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Millet: Key to Alleviate Micronutrient Deficiencies (Calcium & Iron) among Adolescent Girls

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Abstract

Nutritionally and economically millets are superior to staples like rice and wheat. Studies suggest that processes like soaking, germination and fermentation have considerably reduced inhibiting factors and improved mineral bioavailability. Cross-sectional studies reported that a large section of Indian females consume less than 50% of the recommended dietary intake of iron, calcium and folic acid. One of the primary reasons for deficiency disorders is the consumption of plant-based diets containing antinutritional factors that inhibit mineral absorption and utilization. This research was undertaken to study the effect of fermentation on mineral bioavailability, antinutrient reduction in Pearl millet and Italian millet and the acceptability of millet-fermented products (Pearl millet dosa and Italian millet dosa) by adolescent girls. On 12hrs of fermentation Pearl millet and Italian millet showed a significant reduction in the phytate content (752 ±0.03 to 650 ±0.04 mg% in Pearl millet and 700 ± 0.05 to 650 ±0.04 mg% in Italian millet). Similarly, polyphenol content decreased from (190.06 ±0.088 to 100.016 ±0.044 and 130.04 ± 0.00 to 100.06 ± 0.031 mg% respectively). This reduction was found to correlate with the increase in calcium and iron bioavailability. Evaluation of the organoleptic properties of millet dosas revealed overall acceptability among adolescent girls. It can thus be concluded that the intake of fermented millet is key to alleviating micronutrient deficiencies among adolescent girls.

Keywords: *Adolescent Girls, Antinutrients, Micronutrient Deficiency, Millets, Fermentation, Mineral Bioavailability.*

Introduction

Adolescence is a crucial stage lasting from 10 to 19 years of age. Adolescents undergo a transition from childhood to adulthood. During this phase, rapid physical, psychological and social changes are noticed. This period is marked by significant incidents of disease, injury, substance abuse, and irregular eating and sleeping patterns which can affect their health. WHO statistics state that one-sixth of the world's population are aged 10-19 years and 6% account for the global burden of disease and injury. Investing in adolescent health is key for a healthy population [1]. Progress for Children – a report by UNICEF, 2012, states that India has the largest number of adolescents, 243 million [2]. A national review suggests that adolescents suffer from both under and over-nutrition [3]. Adolescent girls are more prone to lowered BMI, health risks, early marriages and violence [4, 5]. Some girls suffer from eating disorders like anorexia nervosa, bulimia and binge eating. Such behaviours can influence their net nutrient intake and thus result in deficiency disorders. Recurrent deficiency disorders include deficiencies of vitamins and minerals (vitamin A, vitamin D, calcium, iron,

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iodine, zinc) which are termed micronutrient deficiencies (MiND) or Hidden Hunger. [6] Micronutrients are usually required <100 mg/day [7]. Some studies state that it is a challenging task to detect or screen MiNDs. [8, 9] Though micronutrients are required in small quantities they play a major role in an individual's health and mild to moderate deficiency can significantly affect an individual's physical as well as mental state. [10] MiND can be the result of several factors – dietary insufficiency, improper cooking and processing, lifestyle, economic, environmental, etc.

The prevalence of osteoporosis among Indian women is high [11]. A cross-sectional study in South India reported that Indian women's intake of vitamin A, B1, B2, B3, B12, zinc, folate and iron is considerably less than that of Indian men [12]. Another Indian survey showed that a large percentage of pregnant women in India consume less than half of the recommended dietary allowances of iron, calcium and folic acid [13]. The incidence of deficiency disorders is higher in developing countries than in developed nations. The target groups are young women during menstruation and pregnancy, old people and infants [14]. One of the primary reasons for deficiency disorders is the consumption of plant-based foods which are poor sources of extractable minerals. A study of adolescent girls consuming plant-based diets in Ethiopia confirms the presence of iron deficiency [15]. The dietary minerals present in plant-based foods form a complex with inhibiting factors or antinutrients like phytates and polyphenols, thus making them unavailable to the body for absorption and utilization [16].

Since ages whole grains and cereals have formed an important part of human nutrition. Millets, termed as small-seeded grains are nutritionally better than staples like rice and wheat. Recent studies point out that millets are economically and ecologically more viable than other cereal crops. The health benefits of millet are immense – containing high protein, essential fatty acids, dietary fibre, B vitamins, minerals like calcium, iron, zinc, potassium and magnesium. Other positive outcomes of millet consumption include reduction of blood pressure, lowered cholesterol, and decreased risk of cardiovascular diseases and cancers [17, 18]. Recent studies showed the presence of tryptophan in millet which led to the production of more serotonin, thus having a soothing effect on the mind [19]. However, the presence of antinutrients like phytic acid, tannins and polyphenols hinders the bioavailability of certain nutrients like calcium, phosphorus, iron etc. Studies also suggest that processes like soaking, germination and fermentation reduce such inhibiting factors and make minerals available to the body. [20] Fermentation is one of the oldest and most widely used household cooking processes found to decrease several nutrient inhibitors like trypsin inhibitors, amylase inhibitors, phytic acid and tannins. [21, 22, 23, 24] Millets are diverse food and regular intake of millets can alleviate malnutrition, deficiency diseases and metabolic disorders. They can become an important tool to improve food and nutritional security worldwide.

