

Chapter 9

Millet Flours as a Vehicle for Fortification with Iron and Zinc

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Key Points

- Fortification of staples with micronutrients is a feasible strategy to combat micronutrient deficiencies.
- Cereals have been commonly used as vehicles for fortification with minerals and vitamins since more than 8 decades.
- Millets are widely grown and consumed by the lower economic segments of the population especially in the developing countries.
- In spite of their extensive consumption, millets are less explored as vehicles for fortification with minerals.
- Finger millet, sorghum, and pearl millet, which are widely grown and consumed as the staple in several parts of India were examined for their feasibility as vehicles for fortification with iron and zinc.
- These millet flours were found suitable for fortification with iron and zinc, providing significant amounts of bioaccessible minerals.
- EDTA, a known metal chelator, when included as a co-fortificant significantly improved the bioaccessibility of both iron and zinc from the fortified flours.
- Fortification of millet flours with ferrous fumarate and zinc stearate along with EDTA did not have any adverse effect on the shelf-life of the fortified flours, or on the sensory quality of the products prepared from them.
- It would be worthwhile to examine other millets consumed as a staple in several parts of the world for feasibility as vehicles for fortification with micronutrients.
- Fortification of millet flours with minerals therefore seems to be a feasible strategy to combat micronutrient deficiency.

Keywords Fortification • Millets • Iron • Zinc • EDTA

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Introduction

Deficiency of micronutrients, especially iron, iodine, vitamin A, and zinc, are widely prevalent not only in developing countries, but also in the developed countries. Micronutrient deficiencies are often known as “hidden hunger,” since they are less visible than protein energy undernutrition. For the last 2 decades, micronutrient deficiencies especially nutritional anemia, iodine deficiency disorders, and vitamin A deficiency have been a subject of concern in developing countries. Deficiency of iron is a public health problem, particularly in developing countries such as India, where 79 % of children between 6 and 35 months and women between 15 and 49 years of age are anemic [1]. In recent years, the deficiency of zinc is also being recognized as a global health problem [2]. Both iron and zinc deficiency have several functional consequences such as impairment of cognitive function, linear growth impairment, behavioral problems, mood changes, memory impairment, problems with spatial learning, and neuronal atrophy. In addition, iron deficiency anemia is found to be associated with reduced work capacity in adults, an increased risk of maternal and neonatal mortality and premature birth, and altered immune function [3, 4].

Fortification is a cost-effective method that can be used at the national level to prevent deficiency of both iron and zinc without any change in existing dietary patterns or any personal contact with the recipients [2]. Fortification of foods is often regarded as the most cost-effective long-term approach to reducing the prevalence of mineral deficiency [5]. The concept of food fortification with micronutrients was documented as early as 1923, when Switzerland introduced the iodization of salt to prevent goiter and cretinism. Rickets caused by deficiency of vitamin D in children living in the Northern Hemisphere was prevented by addition of vitamin D to infant formula and dairy products [6]. In 1941, United States was the first country to enrich wheat flour with iron and vitamins and subsequently, virtually all white wheat flour and wheat bread, most corn meal, grits, and macaroni products were fortified with iron, as were a large proportion of other cereal products. Mandatory enrichment of white wheat flour with iron was introduced in the United Kingdom and Canada in 1953 and many other countries have since introduced either mandatory or voluntary enrichment (UK) [3]. Fortification with iron has been successfully adopted for wheat flour, rice, sugar, salt, milk, fish sauce, and curry powder. Other foods like wheat biscuits, wheat flour noodles, and maize meal have also been tried [7–15].

In India, food fortification was used in the early years as a strategy to improve the intake of macronutrients, in order to combat protein energy malnutrition [6]. In this direction, wholesome foods were blended to improve the protein content. The Indian multipurpose food is one such blend of edible peanut flour and chickpea flour that provides high protein with added minerals and vitamins. Blending of wheat flour with peanut flour to raise the protein content was another strategy tried in India [6].

While fortification of wheat flour, sugar, and salt with iron is a common strategy in industrialized countries [6], fortification of millet flours with minerals has gained little attention. Millets are used chiefly as food grains in Africa, Eastern Europe, China, India, and other Asiatic countries [16]. In developing countries such as India where a majority of the population consume plant-based foods, cereals, millets, and pulses are major dietary sources of iron and zinc.

