



Development of Functional Calcium Rich Beverage Mix for Lactose Intolerance People

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Authors' contributions

This work was carried out in collaboration among all authors. Authors JG and SK designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Lactose intolerance is the most common digestive disorder caused by the inability to digest the lactose in milk. Lactose intolerance may lead to reduced bone density when accompanied by avoidance of dairy. The only treatment ever known for lactose intolerance is lactose-free food. Thus keeping this in view the objective is to develop the plant based beverage mix for lactose intolerance people using functional ingredients viz., ragi (*Eleusine coracana*) milk powder, green banana and ginger. The techniques used for processing the ragi milk powder are soaking, extraction of milk, dehydration, milling and roasting. Control was developed by using skimmed milk powder, green banana and ginger and in test treatment ragi milk powder was added. The developed product was analyzed for microstructure, physical, functional and sensory evaluated. The developed beverage mix was allowed for proximate nutrient analysis. The result revealed that the ragi milk powder containing treatment was acceptable in terms of all sensory attributes when compared to control. The microstructure analysis result revealed that the structure of test sample was very small and polygonal in shape whereas the control sample had large particle size and round smooth surface. The beverage mix had good protein, high calcium, low fat and no lactose. It

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may be concluded that the developed beverage mix is a lactose free functional food which is suitable for lactose intolerance and also calcium deficient people with improved nutritional content and reduced phytochemical content without any negative impact on sensory attributes.

Keywords: Ragi milk powder; green banana; ginger; calcium; lactose intolerance.

1. INTRODUCTION

Calcium is an essential micronutrient for bone strength and regulation of numerous functions in cells and tissues, such as muscle contraction, blood coagulation and structural support of the skeleton [1]. Maintaining adequate calcium intake during childhood and adolescence is necessary for the attainment of peak bone mass [2]. The recent estimation by International Osteoporosis Foundation (IOF) states that there are 200 million osteoporotic women in the world with an accident of osteoporotic fracture every 3 seconds. World Health Organization estimates that over 270 million people in India are likely to suffer osteoporosis by the year 2020. Andhra Pradesh has highest prevalence among India [3].

As per Indian Council of Medical Research (ICMR) the RDA of calcium is 600 mg for adult and doubles during pregnancy & lactation *i.e.* 1200 mg and 800 mg for children ages 10 to17. But it was found that the average daily intake of calcium (331 mg) by adult population was less than RDA of 600 mg in all the States. The adequate dietary intake of calcium is very important for normal bone mineralization and prevention of bone fracture and diseases [4]. Historically, dairy products were the predominant source of dietary calcium.

The dairy calcium is very important for maintain strong and healthy bone in throughout the lifespan. But some people are unable to digest lactose in the milk due to lack of lactase enzyme which is a major cause of calcium deficiency in those people. Globally lactose intolerance is estimated to affect 33% of the population, but the prevalence rate varies from country to country. It is also estimated that after weaning the intestinal lactase activity had decreased in 75% of human adults. In India, 60-70% of the population was observed as lactose intolerant [5].

The plant based foods are very important source of calcium in lactose intolerance people but the availability of calcium is very low due to presence of anti-nutritional factors [6]. These could be reduced by processing methods such as

soaking, germination, boiling, roasting and pressure cooking [7]. Thus, keeping in view the present study was to develop plant based food for lactose intolerance people. The objective is to develop instant beverage mix using, ragi milk powder (RMP), green banana, and ginger. Skimmed milk powder containing treatment is used as control. The combination of these ingredients will fulfill the RDA of Calcium and the addition of functional ingredients will improve the bioavailability of calcium among lactose intolerant people. Finger millet or ragi (*Eleusine coracana*) is a rich source of calcium which is thirty times more than that of rice and wheat [8]. Green banana contains a high resistant starch acts as prebiotic (17.5%) [9]. Ginger has anti-inflammatory and analgesic effect and it creates acidic condition which helps to increase absorption of minerals [10]. The functional beverage mix incorporated with these ingredients will be assessed for sensory attributes, microstructure physical, functional and proximate composition.

2. MATERIALS AND METHODS

The raw materials such as skimmed milk powder (SKM), ragi grains, green banana and ginger were purchased from local market. Ragi grains were washed and steeped in water for 12 hours at room temperature. Then the ragi milk was extracted and dried in cabinet dryer at 70°C for 6 hrs. Then the dried RMP was subjected to roasting. The schematic representation of steps involved in the preparation of raw ingredients is given in (Fig. 1). The green banana was sliced and ginger was grated and dried at 60°C for 6 hrs in cabinet dryer. Then all dried ingredients were grounded separately and sieved to get uniform size and stored in airtight container for further development of instant beverage mix.