The benefits of millets for sustainable agriculture and a healthy world in a nutshell as – attaining food security through sustainable food crops, assuring nutritional security as they are rich sources of macro and micronutrients, ensuring safety from nutrient deficiencies and diseases like CVD, diabetes etc. and justifying economic security [25]. Pearl millet is a good source of lipids (4-6%), proteins (12-16%), dietary fibre (11.5%), niacin, folate, calcium, iron, magnesium, copper and zinc [26]. Foxtail millet or Italian millet supplies 11% protein, 59.1% starch, 3.9% fat, 19.1% fibre, vitamins, minerals, and phenolic compounds [27].

This study was undertaken to address public health concerns about micronutrient insufficiencies among adolescent girls and the nutrient potentiality of pearl millet and Italian millet. Fermented pearl millet and Italian millet

showed better mineral bioavailability by reducing antinutrient factors. Moreover, the traditional fermented product, Dosa, was found to be a favourite among young girls.

Methodology

Procurement of Sample

Pearl millet and Italian millet were purchased from the local market in Anantapur. The seeds were cleaned of stones and other extraneous matter and dried in the sun. The seeds sample was stored in a polythene bag at room temperature till analyzed for various parameters.

Preparation of Fermented Batter

Pearl millet and Italian millet seeds were soaked for 12 hours in sufficient water and ground to a smooth paste. The ground batters were allowed to ferment for 12 hours at 37°C. The moisture content of the fermented batters was determined, and the batters were dried at 100°C for one hour in an oven and then powdered using a mortar and pestle. The powdered samples were stored in air-tight containers and used for analysis.

Chemical Analysis

Ash Solution for Mineral Estimation

The ash obtained by ashing the sample in the muffle furnace at 550°C± 15°C for four hours (AOAC, 1985) [28] was dissolved in 5 ml of concentrated HCl and heated over a burner for complete dissolving. The contents from the crucible were transferred carefully into a volumetric flask and made up to 100 ml with distilled water. This solution was used for the estimation of minerals viz., calcium, iron and phosphorus.

Estimation of Calcium

Calcium in the ash solution was estimated by the titrimetric method [29]. Two ml of ash solution was pipetted out into centrifuge tubes. A drop of methyl red indicator was added to the tubes and the solution turned light pink. Liquor ammonia was added till it turned to light yellow. This showed that the solution was too alkaline. A drop of glacial acetic acid was added to give a salmon pink colour indicating the correct pH for the precipitation of calcium. This was mixed well, and 1 ml of ammonium oxalate solution was added.

The tubes were kept overnight and centrifuged at 2,000 rpm, the following day. The supernatant was thrown off by inverting the tubes carefully on a clean filter paper and drained. Care was taken to prevent the precipitate from running along the tubes' sides. Four ml of liquor ammonia was added to the precipitate along the sides of the test tube and were centrifuged again. The supernatant was thrown off and the process was repeated 2 to 3 times. Ten ml of 1N sulphuric acid was added and the contents were made to dissolve. The tubes were kept in a water bath till the contents started simmering and titrated against 0.01N potassium permanganate when still hot. The end point was a definite pink colour which persisted for at least one minute.

Standardization of KMnO4 against 0.01N oxalic acid was done. The endpoint was the appearance of a permanent pale pink colour. The titration was repeated till concurrent values were obtained.

Estimation of Iron

The iron in the ash solution was estimated by Wong's method given by Raghuramulu et al., 1983 [30], and the working standard solution was taken in the volumes of 1.0, 1.5, 2.0, 2.5 and 3.0 ml which correspond to 0.01, 0.015, 0.02, 0.025 and 0.03 mg of iron respectively in different test tubes. To all the test tubes 1.0 ml of 30% H_2SO_4, 1ml potassium sulphate solution, followed by 1.5 ml of 40% potassium thiocyanate were added. Distilled water was added to make up the volume to 10 ml in each test tube and was allowed to stand for 20 mins. The intensity of the colour developed was read in the colourimeter at 540 nm.

Two ml of ash solution was taken, and all reagents were added as standard. The readings were plotted against different concentrations of the standard and the concentration of unknown was intercepted from the graph.

Estimation of Phosphorus

Phosphorus in the ash solution was estimated by Fiske Subba Rao's method given by Raghuramulu et., 1983 [30]. Working standard solution was taken in the volumes of 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5 and 4 ml which correspond to 5, 10, 15, 20, 25, 30, 35 and 40 µg of inorganic phosphorus respectively in different test tubes. To all the test tubes 1.0ml of 3.125% TCA and 1ml of Ammonium Molybdate, followed by 0.4ml of ANSA were added. Distilled water was added to all the test tubes to make up the volume to 10 ml and the tubes were allowed to stand for 15 mins. Fisk.

