of Food Science and Technology, 2025, **60(2)**, vvaf132
https://doi.org/10.1093/ijfood/vvaf132
Advance access publication: 1 July 2025
Original Article

Effect of fermentation on the proximate composition, antinutrients, bioaccessibility of minerals, and sensory quality of pearl millet-based *Injera*

Tadesse Fenta Yehuala[1], Minaleshewa Atlabachew[2], Sotirios Karavoltsos[3], Aikaterini Sakellari[3], Mohamad Farshard Aslam[4], Lara Allen[5,6], Howard Griffiths[7,*], Anastasia Kanellou[8], Panagiotis Zoumpoulakis[8], Wanjiku Gichohi-Wainainah[9,10], Helen Walle Endalew[1], Metadel Kassahun Abera[1], Mesfin Wogayehu Tenagashaw[1], Gizaw Desta Gessesse[11], and Hirut Assaye Cherie[1,*]

[1]Faculty of Chemical and Food Engineering, Bahir Dar Institute of Technology, Bahir Dar University, Bahir Dar, Ethiopia
[2]Department of Analytical Chemistry, College of Science, Bahir Dar University, Bahir Dar, Ethiopia
[3]Laboratory of Environmental Chemistry, Department of Chemistry, National and Kapodistrian University of Athens, Athens, Greece
[4]Department of Nutritional Sciences, School of Life Course and Population Sciences, King's College London, London, United Kingdom
[5]Department of Chemical Engineering and Biotechnology, University of Cambridge, Cambridge, United Kingdom
[6]Centre for Global Equality, 8C King's Parade, Cambridge, CB2 1SJ, UK
[7]Department of Plant Sciences, University of Cambridge, Cambridge, United Kingdom
[8]Department of Food Science and Technology, University of West Attica, Athens, Greece
[9]World Fish, Penang, Malaysia
[10]International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Lilongwe, Malawi
[11]International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Addis Ababa, Ethiopia
*Corresponding authors: Faculty of Chemical and Food Engineering, Bahir Dar Institute of Technology, Bahir Dar University, Box 26, 6000, Ethiopia. Email: hirutas2000@gmail.com; Department of Plant Sciences, University of Cambridge, Downing Street, CB2 3EA, UK. Email: hg230@cam.ac.uk

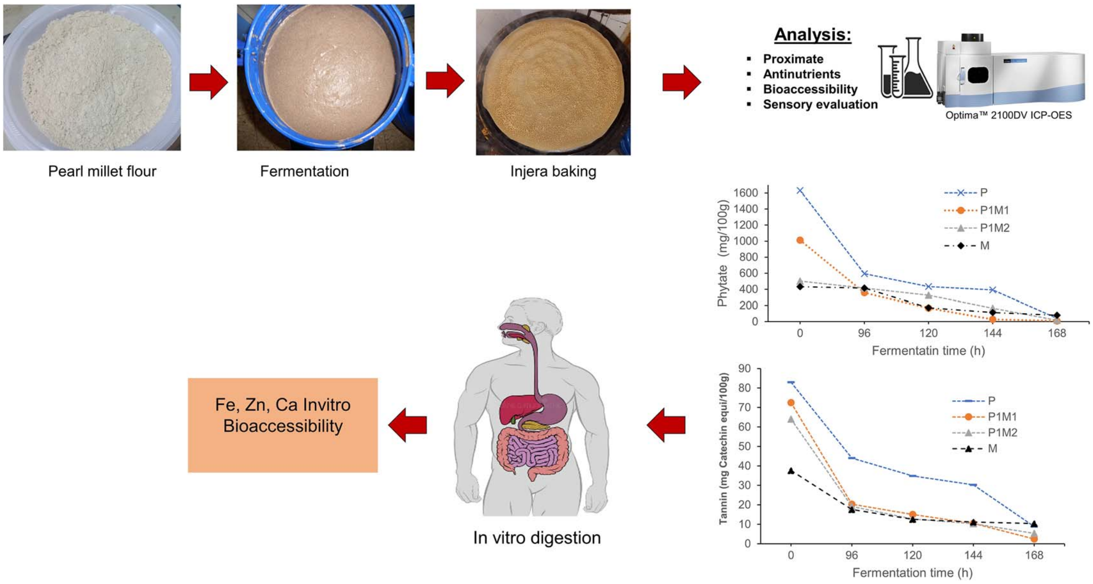
Abstract

Pearl millet is a cereal rich in both macro- and micronutrients; however, it also contains high levels of antinutrients, such as phytate, tannin, and phenols, which can hinder nutrient absorption. This study examined the impact of fermentation on the nutrient composition, antinutritional content, mineral bioaccessibility, and sensory quality of *Injera* prepared from pearl millet flour alone, as well as from a composite flour of pearl millet and maize (in 1:1 and 1:2 ratios). Fermentation significantly improved the nutrient profile and sensory attributes of *Injera* samples. Significant improvements ($p < .05$) were observed in all *Injera* samples, with reductions in phytate (81.5%–99.2%) and tannin (72.4%–96.1%) contents, and increased mineral bioaccessibility for iron (62.1%–73.5%), zinc (53.8%–83.3%), and calcium (19.6%–54.6%). These findings showed that traditional fermentation methods can effectively decrease antinutrients, enhance the nutrient profile, and improve mineral bioaccessibility in pearl millet-based *Injeras*.

Keywords: pearl millet, fermentation, *Injera*, nutrients, antinutrients, bioaccessibility, sensory acceptability

Received: 10 March 2025. **Revised:** 7 June 2025. **Accepted:** 27 June 2025

Graphical abstract



Introduction

Like many developing countries, Ethiopia relies heavily on cereal-based foods for the diet of both children and adults. These foods often contain high levels of antinutrients, mainly phytic acid (PA), which can bind essential nutrients and hinder their absorption (Lopez et al., 2002). Consequently, micronutrient deficiencies, particularly iron, calcium, and zinc are prevalent in Ethiopia (Hailu, 2016). Additionally, protein-energy malnutrition is a significant concern, with nearly 39% of children under five reported to be stunted in a recent survey (FNS, 2023).

Injera, a traditional fermented pancake-like flatbread, serves as a staple food in Ethiopia (Woldemariam et al., 2019). By consuming *Injera* at least once daily, Ethiopians get approximately 70% of their dietary calories from this single food (Ashagrie & A., 2012; Cherie et al., 2018). Teff (*Eragrostis tef*) is the preferred cereal for preparing *Injera*; however, it is also common to prepare *Injera* from other cereals, including wheat, barley, sorghum, millet, maize, and rice (Mihrete & Bultosa, 2017; Neela & Fanta, 2020).

Pearl millet is a cereal rich in both macro- and micronutrients. Its protein content ranges from 9% to 24% (Ali et al., 2003), while its fat content varies from 4% to 9% (Jain & Bal, 1997). Additionally, pearl millet is a good source of micronutrients such as folate, copper, zinc, iron, magnesium, calcium, and various B vitamins, along with unsaturated fatty acids (Dayakar et al., 2018). Unlike teff, which requires fertile soil and significant precipitation for growth, pearl millet can thrive in range of soils and harsh environmental conditions (Dias-Martins et al., 2018). An improved variety of pearl millet, known as Kola-1, has been introduced in Ethiopia, expanding its cultivation to drought-prone areas (Lakew & Berhanu, 2019; Erenso Degu et al., 2009).

Despite its advantageous properties, pearl millet also contains antinutrients, including PA, tannins, phenols, and α-galactosides, which can inhibit nutrient absorption (Rani et al., 2018). Fermentation has been identified as an effective method for reducing antinutritional factors in cereals (Samtiya et al., 2020). This process activates enzymes that break down these compounds, thereby enhancing the nutritional profile by improving mineral bioavailability and the digestibility of proteins and carbohydrates (Adebo et al., 2022; Osman, 2011). Furthermore, fermentation enhances the palatability and organoleptic properties of foods (Şanlier et al., 2019).

The bioaccessibility of problem nutrients in Ethiopia, such as iron and zinc, from *Injera* samples prepared from different flour blends, such as teff–white sorghum, barley–wheat, and wheat–red sorghum, was reported by Baye et al. (2014). However, studies on mineral bioaccessibility and impacts of fermentation on other attributes such as proximate composition and antinutritional levels of pearl millet and pearl millet-based *Injeras* are limited. This study thus aimed to examine the effects of fermentation on the proximate composition, levels of antinutrients, bioaccessibility of selected minerals, and sensorial quality of pearl millet and pearl millet-based *Injeras*.

