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Antioxidant properties of germinated millets (*Eleusine coracana* & *Pennisetum glaucum*): A Comparative study

Ekta Singh Chauhan, *Sarita

Department of Food Science & Nutrition, Banasthali University, Tonk, Rajasthan, India

Abstract
India is largest producer of millets, can be excellent option for the sake of overcoming the intense problem of food insecurity and malnutrition, the quality of diet should be given significant weightage. Finger and pearl millet, both are major one in millets, easily cultivated over the year, have excellent storage properties of the grains and high nutritive value (equal to rice and wheat), major and minor nutrients such as carbohydrate, protein, dietary fiber, calcium and iron. Finger and pearl millet have many antioxidant properties that are helpful to prevent many health problems such as lowering blood pressure, risk of heart disease, prevention of cancer and cardiovascular diseases, decreasing tumor cases etc. Changes antioxidants were followed during 48 hours germination of finger and pearl millet. There was significantly increased in finger millet from 230.9±1.01 to 262.3±1.10 mg/100g of total phenol. In pearl millet, total phenol was significantly increased from 180.6±2.05 to 240.4±1.05 mg/100g after germination process. FRAP (ferric reducing antioxidant property) was also founded in increasing trend during germinated of finger and pearl millet.

Keywords: millet, malnutrition, germination, antioxidants, total phenols, FRAP

1. Introduction
In developing countries, children develop malnutrition when introducing nutritionally inadequate complementary foods. Malnutrition is considered as a big hindrance in national development. This time food must be formulated nutritionally energy- rich, easily digestible and functional food with easy availability and cost effective properties to promote optimization of the body for future prospective. To obtain this aim, must be use local, seasonal, cost effective raw food ingredients having rich nutritional and functional properties such as cereals and millets, pulses, dairy raw material. Wheat and corn flour are commonly used by large population with health benefit properties. India is considered as hub of minor crops like millets. Millets have been found to have high nutritive values and are comparable to other major cereals like wheat and rice. Millet constitute a cheap source of protein, vitamins and minerals for poorest people whom they need vital nutrients but their utilization is limited due to the presence of anti-nutrients, poor digestibility and low palatability occurs restricted their use. To increase millet's utilization, some food processing techniques can be used at household level such as germination, popping etc. stated by Choudhury, *et al*., (2011) [4].

Food processing techniques are used to enhance nutritional quality, improve the digestibility and bioavailability of food nutrients with reducing anti-nutrients. Some food techniques are decortications, milling, soaking, cooking, germination, fermentation, malting, popping etc. Germination is a biochemical process which involves transition of a seed from dormant state to vital active state. Germination can increase protein content, mineral bioavailability and dietary fibre. It reduces anti nutrients like tannin, phytic acid content and polyphenols (Ghavidal and Prakash, 2007) [8]. Controlled germination conditions provide high quality of malt products. Malting increases the nutrient content and digestibility of food (Platel *et al*., 2010) [11]. Germination with other processing treatments to prepare malt rich nutrients can be used for preparation of several healthy and nutritional food products.

Finger millet is a very good source of micronutrient, which could be alleviate of micronutrient malnutrition in the developing countries. Finger millet (Eleusine coracana L.) is also known as ragi and mandua (India). Finger millet is a good source of nutrients especially of fiber, calcium, phosphorus, iron, zinc, potassium and other minerals. Dykes and Rooney, (2006) [6] have reported that finger millet is a very good source of variety of phenolic compounds which may have health benefits. The main polyphenols are phenolis acid and tannins (Shobana *et al*., 2013) [17] while flavonoids are present in small quanties (Rao and Muralikrishna, 2002) [13, 14]. Polyphenols has been known to impart antimicrobial (Viswanath *et al*., 2009) [22], anti-diabetic, antimutagenic properties. Along these, functional property (gelatinization) is also present in finger millet (Mathanghi and Sudha, 2012) [9].

Pearl millet (Pennisetum glaucum L.), also known as "bajra" is negligible, low demanded by people for food. It has high level of calcium, iron, zinc, lipids, and amino acids (Sade. 2009) [15] such as lysine, tryptophan, threonine and fatty acid like omega-9, omega-6 and omega-3 fatty acid ratio. The phytochemicals like tannins, phytates (Onyango *et al*., 2013) [10] act as antioxidant properties. It has low glycemic index, small amount of flavonoids are present and it is gluten free millet. Pearl millet contains phospholipids which are useful in brain functions, behavioural disorder and stress. It may have therapeutic effects in some health problems like anaemia, constipation, diarrhoea, diabetes, CVD, celiac diseases, cancer and it is referred to as anti-inflammatory and it also acts as probiotic food (Vanisha *et al*., 2011) [21].