2.1 Development of Beverage Mix

Five different combinations of beverage mix were developed by using functional ingredients. Skimmed milk powder, green banana and ginger containing sample was used as a control as T₀. The development of beverage mix was shown in Fig. 2.

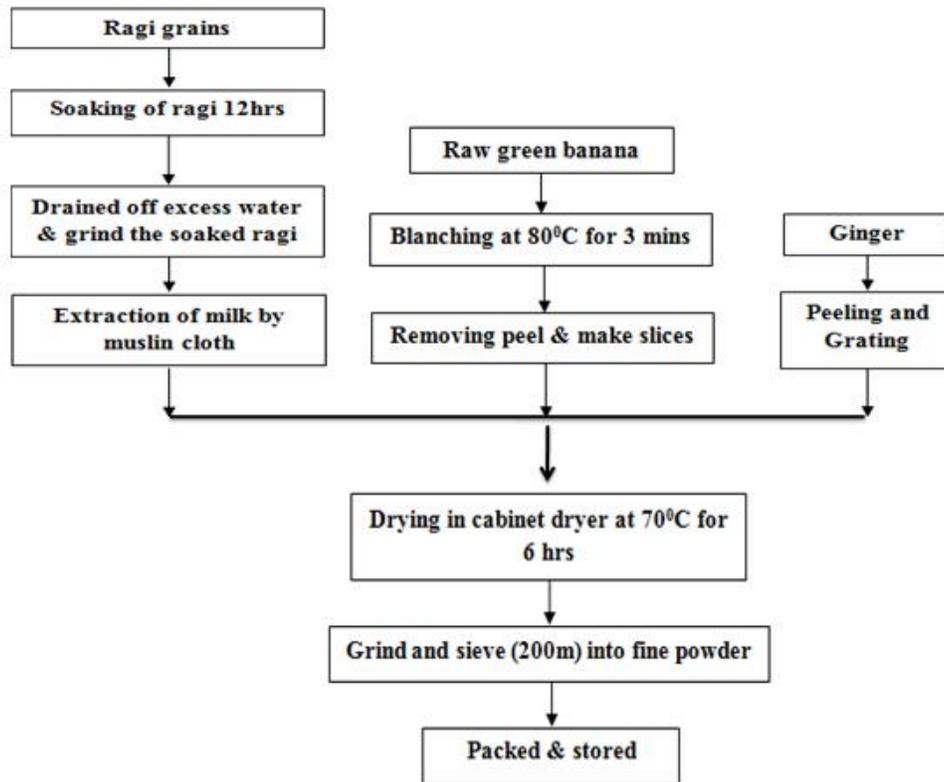


Fig. 1. Flow chart for preparation of raw ingredients for beverage mix

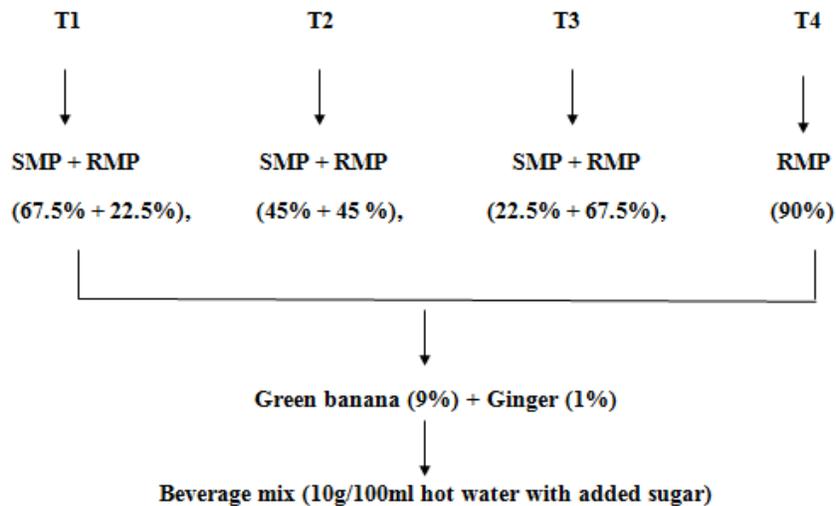


Fig. 2. Development of beverage mix

2.2 Sensory Evaluation of Beverage Mix (Hedonic Scale)

Organoleptic performance was performed as hedonic test to measure the degree of liking for

every formula in terms of taste, flavour, aroma /smell, colour and overall acceptability. A quantitative descriptive analysis (QDA) was carried out for the assessment of the sensory properties, and sensory profile was created for

prepared beverage. The 10 g of beverage mix and 4 g of sugar were mixed into 100 ml boiling water and it was allowed for sensory evaluation. The following attributes were evaluated: overall odour, visual thickness, homogeneity (in the mouth), overall taste intensity, richness of taste, astringent taste, non-typical taste, and mouth coating.