Two ml of ash solution was taken in triplicates and treated in the same way as the standards. The intensity of the colour developed was read in the colourimeter at 660nm. The readings obtained were plotted against different concentrations of the standard and the concentration of unknown was intercepted from the graph.

Extractable Minerals

The extractable mineral content was estimated by adding one gram of the sample taken in a conical flask. To this 50 ml of 0.03 N HCl was added and incubated at 37°C in a shaker cum water bath for 3 hours to stimulate conditions that occur in the human stomach. The mixture was filtered through Whatman 42 filter paper and the filtrate was oven dried. The residue was digested in the diacid mixture and proceeded for the determination of individual minerals [31].

Calculation

$$\text{Mineral extractability} = \frac{\text{Mineral Extractable in 0.03N HCl}}{\text{Total Mineral}} \times 100$$

Estimation of Antinutritional Factors

Estimation of Oxalic Acid

Oxalic acid in the samples was estimated by the method given by Raghuramulu [30].

Extraction

5 grams of the well-ground sample, was added to 100 ml of 2N HCL and the mixture was shaken well for about 2 hours in a mechanical shaker. The mixture was transferred to the same beaker and weighed. It was then boiled for about 15 minutes and cooled. The mixture was adjusted to the previous weight with distilled water, and the volume was made up to 100 ml with 2N HCL, shaken well and filtered.

To 25 ml of the filtrate 5 ml of phosphoric tungstate reagent was added stirred well and kept overnight. The next day, it was centrifuged and filtered. To 20 ml of the filtrate, 2 to 3 drops of methyl red was added and neutralised with ammonia. Then 5 ml of calcium chloride buffer was added and stirred well. The mixture was allowed to stand overnight at the end of which it was filtered through Whatman 40 or 44 filter paper and washed till free from chloride using distilled water. (Silver nitrate test). The precipitate along with the filter paper was transferred to the same beaker and some distilled water was added followed by 5 ml of 2N H_2SO_4. The mixture was heated to 80°C over a burner and titrated against 0.01N $KMnO_4$.

Calculation

$$1\text{ml of } 0.01\text{N } KMnO4 = 0.045 \text{ mg oxalic acid}/100 \text{ gram sample} = \frac{(\text{Titre value} \times \text{Normality of } KMnO4 \times \frac{0.45}{0.01} \times \text{Dilution factor}}{\text{weight of the sample}} \times 100$$

Estimation of Phytates

Phytic acid content was determined by the method of Davies and Reid [32].

Extraction

One gram of finely ground sample was extracted with 25ml of 0.5N nitric acid for 3 hours with continuous shaking in a shaker. After proper shaking, it was filtered through Whatman No. 1 filter paper.

Calibration of Standard Graph

For plotting a standard curve different concentrations i.e., 0.4 to 1.0ml of standard phytic acid were taken and made up to 1.4ml with water.

Estimation

Into three tubes 1.0ml of the filtrate was taken and diluted with distilled water to a final volume of 1.4ml. Then to the standard sample tubes, 1 ml of ferric ammonium sulphate solution was added. The tubes were mixed thoroughly and placed in a boiling water bath for 20 minutes. After cooling down to room temperature under running water, 5 ml of isoamyl alcohol was added. Immediately after mixing the tubes by inversion method 0.1 ml of Ammonium thiocyanate solution was added. The tubes were shaken well and then the tube contents were centrifuged at 3000 rpm for 10 minutes. The alcoholic layer was separated with a Pasteur pipette.

Finally, the colour intensity of the alcoholic layer was read at 465 nm against amyl alcohol blank exactly after 15 minutes of the addition of ammonium thiocyanate. The extinction of 465nm in the amyl alcohol layer was inversely proportional to phytate concentration. The readings obtained were plotted against different concentrations of the standard and the concentration of the sample was intercepted from the graph.

Estimation of Polyphenols

Polyphenolic substances were estimated in the sample of the Folin Denis method (AOAC, 1984) [33] by weighing 200mg defatted sample in a 250ml round bottom flask. To this 100ml of methanol–HCL was and the contents were refluxed for 2 hours and allowed to cool. The extract was filtered through Whatman 40 filter paper into a 100ml volumetric flask and the volume was made up with methanol-HCL after a few washings. The extract was taken for the estimation of polyphenols.

Standard curve

Pipetted out 0.2, 0.4, 0.6, 0.8 and 1ml of standard tannic acid solution in 20ml test tubes. Volume was made up to 8.5ml with water. To this 0.5ml Folin Denis reagent was added and mixed using cyclomixer. To all the tubes 1ml of saturated sodium carbonate solution was added and mixed well using cyclomixer and allowed to stand for 30 minutes and absorbance was read at 760 nm.

To 1.0 ml of the extract 7.5 ml water was added followed by 5.0 ml Folin-Denis reagent and 1.0 ml Sodium Carbonate and mixed well using cyclomixer and allowed to stand for 10 minutes and absorbance was read at 760nm. Using the standard curve the results were calculated as mg tannic acid equivalent /g sample.