Materials and methods

Focus group discussion for recipe selection

Injera preparation recipes were identified and adapted after a focus group discussion with women from the Dangeshita sub-district (*Kebele*), Dangila district, which is located approximately 90 km southwest of Bahir Dar City, Ethiopia. These communities are known as both producers and consumers of millet. Twelve randomly selected women from the community were involved in a focus group discussion and were asked about how they prepare *Injera* from finger millet. Specifically, they were asked if they have a practice of blending millet with other crops in the *Injera* preparation process, duration of fermentation, use of back slopping, and so on.

Focus group discussants elaborated the practice of mixing millet with maize in different proportions based on availability of millet. In cases of high availability of millet, *Injera* is either prepared exclusively from millet or mixing millet and maize in a one-to-one proportion (1-part millet:1-part maize flour, P1M1) whereas, in cases where there is less access, millet is mixed with maize in a one-to-two proportion (1-part millet:2 parts maize flour, P1M2). Three *Injera* preparation recipes were identified: finger millet flour alone, finger millet and maize flours in a one-to-one (1 finger millet:1 maize) proportion, and finger millet and maize flours in a one-to-two ratio (1 finger millet:2 maize)

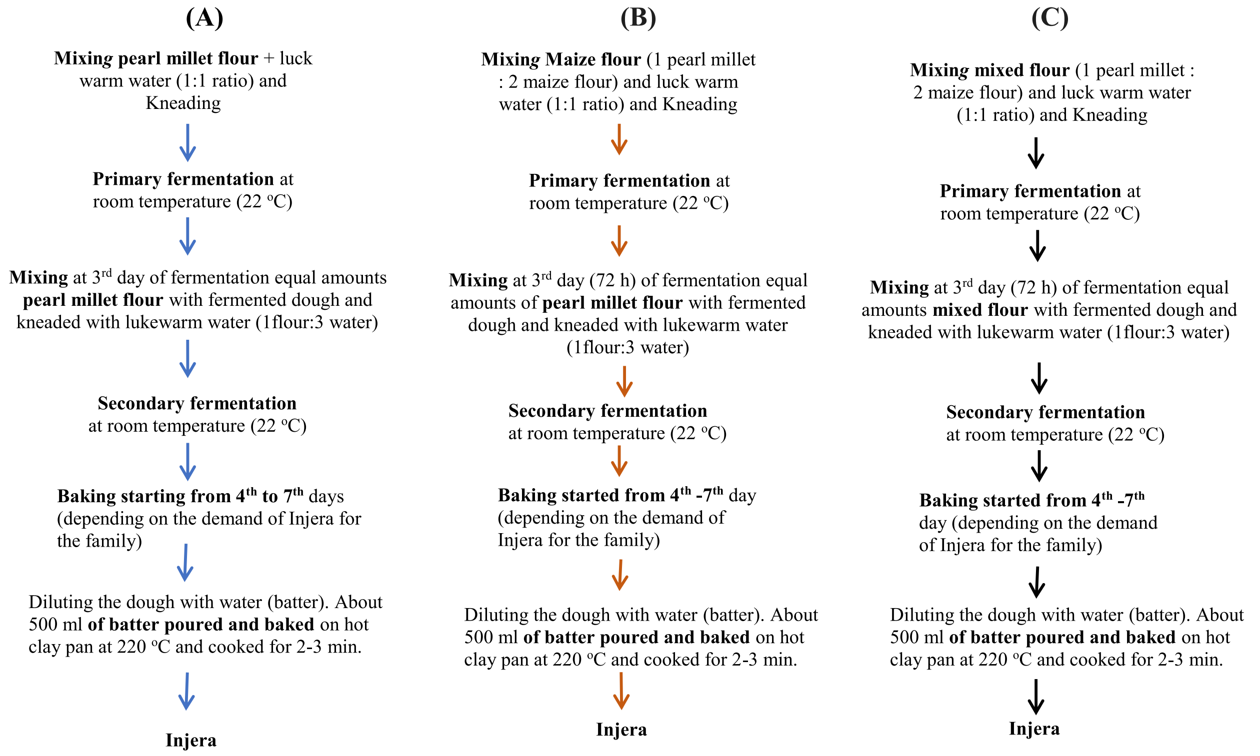


Figure 1. Flow diagram of *Injera* preparation: (A) from pearl millet (P), (B) from pearl millet and maize flour (1 pearl millet:1 maize, P1M1), and (C) from pearl millet and maize flour (1 pearl millet:2 maize, P1M2).

proportion. Differences were also recorded at the mixing stage of the two cereals. For women who use the one-to-one ratio, the cereals (finger millet and maize) are ground to flour separately. Initially, preparing maize dough and subsequently adding finger millet flour after 72 hr (3 days) of fermentation. When using the 1:2 ratio, the cereals (finger millet and maize) are ground together, and the composite flour is used to prepare dough. After 72 hr of fermentation another batch of composite flour is added to the previous dough. In both cases, after 96 hr (fourth day) of fermentation, the dough can be baked and baking may continue until the 168 hr (seventh day). In our study, the *Injera* preparation process from finger millet and maize mixture by the local communities was adopted to prepare *Injera* from pearl millet and maize flours in the laboratory.

Pearl millet-based *Injera* preparation in the laboratory

Pearl millet (kola-1 variety, accession number: ICMV221) was brought from Sekota Dryland Agricultural Research Centre, Ethiopia, and maize (BH 660 variety) and maize was obtained from farmers in Dangila district, Ethiopia. The grains were cleaned and then milled individually into flour using a laboratory hammer mill (ModTPS–JXFM110, Shanghai, China) and sifted to a sieve size of 500 μm. With the assistance of women from Dangeshita kebele, the traditional recipes for *Injera* preparation as described in Figure 1A–C were applied in the Food Processing Laboratory, Bahir Dar University.

Sample preparation

Injera samples prepared following the recipes identified were spread out on aluminium foil and dried individually in hot air for 24 hr at 65 °C using an air-drying oven (DHG-9140A; Zenith Lab Inc., China). They were then milled into a fine powder, sealed in polyethylene plastic bags, and stored at −20 °C for analysis to ensure their integrity was maintained. Samples for the determination of minerals and bioaccessibility of minerals were transported under cooled conditions to Laboratory of Environmental Chemistry, University of Athens, Greece, and immediately frozen to −20 °C.

Proximate composition

The proximate composition (moisture, total ash, crude protein, crude fat, and crude fibre content) of *Injera* samples prepared from the different recipes were determined as per AOAC (2005), while total carbohydrate was calculated indirectly by difference method (Tontisirin et al., 2003). All results were expressed on a dry weight basis.

Determination of mineral content

The mineral content of samples was determined following the methods of Grigoriou et al. (2022) and Lee et al. (2022), with slight modification. The dried sample (0.1 g) was wet digested under microwave digester (Multiwave Go Plus, Anton Paar, Graz, Austria) using HNO_3 (65%). The digested samples were diluted using 20 ml of Milli-Q water, filtered, and the final volume was adjusted to 25 ml. Inductively coupled plasma optical emission spectrometry (Optima 2100DV ICP-OES, PerkinElmer, Waltham, MA, USA) was employed for determining the concentration of the selected minerals. The standard calibration solutions for ICP analyses were prepared in the ranges of 0.1–5 mg/L (Ca, Fe, and Zn), diluting a multielement standard (ICP multi-element standard IV, Merck). Emission intensities were measured at 317.9, 238.2, and 206.2 nm for Ca, Fe, and Zn, respectively. Limits of detection (LODs) for Ca, Fe, and Zn were calculated using the formula 3.3 × S, where S is standard deviation of 10 replicates samples prepared at low concentrations (Long & Winefordner, 1983), equal to 0.140 μg g^{-1} of dry weight. For quality assurance purposes, at least one blank was included in each sample batch and certified reference material BCR191 (brown-red) was analysed for the three metals (iron [Fe], zinc [Zn], and calcium [Ca]) providing a recovery between 90% and 110%.

Determination of mineral bioaccessibility

The bioaccessibility of calcium (Ca), iron (Fe), and zinc (Zn) from *Injera* samples was assessed using a simulated gastrointestinal digestive method developed by Glahn et al. (1998) and Khoja et al. (2021) with minor modifications. About 1 g of sample was mixed with 10 ml of saline solution (140 mmol/L NaCl and 5 mmol/L KCl) and left for 15 min. The pH was adjusted to 2.0 with 1 M HCl. Then, 1 ml pepsin (Sigma-P-7012) (9.6 mg ml^{-1}) was added, and the solution was vortexed. After 90 min incubation at 37 °C in a shaking water bath at 150 rpm, the pH was adjusted to 7 using 1 M NaOH. A solution of 2.5 ml from bile extract (Sigma-B8631) and pancreatin (Sigma-P7545) (8.5 mg ml^{-1} bile extract and 1.4 mg ml^{-1} pancreatin) were added. The solution was diluted to a final volume of 18 ml with saline solution and incubated at 37 °C for 90 min before being centrifuged (at 1,000 rpm for 10 min) using a centrifuge (Sigma 3-18KS, Germany). Then after, the supernatant was decanted; 1 ml was transferred into a 15 ml tube. The supernatant (1 ml) of each sample was digested with 65% concentrated pure HNO_3 acid in an oven at 65 °C for 12 hr. The digested samples were diluted by de-ionised water and Ca, Fe, and Zn contents were measured using ICP-OES (Optima 2100DV, ICP-OES, PerkinElmer, Waltham, MA, USA).