Beverages were kept in closed bottles in a refrigerator ($4 \pm 2^{\circ}\text{C}$) until testing and were removed from refrigerator 30 min prior to a sensory evaluation which allowed the samples to equilibrate to room temperature ($21 \pm 2^{\circ}\text{C}$). Then the samples of the beverages (approximately 20 mL) were presented to the assessors in 30 mL plastic cups, coded with three digit numbers. The 28 semi trained panelists were selected by method of preference / acceptance testing in lab environment. Of them 14 were lactose intolerance people as reported by the individual. The selected panelists evaluated all formulas for degree of liking on 9 point Hedonic scale.

2.3 Microstructure of Beverage Mix

Morphological properties of beverage mix were analyzed by TESCAN scanning electron microscope (SEM) with EDS Analyser. Beverage mix was mounted on aluminum stub using a double backed cellophane tape, coated in auto fine coater, JEOL-JFC-1600, with gold palladium (60:40, g/g). The electron source used for this study is tungsten filament.

2.4 Physical Properties of Beverage Mix

The physical parameters such as bulk density and tapped density [11], flowability and cohesiveness [12], dispersibility [11], Solubility [13] were analyzed for beverage mix as per the standard procedures.

2.5 Functional Properties of Beverage Mix

Functional properties such as water solubility index (WSI) and water absorption index (WAI) were determined by the method of Obilana et al. [14] using standard procedures.

2.6 The Proximate Nutritional Composition of Beverage Mix

The nutritional composition of beverage mix was analyzed for protein by Kjeldhal method [15], total starch by using anthrone method [16], fat by soxhlet extraction method [15], ash [5] moisture

[15] and Calcium by titration method and Phosphorus by spectrophotometric method [17], polyphenol and tannin by spectrophotometric method [16] using standard procedures.

2.7 Statistical Analysis

All the analytical measurement was carried out in 4 replicates and results were expressed in mean values of standard deviation. The collected analytical data was analyzed by using AGRES Software statistical tool.

3. RESULTS AND DISCUSSION

3.1 Sensory Evaluation of Beverage Mix

All five formulas were subjected to organoleptic hedonic evaluation. The sensory scores revealed that in general all formulas scored above 7.0 which was the acceptable level and coinciding with the like very much sensory score. All the parameters such as Colour, Flavour, Texture, Taste and Overall acceptability scored highest in (T_1) (67.5% SMP beverage with 25 percent ragi milk powder) followed by T_0 , T_2 , T_3 and T_4 . It was found that the lactose intolerance people preferred the T_4 treatment as the developed beverage mix was lactose free. Lactose intolerance people may agree to compromise sensory attributes for other perceived benefits. To increase the sensory score of beverage mix sugar concentration can be increased to 4, 4.5 and 5 g/100 ml of beverage mix.

3.2 Microstructure of Beverage Mix

The microstructure of beverage mix is presented in (Fig. 2). The shape of the particles varies like oval, round, polygonal and fibrous shape which is analysed by scanning electron microscopy. The shape of particles size appeared as different geometrical shapes. The granule size of skimmed milk powder was bigger than ragi milk powder, green banana and ginger. When compare to all the size RMP granules had small particle size and fine structure due to processing steps in RMP preparation. Gull et al. [18] reported that microstructure of finger and pearl millet flour is visible with no pores at surface and polygonal in shape with edges. In Fig. 1, the surfaces of all the granules appeared to be smooth and looser agglomerate structures which may be caused by soaking, milling and roasting process. These processes alter the microstructure of ragi which could increase the bioavailability of nutrients such as minerals as reported by Saleh et al. [19].

3.3 Physical Properties of Beverage Mix

The physical parameters such as bulk density and tapped density, flowability and cohesiveness, dispersibility, solubility has major influence on the formation, stability and functionality of the emulsion properties of the instant mixes and presented in (Table 1).