Estimation of Tannins

Tannins were estimated by the Vanillin-Hydrochloric acid method [34].

Extraction

Five hundred milligrams of defatted sample was weighed and transferred to a centrifuge tube. To this 10ml of acidic – methanol (1%) was added and shaken for 20mins on a shaker. The tubes were centrifuged for 10 minutes and the supernatants were transferred to a 25-volumetric flask.

To the residue in the centrifuge tubes, 5 ml of acidic methanol (1%) was added and shaken for 20 minutes. The tubes were centrifuged for 10 mins and the supernatants were transferred to the first extraction. The volume was made up to 25ml and mixed well.

Calibration of the Standard Curve

The standard solution was taken in the volume of 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7 and 0.8 in different test tubes which corresponds to 0.2, 0.4, 0.6, 1.0, 1.2, 1.4 and 1.6mg of standard. The volume was made up to 1 ml in all the tubes. 5ml of vanillin-HCL reagent was added and the intensity of the colour developed was read in the calorimeter at 500nm against the sample blank. The sample blank was prepared by the addition of 5 ml of 4% HCL in methanol to 1 ml of the sample solution. 1 ml of extract was taken in triplicated tubes. To this 5ml of freshly prepared vanillin-HCL regent was added slowly and the intensity of the colour developed was read in the calorimeter at 500nm. The absorbance was plotted against different concentrations of standard and the concentration of the sample was intercepted from the graph.

Calculation

The absorbance of the sample blank was subtracted from that of the sample. The absorbance reading was converted in the concentration of catechin/ml and catechin equivalents (CE%) were calculated as follows:

$$\mathrm{CE}(\%) = \frac{1\ (\mathrm{mg})\ \text{Catechin X Volume made}}{\text{Volume of extract taken X weight of the sample}} \times 100$$

Statistical Analysis

Mean values were calculated and variation was found by calculating the standard deviation. Student's 't' test was applied to find the significant difference among the samples. Statistical analysis was done to find out the significant difference among the variations of the groups by applying analysis of variance (ANOVA) [35].

Preparation of Dosa

South Indian standard dosa was prepared using rice and black gram dhal in a ratio of 2:1. The weighed ingredients were soaked for 12 hrs and then ground to a smooth paste. Salt was added to taste. The batter was fermented at 37°C for 12 hrs and dosas were prepared. For Pearl millet and Italian millet dosas, Pearl millet and Italian millet were used instead of rice and the procedure as above was followed.

Sensory Evaluation

Rice, Pearl millet and Italian millet dosas were code-numbered and presented randomly to a panel of 10 panellists (adolescent girls) who scored them for hardness, stickiness, appearance, odour and taste using the 5-point Hedonic rating, in which the highest score corresponded to the highest perception of quality (1- dislike very much and 5- like very much). The panellists rinsed their mouths before testing each sample.

Results

Mineral Composition

Calcium, iron and phosphorus content of Pearl millet and Italian millet in the raw sample and at 12 hrs of fermentation period is given in Table 1 and Figure 1&2. The calcium content of Pearl millet and Italian millet was estimated to be 116.12 ±0.098 and 185.97 ±0.108 mg%. At 12 hrs of fermentation, the calcium content was 112.7 ±0.088 and 159.12 ± 0.139 mg% respectively which is significantly lower than that present in the raw sample. It has been noted that the period of fermentation showed a considerable reduction in the calcium content. Raw Pearl millet and Italian millet were found to contain 24.93 ±0.046 and 22.43 ±0.046 and at 12 hrs of fermentation, it was 17.49 ±0.011and 17.47 ± 0.046 mg%. A marked decrease was seen in the iron samples at 12 hrs fermentation. The phosphorus content of Pearl millet and Italian millet was found to be 292.07 ±0.020 and 247.13 ±0.021 mg% in the raw sample while the 12 hrs fermented sample showed 240.13 ±0.088 and 269.97 ± 0.177 mg%. A noticeable increase of phosphorus was noted on fermentation in both the millets.

Table 1: Mineral Composition of Raw and Fermented 12 hrs in Pearl Millet and Italian Millet (Mean±SD)

Sample	Treatment	Calcium	Iron	Phosphorus
Pearl millet	**Raw**	116.12 ±0.098	24.93 ±0.046	292.07 ±0.020
	Fermented 12 hrs	112.7 ±0.088	17.49 ±0.011	240.13 ±0.088
Italian Millet	**Raw**	185.97 ±0.108	22.43 ±0.046	247.13 ±0.021
	Fermented 12 hrs	159.12 ± 0.139	17.47 ± 0.046	269.97± 0.177

(All values are expressed as mg/100g on dry weight basis)