The antioxidants like poyphenols, phytates, tannins and condensed tannins (proanthocyanidins) are present in millets. Now a days, polyphenols considered as "life span essential" due to their role in maintaining body functions and health throughout end phase of the life described by Chandrasekara & Shahidi, (2010) [2]. Devi, *et al*., (2011) [5] explained the millet polyphenols is a complex mixture of cinnamic acid derivatives and benzoic acid perform enzyme inhibitory and anti-cataractogenic

activities. Rao & Muralikrishna, (2002) [13, 14] founded that main polyphenols such as phenolic acid and tannins are present in cereals whilst flavonoids are present in small quantities. Siwela, *et al*., (2007) [19] founded that the pigment testa in the red colored is identify to have much tannin content which is located in the said tissue of the grain. Total polyphenol contents founded in wide range (0.19-3.37%) among 85 Indian finger millet varieties (Shankara, 1991) [16]. These compounds have no known direct role in nutrition but have some health friendly properties like anti-nutrients (Sripriya, *et al*., 1996) [20], anti-oestrogenic, anti-mutagenic, anti-carcinogenic, antiviral effects, anti-inflammatory, platelet aggregation inhibitory activity that might be potential benefit in minimizing and preventing the incidence of diseases (Ferguson, 2001) [7]. Amadou, *et al*., (2013) [1] explained that millets are gluten-free and excellent option for people suffering from celiac disease and also useful for those people who are suffering from artherosclerosis and diabetic heart disease. The objective of this research is to comparison study of antioxidants in finger and pearl millet influenced by germination process.

2. Materials and Methods

Selection and procurement of finger and pearl millets.

Processing of finger and pearl millets.

Preparation of processed forms

Whole raw finger millet flour (WRFMF): Finger millet seeds were thoroughly cleaned, remove foreign material and dirt. Thereafter, they were sundried and ground into fine flour or powder in a mixer and stored.

Whole raw pearl millet flour (WRPMF): Pearl millet seeds were cleaned, free from dirt and foreign matter. Then, they were sundried and ground in a mixer into fine flour and stored in another container.

Germinated finger millet flour (GFMF) & Germinated pearl millet flour (GPMF): One portion of finger & pearl millet seeds was soaked overnight. Next day, water was drained and wrapped of seeds in a muslin cloth and hung in a humid atmosphere for germination. After 48 hours germination, seeds were sundried to make moisture free. Germinated seeds were ground in a mixer and stored in container for analysis.

2.1 Antioxidant analyses

Total phenols (Singleton and Slinkard, 1977) [18]

Total free phenolics were determined using Folin-Ciocalteu reagent. Phenols react with phosphomolybdic acid in Folin-Ciocalteau reagent in alkaline medium and produce blue colored product (molybdenum blue).

2gm finely grounded sample was extracted with 5-10 ml of 80% alcohol in a pestle mortar and the homogenate was boiled in water bath for 5-10 minutes, centrifuged and supernatant was collected and volume made up to 200 ml in the same flask (T), then 1.0 ml Folin-Ciocalteu's reagent and 0.8 ml sodium carbonate (7.5%) were added into 'T' test tube. The absorbance of sample was measured at 760 nm after incubating at 30^0 C for 1.5 h.

Calculation: Results were expressed as milligram of gallic acid equivalent (GAE) per gram of fresh weight. Standard curve was drawn by plotting the absorbance against concentration of gallic acid.

Ferric reducing antioxidant property (FRAP) (Pulido *et al*., 2000) [12]

The reducing property is determined by assessing the quantitative ability to reduce of Fe^{3+} to Fe^{2+} by antioxidant abundant food sample.

0.5 g of food sample was taken and extracted with 20 ml of 80% acetone. 2.5 ml of aliquot was pipette into test tube. Then, mixed with 2.5 ml of 200Mm sodium phosphate buffer (ph6.6) and 2.5 ml of 1% potassium ferric cyanide. Incubated the mixture at 50^0 C for 20 min. Thereafter, 2.5 ml 10% trichloroacetic acid was added and subsequently centrifuged at 650 rpm for 10 minutes. 5 ml of supernatant was taken and mixed with equal volume of water. Then, 1ml of 0.1% ferric chloride was added and the absorbance was measured at 700nm against a reagent blank.

Calculation: Methanolic solution of Fe (II) was used for the preparation of the calibration curve. The FRAP value was expressed as µg of Fe (II) equivalent per milligram extract.

Statistical analysis

The statistical methods using for analysis of data in the present investigation were mean, standard deviation (SD) and student t-test.

3. Result and Discussion

Fig.5 depicts the antioxidant content of all versions of finger and pear millet. In WRFMF, total phenol content was higher in finger millet compare to pearl millet. Total phenol content was found as 1000±1.01 mg/100g in WRFMF and 755±2.05 mg/100g in WRPMF. Germination caused a significant increase in total phenols to 1106±1.10 mg/100g in GFMF and 804±3.05 mg/100g in GPMF. A significant improvement in total phenol content was counted 10.6% in germinated finger millet and 6.49% in germinated pearl millet.