In the Indian scenario, this country is the largest producer and consumer of millets, sharing nearly 60 % of the area and output of the millets grown in the world [17]. According to recent statistics, the production of pearl millet in India is about 8.89 million tons, that of sorghum is 7.25 million tons, and of coarse cereals including finger millet is 40.04 million tons [18].

Finger millet is predominantly cultivated in Karnataka, Andhra Pradesh, and Tamil Nadu, where this millet is the staple to a large section of the rural population. Finger millet is also grown in the Himalayas, but its cultivation there is scattered [19].

Pearl millet is consumed predominantly in western and central states of India, and is the staple mainly in Gujarat and Rajasthan. Across income classes, pearl millet is consumed mainly by the low and middle income groups; about 46 % of pearl millet in urban India is consumed by the low income groups.

Sorghum is primarily produced in Maharashtra and southern states of Karnataka and Andhra Pradesh, these three states together accounting for nearly 80 % of the all-India production. Madhya

Pradesh, Gujarat, and Rajasthan are the other states producing sorghum. India is the third largest producer of sorghum in the world. The low income consumers account for 35 and 49 % of sorghum consumption in rural and urban areas of India respectively [20].

In view of the extensive production and consumption of millets especially among the lower economic groups of the population, their fortification with minerals such as iron and zinc is certainly a rational strategy to enhance the intake of these minerals thereby reducing their deficiency.

Millet Flours as Carriers of Iron

While cereal flours are common vehicles for fortification with micronutrients, millets are less explored in this context. Fortification of millet flours with iron would be a feasible public health strategy to combat iron deficiency, since millets form the staple for large segments of the population, especially the poorer sections, in developing countries. Finger millet (*Eleusine coracana*) is widely consumed in the southern parts of India and is a good source of minerals. Sorghum (*Sorghum bicolor*) is an important food crop providing energy, protein, vitamins, and other nutrients to millions people living in semi arid tropical regions of the world [21]. Although millets such as pearl millet (*Pennisetum glaucum*), finger millet, and sorghum are generally good sources of trace minerals [22], the bioavailability of these minerals may be limited because of the presence of high levels of phytates and fiber which are major inhibitors of bioavailability of iron and zinc [23]. Inclusion of promoters of iron absorption in addition to the mineral would thus be beneficial in providing higher amounts of bioavailable iron.

Finger millet, sorghum, and pearl millet were recently examined for feasibility of fortification with iron [24, 25]. Initially, ferrous fumarate and ferric pyrophosphate added at levels to provide 6 mg iron per 100 g flour, were examined for fortification of finger millet flour [24]. Both the salts were found to be equally effective with respect to iron bioaccessibility; however, the bioaccessible iron content declined in the ferric pyrophosphate fortified flour after 30 days of storage. Therefore ferrous fumarate was subsequently used as the fortificant, and was added to the millet flours a level that provided 60 mg iron per kg flour. EDTA, a known metal chelator, was added along with ferrous fumarate at levels equimolar to the added iron. The bioaccessible iron content of the fortified flours was determined by the in vitro simulated gastrointestinal digestion method, involving equilibrium dialysis [24]. Bioaccessibility of iron was determined periodically from the fortified flours stored at ambient temperature for a period of 60 days [24, 25].

Bioaccessibility of iron from the fortified millet flours: Fortification of the millet flours with ferrous fumarate to provide 6 mg of iron per 100 g of the flour brought about an increase in the bioaccessible iron content of the fortified flours. Finger millet flour had a bioaccessible iron content of 0.23 mg/100 g, which increased to 0.29 mg/100 g upon fortification (27 % increase). Similarly, fortification of sorghum and pearl millet flours brought about 41–44 % increase in the bioaccessible iron content (Table 9.1). There was no significant decline

Table 9.1 Bioaccessible iron content of iron-fortified millet flours

Flour	Bioaccessible iron (mg/100 g)		
	Days of storage		
	0	30	60
Finger millet	0.23	0.21	0.20
Finger millet + iron	0.29 ^a	0.26 ^a	0.24 ^a
Sorghum	0.39	0.37	0.35
Sorghum + iron	0.56 ^a	0.53 ^a	0.42 ^{a,b}
Pearl millet	0.39	0.35	0.32 ^b
Pearl millet + iron	0.55 ^a	0.53 ^a	0.44 ^{a,b}