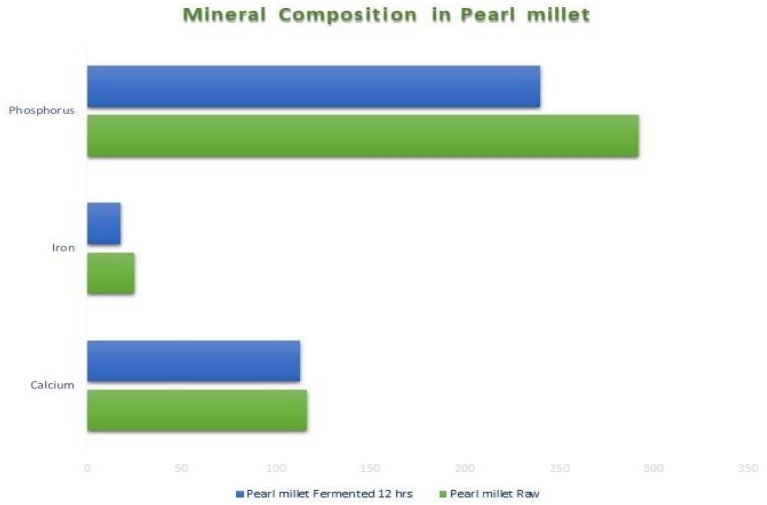

Figure 1. Graphical Representation of the Mineral Composition of Raw and Fermented 12 hrs in Pearl Millet

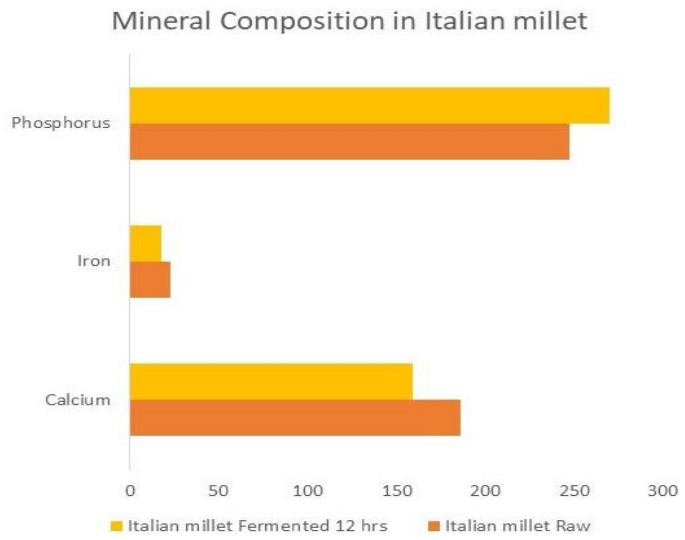

Figure 2: Graphical Representation of the Mineral Composition of Raw and Fermented 12 hrs in Italian Millet

Mineral Extractability

The extractability of Calcium, Iron and Phosphorus in Pearl millet and Italian millet in the raw sample and at 12 hrs of fermentation period is given in Table 2 and Figures 3 & 4. The extractability of calcium in the raw Pearl millet and Italian millet was found to be 60.03 ±0.044 and 25.7 ± 0.17. On fermentation for 12 hrs, it increased significantly to 85.67 ±0.038 and 66.66 ± 0.049 respectively. Raw Pearl millet and Italian millet showed an iron extractability of 50.12 ±0.084 and 66.66 ± 0.049 while on fermentation the extractability increased considerably to 71.42 ±0.00 and 85.67 ± 0.038 respectively. The extractability of phosphorus was estimated to be 60.13 ±0.015 (raw Pearl millet), 66.1 ±0.079 (raw Italian millet), 72.27 ±0.123 (12 hrs fermented Pearl millet sample) and 86.27 ± 0.098 (12 hrs fermented Italian millet sample). The extractability of all minerals i.e. calcium, iron and phosphorus improved remarkably on fermentation.

Table 2: Mineral Extractability of Raw and Fermented 12 hrs in Pearl Millet and Italian Millet (%) (Mean±SD)

Sample	Treatment	Calcium	Iron	Phosphorus
Pearl millet	**Raw**	60.03 ±0.044	50.12 ±0.084	60.13 ±0.015
	Fermented 12 hrs	85.67 ±0.038	71.42 ±0.00	72.27 ±0.123
Italian Millet	**Raw**	25.7 ± 0.17	66.66 ± 0.049	66.1 ±0.079
	Fermented 12 hrs	66.66 ± 0.049	85.67± 0.038	86.27± 0.098

(All values are expressed on dry weight basis)

Table 2a: Comparison of the Mineral Extractability of Raw and Fermented Sample of Pearl Millet and Italian Millet

	Calcium		Iron		Phosphorus	
	Pearl millet	**Italian millet**	**Pearl millet**	**Italian millet**	**Pearl millet**	**Italian millet**
Fcal	7266.858	142248.2	217687.0	2895.827	2892.245	58042.25
Ftab (P<0.05)	5.143	5.143	5.143	5.143	5.143	5.143
(P<0.01)	10.925	10.925	10.925	10.925	10.925	10.925
Sem	0.004	0.01	0.003	0.139	0.167	0.007
CD (P<0.05)	0.100	0.158	0.086	0.591	0.648	0.132
CD (P<0.01)	0.162	0.256	0.140	0.956	1.0048	0.214