Determination of phytate concentration

The phytate concentration of the samples was determined using McKie and McCleary (2016) method, employing a Megazyme assay kit (Megazyme-KPHYT, Bray, Ireland). Samples weighing 1 g were placed in a 75 ml glass beaker, 20 ml of 0.66 M HCl was added, and the beaker was covered with foil and stirred vigorously for at least 3 hr. Afterward, 1 ml of the extract was transferred to a 1.5 ml microfuge tube and centrifuged for 10 min. From the resulting supernatant, 0.5 ml was transferred to a new 1.5 ml microfuge tube, and 0.5 ml of 0.75 M NaOH was added. The phosphorus releases were measured using the colorimetric method (G6860A, Agilent, Malaysia) and the absorbance was recorded at 655 nm against an appropriate blank. Phytate content is reported in g of PA per 100 g of sample.

Determination of tannin content

A modified Vanillin–HCl Assay was used to determine the tannin content as described by the method of Dykes (2019). Three replicates of 0.3 g each of dried sample were measured and transferred into centrifuge tubes and 8 ml of 1% HCl in methanol were added to each tube. Then, the contents were mixed in a vortex mixer for 10 s, each tube remained in a water bath for 20 min for incubation and was afterward mixed on the vortex mixer for 10 s. Then, the samples were centrifuged at 4,000 × *g* (Sigma 3-18KS, Germany) for 10 min. The upper layer was taken by decantation. From the decanted solution an aliquot were taken and placed into separate two test tubes with 1 ml each. One tube was labelled as the "sample" tube and the other as the "blank" tube. All tubes were placed into the water bath (30 °C). The vanillin reagent of 5 ml was added to each "sample" tube in each pair of samples and incubated for 20 min. After that 5 ml of 4% HCl in methanol were added to the "blank" tubes in each pair at 15-s intervals. The absorbance of each "sample" and "blank" tube was measured after 20 min at 500 nm using a spectrophotometer (G6860A, Agilent, Malaysia). For the final determination of the tannin concentration, the value of the "blank" from that of the "sample" was subtracted. Tannin concentration was calculated as mg CE/100 g.

Sensory quality of *Injera*

The sensory attributes of *Injera* samples prepared from pearl millet flour alone and pearl millet and maize composite flours was evaluated according to the method proposed by Lim et al. (2022), using a seven-point hedonic scale. A total of 37 untrained panellists (staff and students from human nutrition, food engineering, and chemical engineering programmes) were chosen randomly from Bahir Dar University. The sensory attributes considered were colour, texture (softness degree), eye uniformity, top and bottom surface (stickiness), rollability, taste, and overall acceptability. A glass of water was offered to the panellists, and they were informed to drink water between tastings (Mutshinyani et al., 2020).

Statistical analysis

Data analysis was performed using SPSS version 26 (SPSS, Chicago, IL, USA). Results were presented as $M \pm SD$. Means were compared using ANOVA (analysis of variance) and Tukey's multiple comparison test to establish where significant difference between treatments at a 95% level of confidence exist.

Results and discussion

Effect of fermentation on the nutritional values of pearl millet-based *Injera*

Table 1 shows the nutritional values of pearl millet-based *Injera* samples. The total ash content of the *Injera* samples ranged from 1.46% to 2.04%. Inyang and Zakari (2008) also reported ash levels of 1.87%–2.72% for instant Fura, a Nigerian cereal fermented food prepared from pearl millet and sorghum. *Injera* samples had a higher protein content compared to their respective unfermented flour samples. The crude protein levels of prepared *Injera* samples from P1M1 flour increased by 34%, while that prepared from P1M2 flour increased by 20.5% compared to their unfermented flours. Even though no significant differences were reported among *Injera* samples, the highest protein content (12.8%) was recorded for *Injera* of pearl millet (P) fermented for 144 hr, and the lowest (8.94%) was recorded for *Injera* from maize (M) fermented for 168 hr. The increase in the protein levels in the *Injera* samples might be associated to the synthesis of enzymes and the formation of new proteins during fermentation, and the release of proteins from the breakdown of antinutritional components (Endalew et al., 2024). The protein level of *Injera* from pearl millet (P) and maize (M) showed a slightly decreasing trend between 144 and 168 hr of fermentation, attributed to multiple factors that can exert an influence on the dynamics of protein during fermentation, including microbial activity, enzymatic reactions, and availability of substrates (Diether & Willing, 2019). The crude protein composition of *Injera* samples recorded in this study was within the range (6.8%–18.2%) reported for other pearl millet-based food products like ready to eat foods, different types of porridges, steamed cooked products, and fermented blended flour (Onyango et al., 2021; Saleh et al., 2013).

Statistically significant differences ($p < .05$) were observed in the fat content of flour samples at initial time of fermentation and their corresponding *Injera* samples. *Injera* samples' fat content decreased during fermentation, probably due to increased activities of lipolytic enzymes, which hydrolyse fats into fatty acids

Table 1. Effect of fermentation on nutritional values (g/100 g d.w.) of *Injera* from pearl millet and pearl millet-maize composite flour.

Fermentation time (hr)	Total ash (%)	Crude protein (%)	Crude fat (%)	Crude fibre (%)	Total CHOs (%)	Gross energy (kcal/100 gm)
Pearl millet (P) *Injera*						
0	1.61 ± 0.01^{ab}	11.1 ± 0.1^{a}	5.28 ± 0.08^{b}	4.60 ± 0.04^{c}	82.1 ± 0.1^{b}	419 ± 1^{c}
96	1.46 ± 0.19^{a}	12.2 ± 0.2^{a}	4.62 ± 0.44^{ab}	4.22 ± 0.10^{cd}	81.3 ± 0.2^{b}	415 ± 3^{ab}
120	1.81 ± 0.01^{b}	12.3 ± 0.2^{a}	5.32 ± 0.29^{b}	4.19 ± 0.36^{e}	80.4 ± 0.4^{ab}	418 ± 1^{bc}
144	1.68 ± 0.13^{ab}	12.8 ± 1.2^{a}	4.13 ± 0.28^{a}	2.20 ± 0.09^{a}	81.1 ± 1.4^{ab}	412 ± 1^{a}
168	1.78 ± 0.05^{b}	11.6 ± 0.2^{a}	6.90 ± 0.13^{c}	2.16 ± 0.01^{d}	79.5 ± 0.2^{a}	426 ± 1^{d}
1 Pearl millet:1 Maize (P1M1) *Injera*						
0	1.15 ± 0.03^{a}	8.36 ± 0.2^{a}	2.81 ± 0.24^{a}	4.56 ± 0.26^{bc}	87.7 ± 0.4^{b}	409 ± 1^{a}
96	1.67 ± 0.03^{b}	11.2 ± 0.2^{bc}	4.50 ± 0.12^{b}	1.93 ± 0.08^{a}	82.7 ± 0.1^{a}	415 ± 1^{bc}
120	1.88 ± 0.05^{a}	11.4 ± 0.2^{bc}	4.51 ± 0.31^{b}	1.83 ± 0.03^{a}	82.2 ± 0.4^{a}	414 ± 1^{bc}
144	1.78 ± 0.13^{b}	11.2 ± 0.2^{bc}	4.18 ± 0.24^{b}	4.24 ± 0.27^{b}	82.9 ± 0.5^{a}	413 ± 1^{b}
168	1.70 ± 0.10^{b}	11.6 ± 0.2^{c}	4.73 ± 0.16^{b}	2.10 ± 0.11^{a}	82.0 ± 0.3^{b}	416 ± 1^{c}
1 Pearl millet:2 Maize (P1M2) *Injera*						
0	1.26 ± 0.06^{a}	9.27 ± 0.2^{a}	4.54 ± 0.04^{a}	3.62 ± 0.15^{d}	84.9 ± 0.3^{a}	417 ± 0^{a}
96	1.88 ± 0.09^{a}	11.2 ± 0.4^{cd}	3.74 ± 0.12^{a}	2.91 ± 0.13^{bc}	83.6 ± 0.3^{a}	412 ± 1^{a}
120	2.04 ± 0.21^{b}	10.8 ± 0.2^{ab}	3.96 ± 0.97^{a}	2.21 ± 0.04^{a}	83.5 ± 1.2^{a}	412 ± 5^{a}
144	1.90 ± 0.16^{b}	10.8 ± 1.3^{ab}	3.66 ± 0.12^{a}	$2.57 \pm 0.18a^{b}$	83.9 ± 1.4^{a}	411 ± 1^{a}
168	2.00 ± 0.04^{b}	11.1 ± 0.2^{b}	3.99 ± 0.36^{a}	3.07 ± 0.20^{c}	81.7 ± 3.0^{a}	419 ± 14^{a}
Maize (M) *Injera*						
0	1.23 ± 0.03^{a}	8.86 ± 0.23^{ab}	3.72 ± 0.11^{d}	2.83 ± 0.05^{e}	86.3 ± 1.3^{a}	414 ± 7^{a}
96	1.35 ± 0.04^{ab}	8.96 ± 0.01^{b}	2.71 ± 0.21^{a}	2.21 ± 0.13^{c}	87.1 ± 0.4^{a}	407 ± 3^{a}
120	1.36 ± 0.03^{b}	8.94 ± 0.11^{ab}	2.86 ± 0.22^{ab}	1.94 ± 0.11^{b}	86.8 ± 0.2^{a}	409 ± 1^{a}
144	1.37 ± 0.10^{b}	8.85 ± 0.22^{ab}	3.12 ± 0.20^{abc}	1.62 ± 0.05^{a}	87.1 ± 0.4^{a}	410 ± 1^{a}
168	1.45 ± 0.03^{b}	8.49 ± 0.22^{a}	3.37 ± 0.17^{cd}	1.58 ± 0.03^{a}	86.2 ± 0.2^{a}	411 ± 1^{a}