Adapted from [20, 21]

Values are average of five replicates

^aSignificantly higher than control (unfortified grain)

^bSignificantly lower than initial (day 0) value

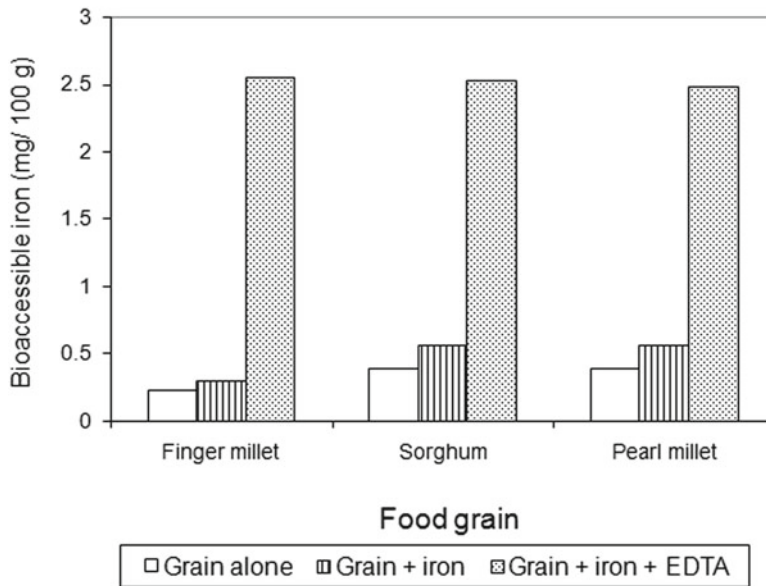


Fig. 9.1 Effect of EDTA on the bioaccessibility of iron from iron-fortified millet flours

in the iron bioaccessibility from the finger millet and sorghum flour upon storage, but the bioaccessible iron content in the pearl millet flour reduced significantly over the period of storage both in the unfortified as well as the fortified flour (Table 9.1). Thus, addition of iron increased the bioaccessible iron content to different extents in the three millet flours. Despite similar total iron content, the bioaccessibility of the native iron from sorghum flour was higher than that from finger millet flour. Fortification of these flours with iron at the same level significantly enhanced the bioaccessible iron content of sorghum flour, but that of finger millet flour was only marginally increased. Both sorghum and pearl millet flour had similar bioaccessible iron content, in spite of a higher amount of total iron in the latter. This could probably be attributable to the higher amounts of inhibitory factors such as phytate and tannin present in pearl millet flour [26].

Influence of EDTA on iron bioaccessibility from the fortified millet flours: Addition of EDTA at levels equimolar to the added iron significantly increased the bioaccessibility of the iron from the fortified millet flours. EDTA brought about a six to eightfold increase in the bioaccessible iron content of the fortified flours (Fig. 9.1). However, this increase tended to decline over the period of storage. Incidentally, EDTA also significantly increased the bioaccessibility of iron from the unfortified flours. Thus, EDTA successfully countered the negative effects of the inhibitory factors inherently present in the millet flours. In spite of the decline in bioaccessible iron content during storage, it continued to be much higher than that of the unfortified flours as well as with the flours fortified with iron alone, even at the end of 60 days of storage [24, 25].

Effect of fortification of millet flours with iron on the bioaccessibility of the native zinc: Iron–zinc interaction is a matter of concern in the case of iron fortification; since the molar ratio of iron to the inherent zinc will be altered several fold as a result of addition of exogenous iron. However, the addition of exogenous iron to millet flours did not negatively influence the bioaccessibility of the native zinc, despite a significant decrease in the Zn:Fe molar ratio as a result of iron fortification. On the other hand, the addition of EDTA as a co-fortificant significantly enhanced the bioaccessibility of the native zinc from all the millet flours examined [24, 25].

Shelf-life of the iron-fortified millet flours: Fortification of finger millet, sorghum, and pearl millet flours did not seem to affect the keeping quality of the flour under ambient conditions up to a period of 60 days, as indicated by their moisture and free fatty acid content that were monitored during the period of storage [24, 25].