Density of a powder decides the container volume, requirement of packaging materials, and selection of machinery for handling. If it is low, the cost of packaging, storage, and transportation are very high. If the bulk density is high, the volume of packaging is reduced. The bulk density and tapped density of control and beverage mix is 0.57-0.34 & 0.74-0.54 g/ml, respectively. T₀ (SMP) sample had significantly higher ($p > 0.05$) bulk density (0.577 g/ mL) and tapped density (0.74 g/ml) whereas beverage mix was decreased due to presence of fiber content in RMP. The similar studies shows that the aerated bulk densities of various milk

powders, SMP, whole milk powder(WMP) ranged from 0.30 to 0.62 g/cm³; whereas the tapped densities ranged from 0.44 to 0.88 g/cm³ [20].

The flowability and cohesiveness of the instant powder is based on particle size, fat content, moisture content and storage condition. It is a determining factor for the designing of machinery to ensure proper flow of powder and to avoid the formation of clogs [21]. The Flowability (Hausner ratio) of control and beverage mix is 1.31 and 1.73. The RMP containing treatment had significantly higher ($p > 0.05$) flowability due to smaller particle size. The similar studies showed that the flowability of SMP ranged from 1.17-1.42 [22]. The cohesiveness (Carr s index) of the beverage mix ranged from 18-27%. The RMP containing treatment had significantly higher cohesiveness when compare to control the high cohesion percentage is because of large particle size. Ilari [23] reported that the cohesiveness of whole milk powder and skimmed milk powder ranged between (14.2-29.50%). It concluded

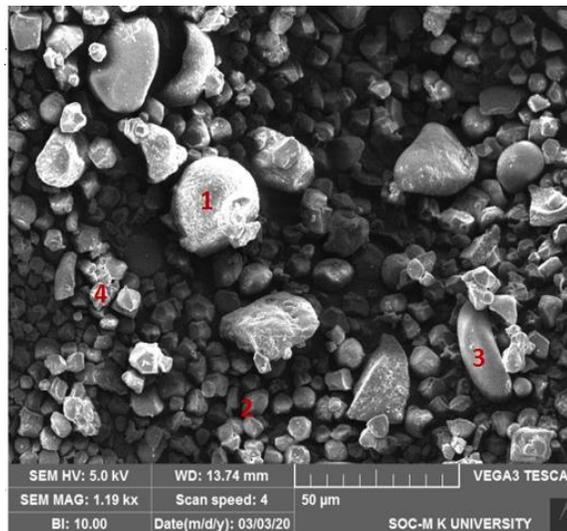


Fig. 3. Microstructure of beverage mix (1. SMP round shape, 2. RMP polygonal shape, 3. Green banana powder oval shape, 4. Ginger powder coarse structure)

Table 1. Physical properties of beverage mix

Treatments	Control (T ₀)	Beverage mix
Bulk density (g/ml)	0.57±0.01 ^a	0.34±0.01 ^b
Tapped density (g/ml)	0.74±0.01 ^a	0.54±0.01 ^b
Flowability	1.31±0.02 ^b	1.73±0.03 ^a
Cohesiveness (%)	18 ±0.03 ^a	27±0.56 ^b
Dispersibility (%)	98.6±0.46 ^a	85±1.33 ^b
Solubility (%)	67.6 ± 1.19 ^a	54.8±0.78 ^b

Values represent mean ± standard error (4 replicates). Means in the same column followed by different superscripts or significantly different at ($p > 0.05$)

that the developed beverage mix had acceptable flowability and cohesion values as it is fine powders.

The dispersibility of instant powder indicates its reconstitutability. The beverage product reconstitutes well in water for leaving little or no sediment, it will show high dispersibility. Dispersibility of control and beverage mix is 98.6 - 85%. The variation is due to addition of RMP. The dispersibility slightly decreased with addition RMP. Because of the fiber content of RMP delayed swelling of the granules which leads to poor reconstitutability and lower dispersibility in beverage mix. The SMP containing treatment had significantly higher ($p > 0.05$) dispersibility that is 98.6% whereas developed beverage mix reconstitutes well in water and leaving little sediment. The similar studies shows that the dispersibility of SMP ($\geq 90\%$) is more than that of WMP ($\geq 85\%$) [24].

The ideal instant powder would wet quickly, sink rather than float and disperse/dissolve without lumps. Hence the solubility is the important feature for any food powder. The solubility of the beverage mix ranged from 67.6% to 54.8%. The solubility of the mix decrease with increasing concentration of ragi milk powder. Tamime [24] reported that the solubility of spray-dried SMP, WMP, and partially skimmed milk powder ($>99\%$) is more than that of tray-dried cheese powder (about 91%). The low values is due to various factors viz., cabinet drying, dry roasting of RMP resulting low aggregation and addition of other ingredients such as green banana and ginger which contain high starch and fiber. But statically all the treatments showed best association.