Note. The result values are presented in mean ± *SD*. The means with different letters across the columns were significantly different (at $p < .05$).

and glycerol for energy production (Inyang & Zakari, 2008). While fat content started to increase between 144 and 168 hr fermentation caused by yeast metabolism creating lipids as a byproduct (Matouk et al., 2025) and short-chain fatty acids formation by some types of micro-organisms (Kitessa, 2024).

The crude fibre content of *Injera* samples ranged from 1.58 to 4.22 g/100 g. There were significant differences ($p < .05$) in the fibre content of *Injera* samples. The fibre content of the studied *Injera* samples decreased with fermentation time which might be due to the partial solubilisation of cellulose and hemicellulose by microbial enzymes (Adebo et al., 2022).

The carbohydrate level of our samples ranged from 79.5% to 87.1%; the minimum recorded for *Injera* from pearl millet fermented for 168 hr and the maximum for maize *Injera* baked after 144 hr of fermentation. When the proportion of maize flour increased, the carbohydrate content of the resulting *Injera* also increased. This might be due to the higher carbohydrate level of maize flour (86.2%) compared to pearl millet (82.1%). The amount of carbohydrate in the studied *Injera* recipes decreased with fermentation time. This might be due to starch–hydrolysing enzymes like alpha-amylase and maltase, which break down complex carbohydrates to simpler sugars that fermenting microorganisms can metabolise (Kitessa, 2024). Similarly, Inyang and Zakari (2008) reported a decrease in the carbohydrate levels of fermented foods prepared from pearl millet during fermentation whereas *Injera* samples' gross energy value ranged from 408 to 419 kcal/100 g. The maximum energy value (419 kcal/100 g) was recorded for pearl millet *Injera*, while the minimum (408 kcal/100 g) was for maize *Injera* prepared after fermentation for 96 hr. *Injera*'s gross energy value also increased with the amount of pearl millet flour in the composite flour was increased. This could be related to higher value of fat, and protein in pearl millet flour compared to maize flour.

Effect of fermentation on antinutritional content

Table 2 shows the content of phytate and tannin from *Injera* samples and their respective flours. Phytic acid acts as the primary phosphorus storage compound in cereals, legumes, oilseeds, and nuts. However, PA is considered as an antinutrient due to its ability to form complexes with minerals, proteins, and carbohydrates, which can reduce their bioavailability and absorption. This is a particular concern in low-income countries, where diets rely heavily on plant-based foods (Feizollahi et al., 2021). Pearl millet (P) flour has highest phytate concentration (1,633 mg/100 g). Regarding the samples of *Injera*, the highest PA content (594 mg/100 g) was recorded for pearl millet (P) *Injera* fermented for 96 hr, whereas the lowest (8.31 mg/100 g) was recorded for *Injera* samples prepared from P1M1 flour fermented for 168 hr. Significant ($p < .05$) reductions were recorded for the phytate content (ranging from 81.5% to 99.2%) for all *Injera* samples with increased fermentation time.

Phytate content reduction during extended fermentation time observed in this study might be attributed to the presence of microbes, and natural phytase enzymes found in the cereals and the synthesis of organic acids, lowering pH of the substrate and creating favourable conditions for the function of phytases (Adebo et al., 2022; Osman, 2011). Similar findings were reported by Mihrete and Bultosa (2017) when the teff-*Injera* is fermenting.

The tannin content of samples (both unfermented flour and *Injera* samples) spanned from 2.50 to 83.0 mg CE/100 g. The maximum condensed tannin (83.0 mg CE/100 g) content was recorded for pearl millet flour. After 168 hr fermentation, the lowest tannin content (2.50 mg CE/100 g) was recorded for P1M1 *Injera*. The significant ($p < .05$) reduction of condensed tannin of *Injera* samples during fermentation was due to various reasons including hydrolysis of ester and dipeptide linkages catalysed by the enzyme tannase, resulting from formation of gallic acid and

Table 2. Phytic acid and tannin content of pearl millet-based *Injera* and flour samples (in d.w.).

Sample			Phytic acid (mg/100 g)	Condensed tannin (mg CE/100 g)
Flour	P		1,633 ± 8.9^{a}	83.0 ± 5.1^{c}
	P1M1		1,012 ± 93^{d}	72.5 ± 0.9^{a}
	P1M2		502 ± 8.98^{b}	64.1 ± 1.9^{a}
	M		432 ± 3.9^{b}	37.7 ± 1.3^{d}
		Fermentation time (hr)		
Injera	P	96	594 ± 5.2^{c}	44.0 ± 2.43^{c}
		120	433 ± 2.2^{b}	34.9 ± 1.26^{b}
		144	393 ± 1.7^{b}	30.2 ± 0.63^{b}
		168	42.0 ± 4.1^{a}	8.83 ± 0.80^{a}
	P1M1	96	358 ± 35^{c}	20.3 ± 1.4^{c}
		120	168 ± 1.1^{ab}	15.1 ± 1.2^{b}
		144	28.0 ± 0.3^{a}	10.5 ± 0.8^{b}
		168	8.31 ± 0.7^{a}	2.50 ± 0.13^{a}
	P1M2	96	415 ± 2.9^{c}	19.1 ± 0.8^{c}
		120	326 ± 2.0^{c}	12.7 ± 0.7^{b}
		144	167 ± 1.0^{b}	10.3 ± 0.9^{b}
		168	18.2 ± 1.5^{b}	5.31 ± 0.27^{a}
	M	96	215 ± 2.1^{d}	17.6 ± 0.1^{bc}
		120	172 ± 1.7^{c}	12.6 ± 3.4^{ab}
		144	114 ± 1.5^{b}	11.1 ± 2.2^{a}
		168	80.0 ± 1.5^{a}	10.4 ± 1.3^{a}

Note. The result values are in mean ± *SD*. Mean with different letters across columns were significantly different ($p < .05$). Where: P (100% pearl millet) *Injera*; P1M1 (1 pearl millet:1 maize) *Injera*; P1M2 (1 pearl millet:2 maize) *Injera*, M (100% maize) *Injera*.

Table 3. Iron, calcium, and zinc content (mg/100 g in d.w.) of pearl millet-based *Injera* recipes with different fermentation time preparation.