As mentioned earlier, millet flours are less explored as vehicles for fortification with micronutrients, and the two reports mentioned above have suggested that millet flours can indeed be employed as carriers of iron. Such qualitatively rich flours can be a part of the nutrition intervention programs to overcome the deficiency of iron.

Millet Flours as Carriers of Zinc

The importance of zinc in human health has been widely recognized in recent years, and zinc deficiency is included as a major risk factor to the global burden of diseases along with iron, vitamin A, and iodine deficiencies since 2002 [26]. Although the major source of zinc in our diet is animal foods, a majority of the population in developing countries derive this micronutrient from plant foods, especially grains. Staple foods in developing countries include cereals and legumes, which are the main sources of zinc for most of the population but even if net zinc intake appears adequate, compromised zinc status is common [27]. Recent evidence from National Food Balance Sheets suggests that the food supply of nearly 50 % of the global population is low in absorbable zinc because of limited availability of animal products and a higher intake of cereals and legumes [28]. Thus, fortification of staple food grains with zinc may be a suitable approach to prevent zinc deficiency in developing countries.

Bioaccessibility of zinc from the fortified millet flours: In a recent study, finger millet, sorghum, and pearl millet that are commonly consumed as the staple in several parts of India, were fortified with zinc. Two zinc salts, namely zinc stearate and zinc oxide were initially used for fortifying finger millet flour with zinc at levels that provided 5 mg zinc/100 g flour. Zinc stearate was found to provide significantly higher amounts of bioaccessible zinc as compared to zinc oxide [29]. Fortification of the flours of finger millet, sorghum, and pearl millet with zinc stearate to provide 5 mg zinc per 100 g flour brought about a significant increase in the bioaccessible zinc content [29, 30]. The native zinc content in finger millet, sorghum, and pearl millet flours was 1.72, 1.68, and 4.04 mg/100 g, respectively, and the bioaccessible zinc content was 0.18, 0.37, and 0.69 mg/100 g, respectively. Addition of zinc stearate at the level mentioned above increased the bioaccessible zinc content to 0.49, 0.61, and 0.79 mg/100 g in finger millet, sorghum, and pearl millet flours, respectively. These levels remained stable during a 60-day period of storage in finger millet and sorghum flours, but tended to decline slightly after 30 days of storage in the pearl millet flour (Table 9.2). Among the three millet flours, pearl millet flour had the highest native as well as bioaccessible zinc content, but despite fortification with zinc stearate which more than doubled the native zinc content, the increase in bioaccessible zinc content was only marginal (0.70–0.79 mg/100 g). Similarly, despite comparable zinc content in fortified finger millet and sorghum flour, the latter provided higher amount of bioaccessible zinc (0.61 mg/100 g). Thus, it is evident that fortification with zinc has

Table 9.2 Bioaccessible zinc content of zinc-fortified millet flours

Flour	Bioaccessible zinc (mg/100 g)		
	Days of storage		
	0	30	60
Finger millet	0.18	0.17	0.15
Finger millet + zinc	0.49 ^a	0.45 ^a	0.44 ^a
Sorghum	0.37	0.33	0.32
Sorghum + zinc	0.61 ^a	0.59 ^a	0.58 ^a
Pearl millet	0.70	0.68	0.63
Pearl millet + zinc	0.79	0.77	0.72

Adapted from [25, 26]

Values are average of five replicates

^aSignificantly higher than control (unfortified grain)