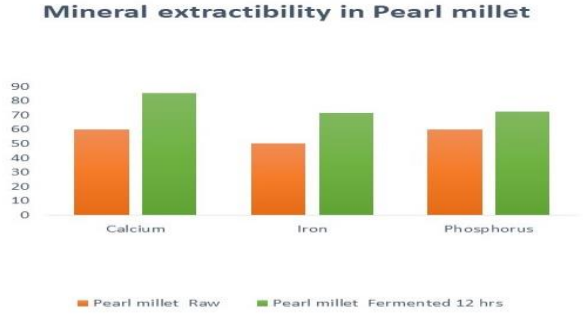

Figure 3: Graphical Representation of Extractability of Minerals in Raw and Fermented 12 hrs in Pearl Millet

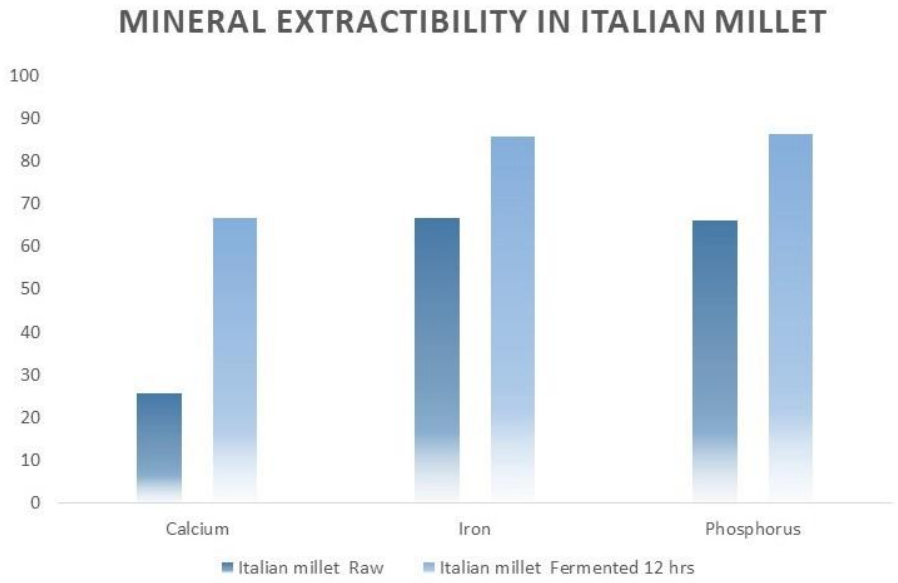

Figure 4: Graphical Representation of Extractability of Minerals in raw and Fermented 12 hrs in Italian Millet

Antinutritional Factors

Oxalates, phytates, polyphenols and tannins were estimated in Pearl millet and Italian millet raw and fermented samples and the values are given in Table 3 and Fig. 5 & 6. Oxalates in the Pearl millet and Italian millet were found to be 50.76 ±0.00 and 13.536 ± 0.00 in the raw sample and 43.98 ±0.015 and 10.152 ± 0.00 (12 hrs fermented sample). Phytates and polyphenols in the raw Pearl millet and Italian millet were estimated to be 752 ±0.03 and 700 ± 0.05 and 620 ± 0.12 (phytates) 190.06 ±0.088 and 130.04 ± 0.00 (polyphenols) respectively. A remarkable decrease was noticed in the phytates and polyphenols at 12 hours of fermentation which was found to be 650 ±0.04 and 620 ± 0.12 (phytates) 100.016 ±0.044 and 100.06 ± 0.031 (polyphenols). Tannins were not detected in any of the samples. The process of fermentation seemed to impact the antinutritional factors in millets.

Table 3: Antinutritional Factors of raw and Fermented 12 hrs in Pearl Millet and Italian Millet (Mean±SD)

Sample	**Treatment**	**Oxalates**	**Phytates**	**Polyphenols**	**Tannins**
Pearl millet	**Raw**	50.76 ±0.00	752 ±0.03	190.06 ±0.088	ND
	Fermented 12 hrs	43.98 ±0.015	650 ±0.04	100.016 ±0.044	ND
Italian Millet	**Raw**	13.536 ± 0.00	700 ± 0.05	130.04 ± 0.00	ND
	Fermented 12 hrs	10.152± 0.00	620± 0.12	100.06± 0.031	ND

(All values are expressed as mg/g on dry weight basis ND – Not detected)