Fermentation time (hr)	Iron (Fe)	Zinc (Zn)	Calcium (Ca)
Pearl millet (P) *Injera*			
0	10.8 ± 0.02^{a}	4.14 ± 0.12^{a}	21.7 ± 0.4^{a}
96	12.3 ± 0.08^{a}	4.34 ± 0.06^{ab}	24.4 ± 0.1^{c}
120	13.6 ± 0.68^{b}	4.42 ± 0.05^{b}	27.3 ± 0.4^{d}
144	15.9 ± 0.07^{c}	4.92 ± 0.01^{c}	29.8 ± 0.1^{c}
168	18.4 ± 0.38^{d}	5.11 ± 0.06^{c}	31.5 ± 0.2^{d}
1 Pearl millet: 1 Maize (P1M1) *Injera*			
0	7.29 ± 0.01^{a}	2.86 ± 0.08^{a}	17.7 ± 0.1^{a}
96	9.08 ± 0.17^{b}	3.77 ± 0.02^{c}	24.5 ± 0.1^{c}
120	9.39 ± 0.16^{bc}	3.81 ± 0.02^{c}	25.8 ± 0.2^{d}
144	9.54 ± 0.06^{c}	5.30 ± 0.01^{d}	26.2 ± 0.1^{d}
168	9.66 ± 0.06^{c}	6.19 ± 0.01^{b}	30.0 ± 0.2^{b}
1 Pearl millet:2 Maize (P1M2) *Injera*			
0	7.66 ± 0.06^{a}	2.31 ± 0.01^{a}	19.4 ± 0.2^{a}
96	8.16 ± 0.01^{b}	3.18 ± 0.03^{c}	21.8 ± 0.3^{b}
120	8.43 ± 0.18^{bc}	3.35 ± 0.04^{d}	22.6 ± 0.6^{c}
144	8.80 ± 0.14^{cd}	3.38 ± 0.06^{d}	25.7 ± 0.4^{d}
168	9.01 ± 0.13^{d}	3.68 ± 0.09^{b}	26.7 ± 0.3^{d}
Maize (M) *Injera*			
0	5.62 ± 0.62^{a}	1.80 ± 0.01^{a}	16.5 ± 0.2^{a}
96	7.01 ± 0.27^{bc}	2.09 ± 0.03^{c}	20.7 ± 0.1^{d}
120	7.46 ± 0.06^{bc}	2.13 ± 0.04^{c}	22.3 ± 0.1^{c}
144	7.84 ± 0.37^{c}	2.17 ± 0.05^{cd}	23.3 ± 0.1^{cd}
168	8.32 ± 0.16^{c}	2.24 ± 0.01^{d}	26.4 ± 0.1^{b}

Note. The result values are in mean ± *SD* (with duplicate experiments). There was a significant difference ($p < .05$) in the mean across the columns with different letters.

glucose. Furthermore, the breakdown of tannins by enzymes is facilitated by a drop in pH and tannins solubility in water, leading to leaching into the fermenting medium (Adebo et al., 2022). Tannin can be also reduced by the enzyme tannin acyl hydrolase and enzymes produced from fermenting microorganisms which break down tannin-protein, tannic acid-starch, and tannin-iron complexes, releasing free nutrients that ultimately enhance nutritional availability (Nkhata et al., 2018).

Proximate composition and antinutrient levels of pearl millet-based *Injera* samples prepared on the fourth day

The present work investigated the nutritional and antinutrient values of *Injera* samples prepared after 96 hr fermentation of the dough following the traditional recipes of women from Dangeshita kebele. *Injera* prepared solely with pearl millet flour (P-*Injera*) exhibited significantly higher ($p < .05$) protein, fat, and fibre content compared to *Injera* prepared from pearl millet–maize composite flours (Supplementary Table 1). These results imply that pearl millet flour is a significant contributor to *Injera's* protein content, as evidenced by the increasing levels with higher millet flour ratios. Maize flour appears to be the main source of carbohydrates in *Injera* samples prepared with pearl millet-maize composite flour. The mean energy value of all *Injera* samples ranged from 408 to 415 kcal/100 g. The study also revealed significant differences ($p < .05$) in antinutrient content (phytate and tannin) among the *Injera* samples examined. *Injera* prepared solely from pearl millet flour exhibited the highest phytate (594 mg/100 g) and tannin (44 mg CE/100 g) concentrations compared to other recipes. However, phytate was reduced by 64.6% and 17.3%, while tannin was reduced by 72% and 70.2%, when pearl millet was mixed with maize flour at a ratio of 1:1 and 1:2, respectively. This suggests that pearl millet-based *Injera* might be a good source of plant protein, and fat, despite containing higher levels of antinutrients which actually decreased to an acceptable level during the later stages of fermentation.

Mineral content of pearl millet-based *Injera*

The iron (Fe), zinc (Zn), and calcium (Ca) content of *Injera* prepared from pearl millet and pearl millet-maize composite flours is presented in Table 3. The unfermented pearl millet (P) flour exhibited the highest content of Fe, Zn, and Ca (10.8 mg/100 g dw, 4.14 mg/100 g dw, 21.7 mg/100 g dw respectively), compared to other unfermented flours. Pearl millet *Injera* contained the highest Fe content (18.4 mg/100 g) followed by 1:1 pearl millet and maize (P1M1) (9.66 mg/100 g) and 1:2 pearl millet and maize (P1M2) (9.01 mg/100 g) *Injera* samples after 168 hr fermentation. Increment in iron content during fermentation of pearl millet was also reported by Balli et al. (2023) and Anberbir et al. (2023). Compared to unfermented flour samples, the iron content of *Injera* samples prepared from pearl millet (P) dough, 1:1 pearl millet and maize (P1M1) composite dough, 1:2 pearl millet and maize (P1M2) composite dough and maize (M) dough was considerably higher at 168 hr fermentation.

With regard to zinc, the highest content (6.19 mg/100 g) was recorded for *Injera* prepared from P1M1 flour fermented for 168 hr followed by pearl millet (P) *Injera* (5.11 mg/100 g), while the lowest zinc content (2.09 mg/100 g) was recorded for maize (M) *Injera* fermented for 96 hr. The zinc content of all *Injera* samples showed significant ($p < .05$) difference during the various fermentation times. Specifically, a significant change was observed for all *Injera* samples at 96 hr of fermentation. Furthermore, for P1M1, and M *Injera*, a significant difference was observed from 120 to 168 hr of fermentation times. The zinc content of pearl millet based

Injera samples (1.80–6.19 mg/100 g) was higher than that reported by Cherie et al. (2018) (1.62–2.11 mg/100 g) for *Injera* prepared from teff, maize, and rice composite flours and that reported by Anberbir et al. (2023) for *Injera* samples prepared from teff, pearl millet, and buckwheat composite flours (1.14–3.32 mg/100 g).

Among unfermented flour samples, pearl millet (P) flour showed the highest calcium content (21.7 mg/100 g), while maize (M) flour contained the lowest (16.5 mg/100 g). Correspondingly, pearl millet (P) *Injera* exhibited the highest calcium (Ca) content (31.5 mg/100 g) after 168 hr of fermentation, whereas, the lowest Ca content (20.7 mg/100 g) was observed in maize *Injera* fermented for 96 hr. Fermentation significantly ($p < .05$) increased the calcium (Ca) content of *Injera* samples. Specifically, *Injera* prepared from 1:1 pearl millet and maize (P1M1) and 1:2 pearl millet and maize (P1M2) composite flours exhibited significant changes in Ca content up to 144 hr of fermentation. In contrast, pearl millet (P) and maize (M) *Injera* samples showed such differences only up to 120 hr of fermentation. The current finding agrees with previous studies by Endalew et al. (2024) and Anberbir et al. (2023), who reported an increase in calcium content during the fermentation of finger millet and teff, respectively, for Injera preparation.

Generally, the mineral content of *Injera* samples prepared from pearl millet and pearl millet-maize composite flours exhibited an increment with fermentation time. This finding agrees with a previous study by Endalew et al. (2024), who reported a significant increment in iron, zinc, and calcium content during finger millet and maize fermentation in the preparation of Injera. Balli et al. (2023) also reported an increment in iron, calcium, and phosphorus content during pearl millet fermentation. The observed increment in the mineral content of *Injera* samples with an increase in fermentation time may be attributed to a reduction in dry matter content, resulting from the microbial degradation of carbohydrates, proteins, and crude fat during the fermentation process (Gabaza et al., 2018; Nkhata et al., 2018).

Bioaccessibility of minerals

The bioaccessibility of iron (Fe), zinc (Zn), and calcium (Ca) of different *Injera* recipes is displayed in Figure 2. Fermentation significantly enhanced the Fe, Zn, and Ca bioaccessibility in all *Injera* recipes, with increments ranging from 62.1%–73.5% for Fe, 53.8%–83.3% for Zn, and 19.6%–54.6% for Ca. *Injera* samples from pearl millet flour alone exhibited the greatest improvement in mineral bioaccessibility. This increment can be related to the decrease in antinutrients during fermentation, specifically phytate and tannin levels. Fermentation by naturally occurring microflora promotes the degradation of antinutrient-mineral complexes through enzymatic activity, and releases free minerals, enhancing their bioaccessibility and bioavailability (Endalew et al., 2024; Nkhata et al., 2018). Furthermore, the pH reduction during fermentation creates a more favourable environment for the activity of phytase enzymes. These enzymes break down phytate, thereby enhancing the Fe, Zn, and Ca bioaccessibility and bioavailability in fermented food products (Frontela et al., 2011; Sharma et al., 2020).

Sensory acceptability of pearl millet-based *Injera*

The sensory value of the different *Injera* samples is presented in Figure 3. The sensory acceptability scores for *Injera* samples especially colour, taste, texture, and overall acceptability were significantly ($p < .05$) affected by fermentation (Supplementary Table 2).