Fig 1: Germinated pearl millet (48 hours)

Fig 2: Germinated finger millet (48 hours)

Ferric reducing antioxidant properties contained more in pearl millet compare to finger millet FRAP was founded as 1.53±0.03, 2.44±0.02, 2.02±0.02 and 2.36±0.12 µg/g in all versions i.e. WRFMF, GFMF, WRPMF and GPMF respectively. FRAP increased significantly in germination of both millet. FRAP of GFMF increased 59.47% and 16.83% in GPMF.

Table 1: Effects on antioxidants in raw and germinated millets (Finger and Pearl)

Antioxidants	WRFMF	WRPMF	GFMF	GPMF
Total phenols (mg/100g)	230.9±1.01	180.6±2.05	262.3±1.10	190.5±1.05
FRAP (µ/g)	1.53±0.03	2.02±0.02	2.44±0.02	2.36±0.12

WRFMF- whole raw finger millet flour, WRPMF- whole raw pearl millet flour, GFMF- germinated finger millet flour, GPMF- germinated pearl millet flour.

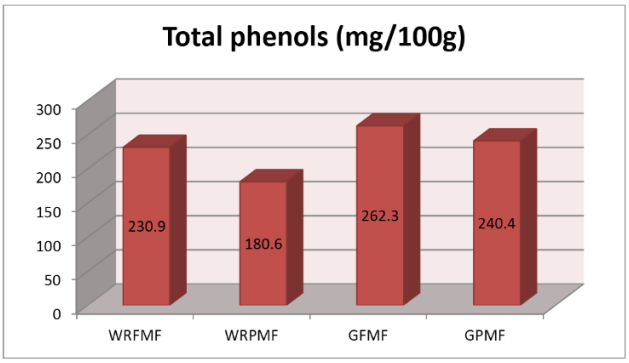

Fig 3: Mean levels of total phenols in raw and germinated finger and pearl millet

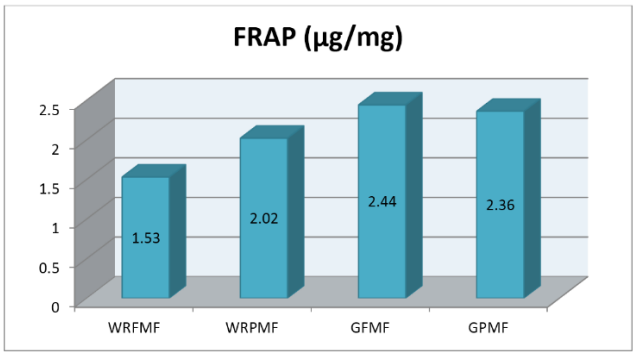

Fig 4: Mean levels of ferric reducing antioxidants properties (FRAP) in raw and germinated finger and pearl millet

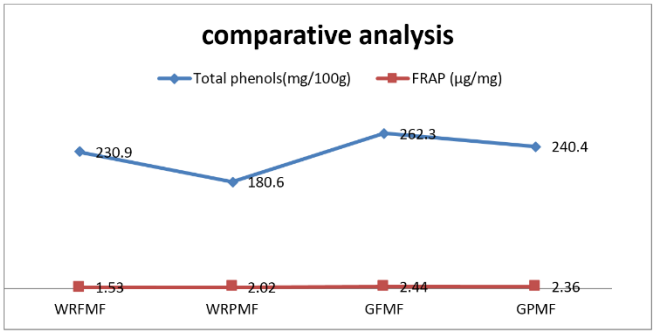

Fig 5: Comparative analysis of antioxidant in WRFMF, WRPMF, GFMF and GPMF. WRFMF- whole raw finger millet flour, WRPMF- whole raw pearl millet flour, GFMF- germinated finger millet flour, GPMF- germinated pearl millet flour.

4. Conclusion

Malnutrition is hindrance for the physical and mental development of human which is important parameter of national productivity and development. The foods also need to be low cost and nutritious too, can be consumed easily for large population. Millets are rich source of nutrient, antioxidants and nutraceutical compounds. Various food processing operations help in making millet more nutritious and palatable. Emphasis is given on the effect of germination on antioxidants such as total free phenols and ferric reducing antioxidant properties (FRAP) of finger and pearl millet. Antioxidants significantly prevent tissue damage and stimulate the wound healing process. Antioxidant effects of finger millet on the dermal wound healing process in diabetes induced rats with oxidative stress-mediated modulation of inflammation. Finger and pearl millet has great potential to substitute of cereals (wheat, rice) because similar nutritious, easily cultivated and rich in calcium and iron. This study showed that germination affects the antioxidant properties of both millets (finger and pearl). Germination increased total phenol content higher in finger millet in compare to pearl millet. Ferric reducing antioxidant properties also more increased in finger millet after germination of both millets. Thus, germination can be an essential prerequisite in harnessing the antioxidants in finger and pearl millet.

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