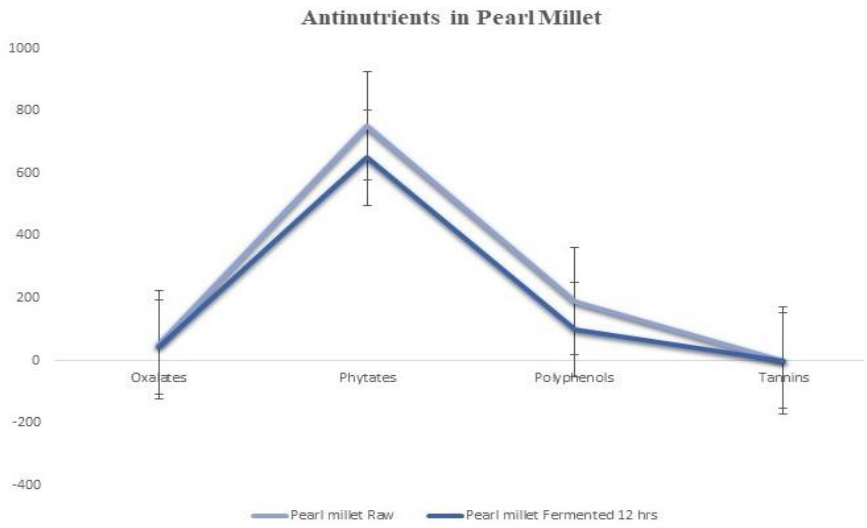

Figure 5: Graphical Representation of Antinutrients in Raw and Fermented 12 hrs in Pearl Millet

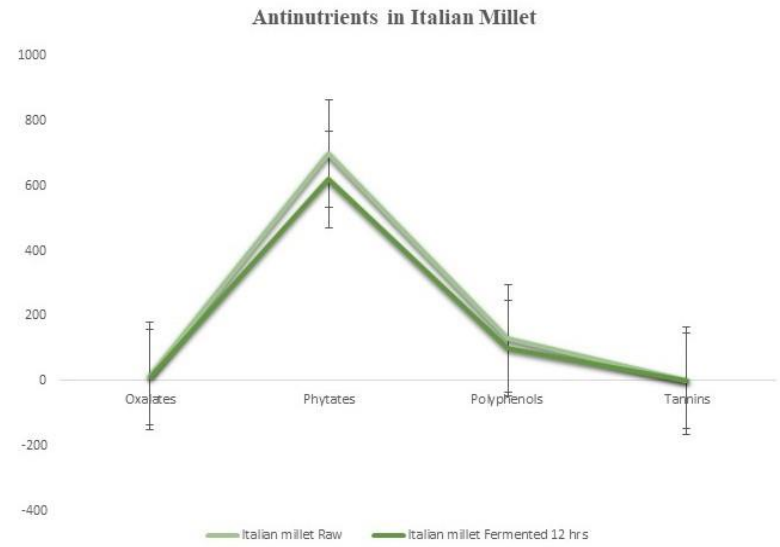

Figure 6: Graphical Representation of Antinutrients in raw and Fermented 12 hrs in Italian Millet

Sensory Evaluation of Dosa

The acceptability of the Fermented product – Pearl millet and Italian millet Dosa was studied. Sensory evaluation of Standard or rice dosa and millet dosas (Pearl millet and Italian millet) were assessed for the following qualities – hardness, stickiness, appearance, odour and taste (Table 4). The texture or hardness of Standard dosa and Italian millet dosa were found to be comparatively softer than the Pearl millet dosa. The stickiness of the three dosas showed that the standard dosa was stickier than the other two which were medium sticky. The porosity of the dosas showed that the standard and Italian millet dosas were more porous than the Pearl millet dosa. Standard dosa and Italian millet dosa were found to exhibit an accepted good odour while the Pearl millet dosa had a slightly off odour. Overall the acceptability of the dosas was good with Italian millet dosa more acceptable than Pearl millet dosa (Figures 7 & 8).

Table 4: Acceptability of the Fermented Product Dosa (Mean±SD)

Sensory Quality	Standard Dosa	Pearl millet Dosa	Italian millet Dosa
Hardness	4.2 ± 0.20	3.6 ± 0.33	4.0 ± 0.80
Stickiness	4.1 ± 0.35	3.3 ± 0.45	3.7 ± 0.002
Appearance	4.6 ± 0.58	3.8 ± 0.08	4.6 ± 0.09
Odour	4.7 ± 0.11	3.4 ± 0.06	4.5 ± 0.10
Taste	4.7 ± 0.17	3.5 ± 0.18	4.5 ± 0.12

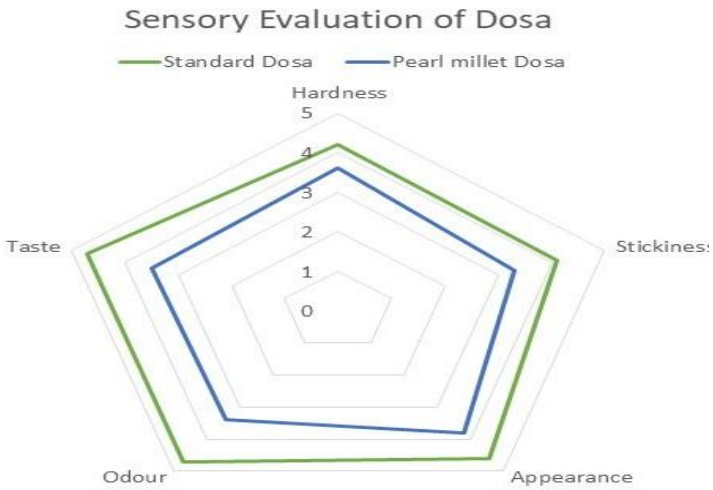

Figure 7: Graphical Representation of Sensory Evaluation of Standard Dosa and Pearl Millet Dosa