Colour is used to assess the basic sensory attribute and acceptability of traditional baked goods. The colour of pearl millet (P) *Injera* was the most preferred by the panellists (6.83). Among all *Injera* samples, a trend of decreasing colour values was observed with increasing fermentation time, possibly being related to the pigment breakdown by fermentation process (Altan et al., 2008). The sensory acceptance scores for colour significantly increased with the proportion of pearl millet flour in *Injera* samples. The observed colour differences in the samples might be attributed to pearl millet flour's higher inherent lightness value due to pigment extraction by starch hydrolysis processes during fermentation, leading to a lower pigment content of maize flour. Additionally, colour variations can occur due to chemical and metabolic reactions during baking (Jebessa et al., 2024).

Taste is the most crucial factor for selecting food and ensuring its success in competition, along with other perceived attributes (Heiniö et al., 2016). In order to get the right flavour of *Injera* when paired with stew (known as "wot" in Amharic), high-quality *Injera* should possess a slight sourness. The maximum (6.83) taste score of *Injera* was observed for pearl millet (P) *Injera* fermented for 96 hr, while the lowest (4.25) for maize (M) *Injera* fermented for 144 hr.

Texture constitutes main sensory property typically used to evaluate the quality of cereal-based baked items. It pertains to the food product's level of softness or hardness and is assessed by touch. *Injera* recipes texture score ranged from 4.50 to 6.83. The highest (6.83) texture or softness score was recorded for pearl millet (P) *Injera* fermented for 96 hr, while the lowest (4.50) for maize *Injera* fermented for 144 hr.

The capacity of *Injera* to be rolled is linked to the reduced tendency of baked goods to become stale. The process of staling in the product caused to an increase in stiffness and a decrease in the acceptability of baked products with time. The acceptability score in the rollability of *Injera* ranged from 3.75 to 6.67. The panellists preferred the rollability of pearl millet *Injera* over the other *Injera* recipes followed by P1M1 *Injera* fermented for 144 and 120 hr, respectively. The inclusion of maize flour in mixing process resulted in a higher level of firmness in the *Injera* samples, even though the change in rollability was not drastic.

The visual aspect (appearance) of *Injera* is a crucial factor that determines its quality, encompassing both its external appearance and the texture beneath the surface. The surface eyes of *Injera* are the small honeycomb-like structures that emerge on the top surface due to the generation and release of carbon dioxide while fermenting and baking (Neela & Fanta, 2020). The highest acceptance score was attained for pearl millet (P) *Injera* (6.33) fermented for 96 hr followed by *Injera* prepared from P1M2 flour (6.20) fermented for 120 hr. The score for eye uniformity ranged from 4.00 to 6.33. Pearl millet (P) *Injera* fermented for 120 hr had superior eye uniformity scores; whereas the lowest was obtained from maize (M) *Injera* fermented for 168 hr. As the fermentation time increased from 96 to 120 hr, the eye uniformity of *Injera* samples increased, decreasing beyond 120 hr.

The *Injera* samples overall acceptability score ranged from 3.63 to 6.83, suggesting a good acceptability of the samples. The highest overall acceptance was obtained for pearl millet (P) *Injera* fermented for 96 hr followed by *Injera* produced from P1M1 *Injera* (6.25) fermented for 144 hr.

Overall fermentation affected the sensory acceptability of our samples. Similar findings were reported for *Injera* samples prepared from various cereals including millet, teff, and maize, attributed to the influence of LAB, which generates organic acids and various flavour molecules during fermentation. Additionally, the decrease in pH changes the composition of substrate metabolites and soluble organic acids released from the fermenting media, further enhancing the sensory profiles

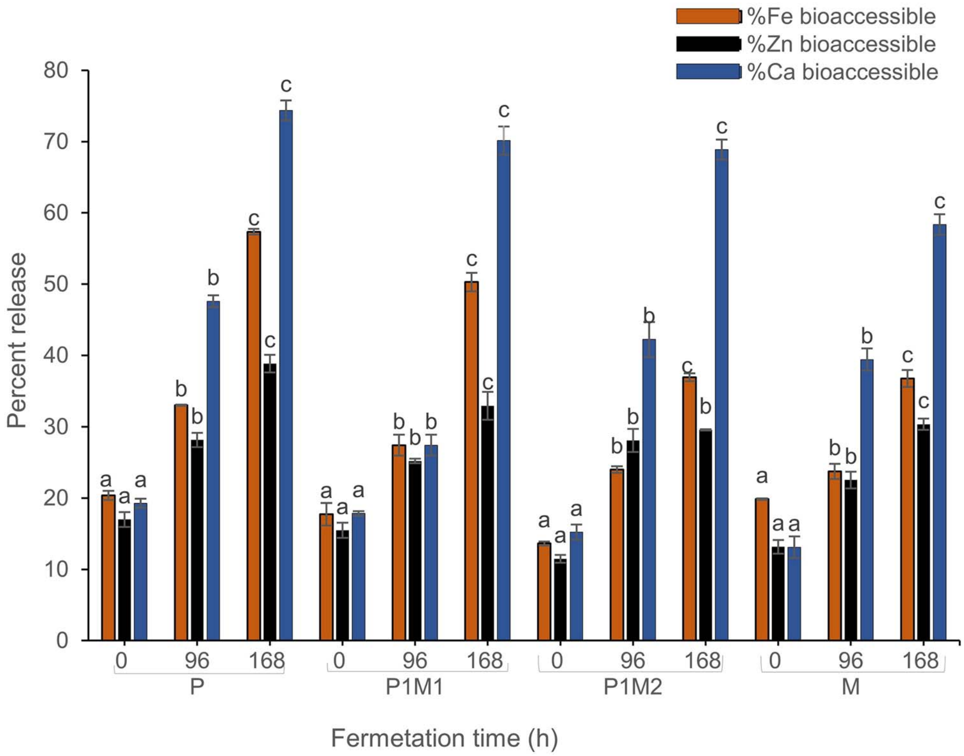


Figure 2. Bioaccessible Ca, Fe, and Zn from pearl millet-based *Injera* during different fermentation times (error bars represents standard deviation of the means).

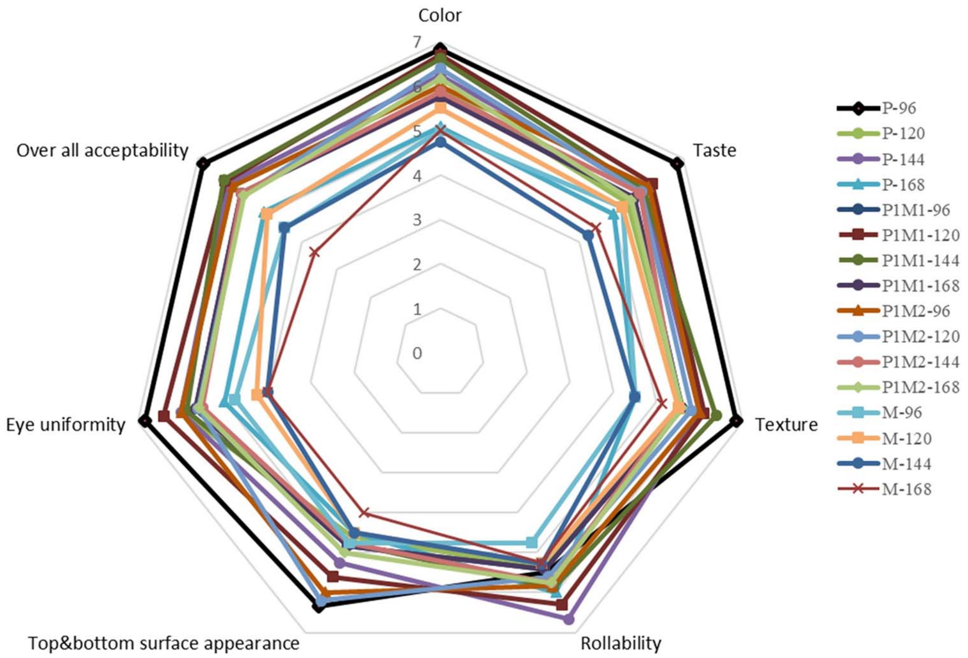


Figure 3. Effect of fermentation on sensory acceptability of *Injera*. 96,120, 144, and 168 in the graph represent fermentation hours.

(Mengesha et al., 2022; Mihrete & Bultosa, 2017; Mutshinyani et al., 2020).