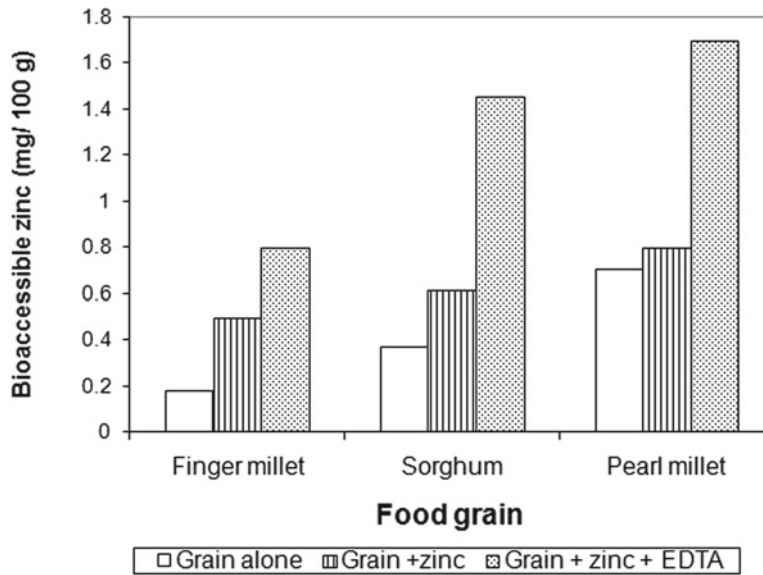


Fig. 9.2 Effect of EDTA on the bioaccessibility of zinc from zinc-fortified millet flours. Values are average of five replicates

a more pronounced effect on sorghum as compared to either finger millet or pearl millet flour with respect to the increase in bioaccessible zinc content. Further, the bioaccessible zinc content in the fortified sorghum flour was more stable as compared to that in pearl millet flour, which tended to decline after a period of 30 days [29, 30].

Influence of EDTA on zinc bioaccessibility from the fortified millet flours: As in the case of iron-fortified millet flours, addition of EDTA at levels equimolar to the added zinc significantly enhanced the bioaccessible zinc content of all the three millet flours examined (Fig. 9.2). The bioaccessible zinc content was increased to an extent of 1.6-fold in finger millet, while in sorghum and pearl millet flours there was more than twofold increase in the same. In addition to enhancing the bioaccessibility of zinc, EDTA also countered the slight reduction in the same during storage that was seen in finger millet and sorghum flours where EDTA was not included; however, the decrease in bioaccessible zinc content of fortified pearl millet flour on storage beyond 30 days was not countered by the addition of EDTA. As in the case of iron fortification, EDTA also increased the bioaccessibility of the native zinc from all the three millet flours. A significant decrease in the iron:zinc ratio as a result of fortification of the millet flours with zinc did not adversely affect the bioaccessibility of the native iron in these flours.

Effect of fortification of millet flours with zinc on the bioaccessibility of the native iron: The iron:zinc ratio of the zinc-fortified millet flours was significantly reduced as a result of addition of exogenous zinc. However, this reduction did not result in any compromise in the bioaccessibility of the native iron from the zinc-fortified millet flours. On the other hand, inclusion of EDTA as a co-fortificant was beneficial in increasing the bioaccessibility of the native iron [29, 30]. Thus, addition of exogenous zinc to the millet flours does not have any negative influence on the bioaccessibility of the inherent iron.

Shelf-life of the zinc-fortified millet flours: Moisture and free fatty acid contents of the stored fortified flours indicated that the fortified finger millet and sorghum flours can be stored up to 60 days under ambient conditions. Pearl millet flour seems to have limited shelf-life as indicated by the FFA content, which increased marginally at the end of 60 days of storage [29, 30]. Thus, millet flours seem to be suitable for fortification with zinc, and inclusion of EDTA as a co-fortificant further improved the bioaccessibility of zinc from these flours.

Double Fortification of Millet Flours with Iron and Zinc

In the view of widespread multiple mineral deficiencies, it would be appropriate to fortify staple foods with two or more minerals simultaneously. In this context, finger millet and sorghum were double fortified with iron and zinc [31]. Ferrous fumarate and zinc stearate were added at levels that provided 6 mg iron and 5 mg zinc per 100 g of flour, respectively. EDTA was used as a co-fortificant, and was added at a level equimolar with the added iron.

The bioaccessible iron content of the finger millet and sorghum flour was 0.33 and 0.37 mg Fe/100 g, respectively (Table 9.3). When the flour was double fortified including iron at the level of 6 mg Fe/100 g flour along with EDTA the bioaccessible iron increased to 2.39 and 2.63 mg Fe/100 g flour in finger millet and sorghum flour respectively. Thus fortification of the millet flours with ferrous fumarate and EDTA led to a significant (sevenfold) increase in bioaccessible iron content of both the millet flours. There was a marginal (13.8 %) decline in the bioaccessible iron content in of the fortified sorghum flour stored for 60 days, while that in the fortified finger millet flour was negligible [31].