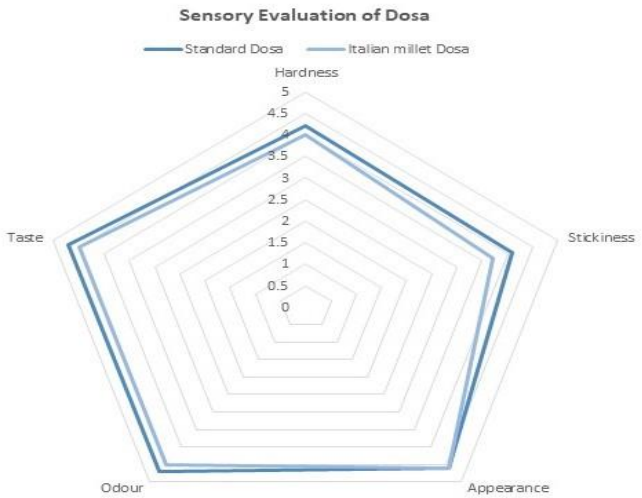

Figure 8: Graphical Representation of Sensory Evaluation of Standard Dosa and Italian Millet Dosa

Discussion

Nutritionally and economically millets are superior to staples like rice and wheat. They are widely used in several parts of the world in varied forms. Millets are small-seeded grains, rich in protein, fat, fibre, calcium, iron, zinc, magnesium and B vitamins [36]. However, millets also contain certain antinutrients like phytates, tannins and polyphenols. Studies suggest that processes like soaking, germination and fermentation of millets can be a solution to the reduction of mineral inhibiting factors and consumption of such processed millets results in mitigating several nutritional problems [37, 38].

This study shows that the minerals, calcium and iron, content in raw samples of Pearl millet and Italian millet decreased considerably in millet samples fermented for 12hrs. Akinyele and Akinlosotu, 1991 [39] report a decrease in mineral content in fermenting which might be due to the utilization by microbes. However, phosphorus content in Pearl millet decreased on fermentation but in Italian millet, a considerable increase was noticed. This increase coincides with the increase (96.2%) shown by Akinyele and Akinlosotu [39].

The extractability of minerals, calcium, iron and phosphorus, showed a significant increase in the fermented samples ($P<0.1$) (Table 2a) than in the raw Pearl millet and Italian millet. The increase in calcium bioavailability may be attributed to the decrease in the phytic acid content during fermentation possibly through hydrolysis by phytase of the fermenting microflora [40]. Fermentation brought about a significant decrease ($P<0.1$) in the antinutrient content – oxalates, phytic acid and polyphenols. The decrease in phytic acid and polyphenols can be attributed to the enzymatic hydrolysis of phytic acid by the endogenous phytase [41] and the activity of polyphenol oxidase of the microbes [42].

The taste of the dosas was good with standard and Italian millet dosas showing a slightly sour and acceptable taste while the Pearl millet dosa was sourer than the other two. Overall, a comparison of the dosas can be interpreted as the millet dosas are acceptable with Italian millet dosa closer in texture, taste and odour to the standard dosa and the Pearl millet dosa is a little harder, slightly off odour and sourer than the standard dosa.

The present investigation was undertaken to study the effect of fermentation on improving the mineral bioavailability and decreasing antinutrients in Pearl millet and Italian millet and the acceptability of millet-fermented products (Pearl millet dosa and Italian millet dosa) by adolescent girls which could be crucial to decreasing micronutrient deficiencies in young girls. It is conclusive that millet consumption thereby helps to mitigate complications and even decrease mortality rates arising due to micronutrient deficiencies in young child-bearing females.

Conclusion

Investment in adolescent health is imperative as a considerable portion of the world's population consists of the younger generation between 10 – 19 years old and a sizeable portion of them are a burden of disease and injury. Poor health, risky behaviours, eating problems, etc. can affect negatively their health in adult life. Child-bearing adolescents are at a greater risk because it is not only their lives that are at stake but they risk the life of the newborn too. Such incidents result in low-birth-weight babies, poor growth, lowered immunity and infant mortality. Agarwal et al. report a high prevalence of calcium and iron deficiency in India. Several factors can be attributed to micronutrient deficiencies – dietary insufficiency, improper cooking methods, lifestyle, income, etc. One of the primary reasons for deficiency disorders is the consumption of plant-based diets which contain antinutritional factors that inhibit mineral absorption and utilization.

This study proves that the mineral availability of Pearl millet and Italian millet

improved when fermented. The organoleptic properties of fermented millet product, dosa, are acceptable by adolescent girls. Hence, it can be concluded that millet can be the key to alleviating micronutrient deficiencies (calcium and iron) among adolescent girls.

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Conflict of Interest

The authors declare no conflict of interest.

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