Conclusion

Fermentation improved the nutritional and sensory attributes of all *Injera* recipes while effectively decreasing phytate, and condensed tannins, which resulted in increased bioaccessibility of minerals. *Injera* prepared from pearl millet flour alone and *Injera* prepared from composite flour of pearl millet and maize in a one-to-one proportion and fermented for 96 hr or up to 144 hr were found to be nutritionally superior with better sensorial quality.

Supplementary material

Supplementary material is available at *International Journal of Food Science and Technology* online.

Data availability

The data that support this study will be made available on request from the corresponding authors.

Author contributions

Tadesse Fenta Yehuala (Writing—original draft, Conceptualization, Methodology, Formal analysis, Investigation [equal]), Sotirios Karavoltsos (Writing—review & editing, Resources, Formal analysis [equal]), Aikaterini Sakellari (Writing—review & editing, Resources, Formal analysis [equal]), Mohamad Farshard Aslam (Writing—review & editing, Resources, Formal analysis [equal]), Howard Griffiths (Supervision, Resources, Writing—review & editing [equal]), Lara Allen (Supervision, Resources, Writing—review & editing [equal]), Anastasia Kanellou (Supervision, Resources, Writing—review & editing, Resources [equal]), Panagiotis Zoumpoulakis (Supervision, Resources, Writing—review & editing, Resources [equal]), Wanjiku Gichohi-Wainaina (Supervision, Writing—review & editing [equal]), Metadel Kassahun Abera (Supervision, Writing—review & editing [equal]), Mesfin Wogahyehu Tenagashaw (Supervision, Writing—review & editing [equal]), Gizaw Desta Gessesse (Supervision, Writing—review & editing [equal]), Helen Walle Endalew (Investigation [equal]), Hirut Assaye Cherie (Conceptualization, Resources, Writing—review & editing, Methodology, Formal analysis [equal]), and Minaleshewa Atlabachew (Conceptualization, Resources, Writing—review & editing, Methodology, Formal analysis [equal])

Funding

This work was supported by a contribution from Cambridge Research Service Division, United Kingdom (Biotechnology and Biological Sciences Research Council [BBSRC] Grant Ref: BB/S013954/1), with Cambridge University.

Conflicts of interest

There is no conflict of interests or relations that might have impact on this paper's work.

Acknowledgements

The authors wish to express their gratitude to ICRSAT, Ethiopia for supplying the pearl millet samples. Special thanks are also due to the women of the Dangeshita sub-district communities for their help in sample preparation. Additionally, we are very grateful for the financial support granted to Tadesse Fenta Yehuala through the ICM Erasmus+ Student Mobility Program in Greece, particularly from the Food Science and Technology Department at the University of West Attica, which partnered with the Laboratory of Environmental Chemistry (at University of Athens, 15784 Athens) for specific lab analyses. Lastly, we acknowledge Professor Seifu Admassu Tilahun for his significant role in facilitating collaboration between Ethiopian and European partners.

References

Adebo, J. A., Njobeh, P. B., Gbashi, S., Oyedeji, A. B., Ogundele, O. M., Oyeyinka, S. A., & Adebo, O. A. (2022). Fermentation of cereals and legumes: Impact on nutritional constituents and nutrient bioavailability. *Fermentation*, *8*, 2. https://doi.org/10.3390/fermentation8020063.

Ali, M. A., El Tinay, A. H., & Abdalla, A. H. (2003). Effect of fermentation on the in vitro protein digestibility of pearl millet. *Food chemistry*, *80*, 51–54.

Altan, A., McCarthy, K. L., & Maskan, M. (2008). Twin-screw extrusion of barley–grape pomace blends: Extrudate characteristics and determination of optimum processing conditions. *Journal of Food Engineering*, *89*, 24–32. https://doi.org/10.1016/j.jfoodeng.2008.03.025

Anberbir, S. M., Satheesh, N., Abera, A. A., Tenagashaw, M. W., Asres, D. T., Tiruneh, A. T., Habtu, T. A., Awol, S. J., & Wudineh, T. A. (2023). Effect of blending ratio and fermentation time on the physicochemical, microbiological, and sensory qualities of injera from teff, pearl millet, and buckwheat flours. *CyTA - Journal of Food*, *21*, 217–236. https://doi.org/10.1080/19476337.2023.2188058

AOAC (2005). *Official methods of analysis of AOAC International Association of Official Analytical Chemists* (18th ed.). Rockville: AOAC International.

Ashagrie, Z., & A., D. (2012). Improvement of injera shelf life through the use of chemical preservatives. *African Journal of Food, Agriculture, Nutrition and Development*, *12*, 6409–6423. https://doi.org/10.18697/ajfand.53.10910

Balli, D., Cecchi, L., Pieraccini, G., Venturi, M., Galli, V., Reggio, M., di Gioia, D., Furlanetto, S., Orlandini, S., Innocenti, M., & Mulinacci, N. (2023). Millet fermented by different combinations of yeasts and lactobacilli: Effects on phenolic composition, starch, mineral content and prebiotic activity. *Foods*, *12*, 748. https://doi.org/10.3390/foods12040748

Baye, K., Mouquet-Rivier, C., Icard-Vernière, C., Picq, C., & Guyot, J. P. (2014). Changes in mineral absorption inhibitors consequent to fermentation of Ethiopian injera: Implications for predicted iron bioavailability and bioaccessibility. *International Journal of Food Science & Technology*, *49*, 174–180. https://doi.org/10.1111/ijfs.12295

Cherie, Z., Ziegler, G. R., Gemede, H. F., & Woldegiorgis, A. Z. (2018). Optimization and modeling of teff-maize-rice based formulation by simplex lattice mixture design for the preparation of brighter and acceptable injera. *Cogent Food & Agriculture*, *4*, 1–19. https://doi.org/10.1080/23311932.2018.1443381

Dayakar, Rao, B., Ananthan, R., Hariprasanna, K., Bhatt, K. V., Rajeswari, S. S., & Tonapi, V. A. (2018). Nutritional and health benefits of nutri cereals. *Nutri hub TBI, ICAR_Indian Institute of Millets Research (IIMR) Rajendranagar, Hyderabad*, *96*, 1–100.

Dias-Martins, A. M., Pessanha, K. L. F., Pacheco, S., Rodrigues, J. A. S., & Carvalho, C. W. P. (2018). Potential use of pearl millet (*Pennisetum glaucum* (L.) R. Br.) in Brazil: Food security, processing, health benefits and nutritional products. *Food Research International*, *109*, 175–186. https://doi.org/10.1016/j.foodres.2018.04.023

Diether, N. E., & Willing, B. P. (2019). Microbial fermentation of dietary protein: An important factor in diet–microbe–host interaction. *Microorganisms*, *7*, 1. https://doi.org/10.3390/microorganisms7010019

Dykes, L. (2019). *Tannin analysis in sorghum grains* (pp. 109–120). Sorghum: Methods and Protocols.

Endalew, H. W., Atlabachew, M., Karavoltsos, S., Sakellari, A., Aslam, M. F., Allen, L., Griffiths, H., Zoumpoulakis, P., Kanellou, A., Yehuala, T. F., Abera, M. K., Tenagashaw, M. W., & Cherie, H. A. (2024). Effect of fermentation on nutrient composition, antinutrients, and mineral bioaccessibility of finger millet based *Injera*: A traditional Ethiopian food. *Food Research International*, *190*, 114635. https://doi.org/10.1016/j.foodres.2024.114635

Erenso Degu, E. D., Asfaw Adugna, A. A., Taye Tadesse, T. T., & Tesso, T. (2009). Genetic resources, breeding and production of millets in Ethiopia.

Feizollahi, E., Mirmahdi, R. S., Zoghi, A., Zijlstra, R. T., Roopesh, M. S., & Vasanthan, T. (2021). Review of the beneficial and anti-nutritional qualities of phytic acid, and procedures for removing it from food products. *Food Research International*, *143*, 110284. https://doi.org/10.1016/J.FOODRES.2021.110284

FNS (2023). *National food and nutrition strategy baseline survey: Key findings preliminary survey*. Addis Ababa, Ethiopia: Ethiopian Public Health Institute.

Frontela, C., Ros, G., & Martínez, C. (2011). Phytic acid content and "*in vitro*" iron, calcium and zinc bioavailability in bakery products: The effect of processing. *Journal of Cereal Science*, 54, 173–179. https://doi.org/10.1016/j.jcs.2011.02.015

Gabaza, M., Shumoy, H., Muchuweti, M., Vandamme, P., & Raes, K. (2018). Iron and zinc bioaccessibility of fermented maize, sorghum and millets from five locations in Zimbabwe. *Food Research International*, *103*, 361–370. https://doi.org/10.1016/j.foodres.2017.10.047

Glahn, R. P., Lee, O. A., Yeung, A., Goldman, M. I., & Miller, D. D. (1998). Caco-2 cell ferritin formation predicts nonradiolabeled food iron availability in an In vitro digestion/Caco-2 cell culture Model1. *The Journal of Nutrition*, *128*, 1555–1561. https://doi.org/10.1093/jn/128.9.1555

Grigoriou, C., Costopoulou, D., Vassiliadou, I., Karavoltsos, S., Sakellari, A., Bakeas, E., & Leondiadis, L. (2022). Polycyclic aromatic hydrocarbons and trace elements dietary intake in inhabitants of Athens, Greece, based on a duplicate portion study. *Food and Chemical Toxicology*, *165*, 113087, ISSN 0278-6915. https://doi.org/10.1016/j.fct.2022.113087

Hailu, A. (2016). Ethiopian national micronutrient survey report. *Ethiopian Public Health Institute*. Ethiopia: Addis Ababa.