Double fortification of the millet flours also resulted in an increase in the bioaccessibility of zinc, the same being enhanced from 0.22 to 0.83 mg/100 g in finger millet flour and from 0.39 to 1.63 mg/100 g in sorghum flour, which amounts to about 3.5- to 4-fold increase (Table 9.4). However, there was a significant decline in the bioaccessible zinc content from the fortified millet flours on the 60th day of storage, the extent of this decline being 14 and 33 % in the finger millet and sorghum flours, respectively. Despite this decline, the bioaccessible zinc content of the fortified millet flours remains fourfold higher than the unfortified flours in either case [31].

Fortification with both iron and zinc would alter the molar ratios of these minerals. Significant alteration of the molar ratios of iron and zinc in the millet flours as a result of double fortification did

Table 9.3 Bioaccessible iron content (mg/100 g) of the double-fortified millet flours

Flour	Bioaccessible iron (mg/100 g)		
	Days of storage		
	0	30	60
Finger millet	0.33	0.31	0.29
Fortified finger millet	2.39 ^a	2.27 ^a	2.06 ^{a,b}
Sorghum	0.37	0.36	0.35
Fortified sorghum	2.63 ^a	2.61 ^a	2.57 ^a

Adapted from [27]

Values are average of five replicates

^aSignificantly higher than control (unfortified grain)

^bSignificantly lower than initial (day 0) value

Table 9.4 Bioaccessible zinc content (mg/100 g) of the double-fortified millet flours

Flour	Bioaccessible iron (mg/100 g)		
	Days of storage		
	0	30	60
Finger millet	0.22	0.21	0.16
Fortified finger millet	0.83 ^a	0.77 ^a	0.71 ^a
Sorghum	0.39	0.37	0.35
Fortified sorghum	1.63 ^a	1.35 ^{a,b}	1.09 ^{a,b}

Adapted from [27]

Values are average of five replicates

^aSignificantly higher than control (unfortified grain)

^bSignificantly lower than initial (day 0) value

not compromise the bioaccessibility of these two minerals, since the bioaccessible iron and zinc values were similar to those in the flours fortified with either of the minerals alone [24–26, 30]. This indicates that the addition of these two minerals together does not interfere with the bioaccessibility of either of them. The shelf-life of the double-fortified flours was also satisfactory up to a period of 60 days, as indicated by their moisture and free fatty acid contents [31].

Millet flours are normally consumed after heat processing; in India, these millet flours are most commonly consumed in the form of dumpling and *roti* (unleavened bread). Sensory analysis of these two products prepared from fortified millet flours indicated that these products were sensorily acceptable [32, 33]. Bioaccessibility of iron and zinc from the cooked products was comparable to that from the raw flour [20, 21, 25–27]. This indicates that fortification with iron and zinc both individually and in combination does not alter the sensory characteristics of heat processed products prepared from fortified millet flours, and that the bioaccessibility of the minerals is not compromised by subjecting the fortified flours to heat processing.

Conclusion

Millet flours seem to be suitable candidates for fortification with iron and zinc, both individually and in combination. Addition of EDTA has a significant beneficial influence on the bioaccessibility of these minerals. Given the fact that diets in India and probably other developing countries are predominantly plant-based with poor mineral bioavailability, any improvement in the bioaccessibility of essential minerals from the same would be significant in the context of improving their intake. Fortification of millet flours with iron and zinc therefore is a feasible strategy to increase the intake of these important micronutrients. Since millets are consumed as the staple, fortification of the same would not call for any drastic change in food habits, and the fortified millet flours would be easily accepted by the target population. Such qualitatively rich flours could also be used in supplementary feeding programs, and promoted through the public distribution systems for a wide outreach.

The studies mentioned above examined the feasibility of fortifying three millet flours that are commonly consumed in India, with iron and zinc. These millets were found suitable for mineral fortification, providing significant levels of bioaccessible iron and zinc, and were also cost-effective. These studies merit extension to other millets that form the staple in several developing countries world over.

Millet flours thus seem to be promising candidates for fortification with minerals, and if successfully employed for this purpose, would have a wide outreach in combating iron and zinc deficiency.

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