Heiniö, R. L., Noort, M. W. J., Katina, K., Alam, S. A., Sozer, N., de Kock, H. L., Hersleth, M., & Poutanen, K. (2016). Sensory characteristics of wholegrain and bran-rich cereal foods – A review. *Trends in Food Science & Technology*, *47*, 25–38. https://doi.org/10.1016/j.tifs.2015.11.002

Inyang, C. U., & Zakari, U. M. (2008). Effect of germination and fermentation of pearl millet on proximate chemical and sensory properties of instant "Fura"- A Nigerian cereal food. *Pakistan Journal of Nutrition*, *7*, 9–12. https://doi.org/10.3923/pjn.2008.9.12

Jain, R. K., & Bal, S. (1997). Properties of pearl millet. *Journal of Agricultural Engineering Research*, 66, 85–91. https://doi.org/10.1006/jaer.1996.0119

Jebessa, T. F., Tolesa, G. N., Bultosa, G., Abera, S., & Hailu, G. G. (2024). Comprehensive study on the effect of fermentation time, baking temperature and baking time on the physicochemical and nutritional properties of *Injera teff* (*Eragrostis teff*). *Food and Humanity*, 2, 100256. https://doi.org/10.1016/j.foohum.2024.100256

Khoja, K. K., Aslam, M. F., Sharp, P. A., & Latunde-Dada, G. O. (2021). *In vitro* bioaccessibility and bioavailability of iron from fenugreek, baobab and moringa. *Food Chemistry*, *335*, 127671. https://doi.org/10.1016/j.foodchem.2020.127671

Kitessa, D. A. (2024). Review on effect of fermentation on physicochemical properties, anti-nutritional factors and sensory properties of cereal-based fermented foods and beverages. *Annales de Microbiologie*, *74*, 32. https://doi.org/10.1186/s13213-024-01763-w

Lakew, A., & Berhanu, T. (2019). Determination of intra and inter row spacing on the yield of pearl millet (Pennisetum glaucum L.) in the dry land areas of Wag Himra, eastern Amhara, Ethiopia. *Archives of Agriculture and Environmental Science*, *4*, 45–49.

Lee, J., Park, Y. S., Lee, H. J., & Koo, Y. E. (2022). Microwave-assisted digestion method using diluted nitric acid and hydrogen peroxide for the determination of major and minor elements in milk samples by ICP-OES and ICP-MS. *Food Chemistry*, *373*, 131483–131483. https://doi.org/10.1016/J.FOODCHEM.2021.131483

Lim, S. H., Chin, N. L., Sulaiman, A., Tay, C. H., & Wong, T. H. (2022). Sensory analysis for cow milk product development using high pressure processing (HPP) in the dairy industry. *Foods*, *11*, 9. https://doi.org/10.3390/foods11091233

Long, G. L., & Winefordner, J. D. (1983). Limit of detection. A closer look at the IUPAC definition. *Analytical Chemistry*, 55, 712A–724A.

Lopez, H. W., Leenhardt, F., Coudray, C., & Remesy, C. (2002). Minerals and phytic acid interactions: Is it a real problem for human nutrition? *International Journal of Food Science and Technology*, *37*, 727–739. https://doi.org/10.1046/j.1365-2621.2002.00618.x

Matouk, A. M., Abu-Elreesh, G. M., Abdel-Rahman, M. A., Desouky, S. E., & Hashem, A. H. (2025). Response surface methodology and repeated-batch fermentation strategies for enhancing lipid production from marine oleaginous Candida parapsilosis Y19 using orange peel waste. *Microbial Cell Factories*, 24, 16. https://doi.org/10.1186/s12934-024-02635-3

McKie, V. A., & McCleary, B. V. (2016). A novel and rapid colorimetric method for measuring total phosphorus and phytic acid in foods and animal feeds. *Journal of AOAC International*, *99*, 738–743. https://doi.org/10.5740/jaoacint.16-0029

Mengesha, Y., Tebeje, A., & Tilahun, B. (2022). A review on factors influencing the fermentation process of teff (Eragrostis teff) and other cereal-based Ethiopian injera. *International Journal of Food Science*, 2022, 4419955. https://doi.org/10.1155/2022/4419955

Mihrete, & Bultosa (2017). The effect of blending ratio of tef [*Eragrostis tef* (Zucc) trotter], sorghum (*Sorghum bicolor* (L.) Moench) and Faba bean (Vicia faba) and fermentation time on chemical composition of Injera. *Journal of Nutrition & Food Sciences*, 07. https://doi.org/10.4172/2155-9600.1000583

Mutshinyani, M., Mashau, M. E., & Jideani, A. I. O. (2020). Bioactive compounds, antioxidant activity and consumer acceptability of porridges of finger millet (*Eleusine coracana*) flours: Effects of spontaneous fermentation. *International Journal of Food Properties*, 23, 1692–1710. https://doi.org/10.1080/10942912.2020.1825485

Neela, S., & Fanta, S. W. (2020). Injera (an ethnic, traditional staple food of Ethiopia): A review on traditional practice to scientific developments. *Journal of Ethnic Foods*, *7*, 32. https://doi.org/10.1186/s42779-020-00069-x

Nkhata, S. G., Ayua, E., Kamau, E. H., & Shingiro, J. B. (2018). Fermentation and germination improve nutritional value of cereals and legumes through activation of endogenous enzymes. *Food Science and Nutrition*, 6, 2446–2458. https://doi.org/10.1002/fsn3.846

Onyango, S. O., Abong, G. O., Okoth, M. W., Kilalo, D. C., & Mwang'ombe, A. W. (2021). Effect of pre-treatment and processing on nutritional composition of cassava roots, millet, and cowpea leaves flours. *Frontiers in Sustainable Food Systems*, 5, 1–8. https://doi.org/10.3389/fsufs.2021.625735

Osman, M. A. (2011). Effect of traditional fermentation process on the nutrient and antinutrient contents of pearl millet during preparation of Lohoh. *Journal of the Saudi Society of Agricultural Sciences*, *10*, 1–6. https://doi.org/10.1016/J.JSSAS.2010.06.001

Rani, S., Singh, R., Sehrawat, R., Kaur, B. P., & Upadhyay, A. (2018). Pearl millet processing: A review. *Nutrition & Food Science*, *48*, 30–44. https://doi.org/10.1108/NFS-04-2017-0070

Saleh, A. S. M. M., Zhang, Q., Chen, J., & Shen, Q. (2013). Millet grains: Nutritional quality, processing, and potential health benefits. *Comprehensive Reviews in Food Science and Food Safety*, 12, 281–295. https://doi.org/10.1111/1541-4337.12012

Samtiya, M., Aluko, R. E., & Dhewa, T. (2020). Plant food antinutritional factors and their reduction strategies: An overview.

Food Production, Processing and Nutrition, 2, 1–14. https://doi.org/10.1186/s43014-020-0020-5

Şanlier, N., Gökcen, B. B., & Sezgin, A. C. (2019). Health benefits of fermented foods. *Critical Reviews in Food Science and Nutrition*, 59, 506–527. https://doi.org/10.1080/10408398.2017.1383355

Sharma, R., Garg, P., Kumar, P., Bhatia, S. K., & Kulshrestha, S. (2020). Microbial fermentation and its role in quality improvement of fermented foods. *Fermentation*, 6, 4. https://doi.org/10.3390/fermentation6040106

Tontisirin, K., MacLean, W. C., & Warwick, P. (2003). Food energy: methods of analysis and conversion factors: report of a technical workshop, Rome, 3-6 December 2002 (No. 77). Fao.

Woldemariam, F., Mohammed, A., Fikre Teferra, T., & Gebremedhin, H. (2019). Optimization of amaranths–teff–barley flour blending ratios for better nutritional and sensory acceptability of injera. *Cogent Food & Agriculture*, 5, 1565079. https://doi.org/10.1080/23311932.2019.1565079

International Journal of Food Science and Technology, 2025, **60(2)**, vvaf132
https://doi.org/10.1093/ijfood/vvaf132
Original Article