3.4 Functional Properties of Beverage Mix

The instant properties of a powder involve the ability of a powder to dissolve in water. Most powdered foods are intended for rehydration. Hence the ideal powder would wet quickly and thoroughly, sink rather than float and disperse/dissolve without lumps [25]. Solubility depends mainly on the chemical composition of the powder and its physical state [20]. Powders which absorb much moisture may cake during storage so the high solubility and low absorption index is essential for any instant powder [24]. The functional properties of beverage mix are presented in (Table 2).

Water absorption index (WAI) of different treatment ranged from 16.37-3.15%. Water

absorption index was significantly ($p > 0.05$) higher (16.37%) in beverage mix whereas lowest (3.15%) amount was found in T_0 . Water solubility index (WSI) of different treatment ranged from 107-101%. Water solubility index was significantly higher (107%) in control whereas lowest (101%) amount was found in beverage mix. Pragati et al. [26] reported that the WAI increase with increasing starch content of given sample. The result shown that the beverage mix had high Starch (polysaccharides) content which was able to bind water as gelatinization of starch took place. Kaur et al. [27] reported that the foxtail millet containing flour blend had highest WAI. Due to difference in protein structure and the presence of different hydrophilic carbohydrates might be responsible for variations in the WAI between the flours and flour blends. Charunuch et al. [11] revealed that the WAI & WSI is based on the presence of starch content in given sample.

Table 2. Functional properties of beverage mix

Treatments	T_0 -Control	Beverage mix
WAI (%)	3.15±0.01 ^b	16.37±0.41 ^a
WSI (%)	107±2.65 ^a	101±0.96 ^b

Values represent mean \pm standard error (4 replicates). Means in the same column followed by different superscripts or significantly different at ($p > 0.05$)

3.5 Proximate Composition of Beverage Mix

The proximate nutrient composition of beverage mix was analysed and shown in Table 3. A significant variation was observed between control and beverage mixes in all the nutritive values. The moisture content of beverage mix is significantly higher ($p > 0.05$) value that is 7.45%. Ramashia et al. [28] reported that the moisture content of finger millet flours ranged from 9.17 ± 1.44 to $11.67 \pm 1.44\%$, respectively. The reduction of moisture content is due to processing steps of RMP. The carbohydrate content of beverage mix is 68.62% which is significantly higher ($p > 0.05$) when compare to control. Similar studies shown that Desai et al. [29] revealed that total carbohydrate content of unmalted and malted ragi flour is 76.51 and 76.18%. The protein and fat content of beverage mix are 9.45% and 1.57% respectively. The data showed that there were significant variations were found between the samples in protein and fat content. Similarly, Tripathi et al. [30] developed beverage powder had the protein and

Table 3. Proximate nutrient composition of beverage mix

Parameters (%)	Control T ₀	Beverage mix
Moisture	4.08± 0.028 ^a	7.45 ±0.25 ^b
Carbohydrate	14.18± 0.26 ^b	68.62±1.91 ^a
Lactose	42.29 ± 0.34 ^a	0
Protein	30.74± 0.08 ^a	9.45±0.16 ^b
Fat	0.84±0.01 ^a	1.57±0.01 ^b
Dietary fiber	0.34±0.01 ^b	10.75±0.29 ^a
Ash	7.53±0.19 ^a	2.2±0.05 ^b
Calcium (mg/100g)	1165.6 ± 17.85 ^a	583 ± 9.12 ^b
Phosphorus (mg/100g)	898.7 ±13.96 ^a	506.4 ± 3.44 ^b
Tannin (mg/100g)	7.4±0.14 ^a	59.15±0.16 ^b
Polyphenols (mg/100g)	28.56±0.54 ^a	84.35±0.86 ^b

Values represent mean ± standard error (4 replicates). Means in the same column followed by different superscripts or significantly different at (p > 0.05)

fat content of beverage powder is 12.25% and 1.78%, which is comparable to the values obtained in present study. This may be due to vary the proportion of SMP and RMP. The dietary fiber and ash content of beverage mix is 10.75% and 2.2% as reported by Tripathi et al. [30]. Some difference may be attributed to the fact that the millet milk was extracted and roasted slightly during RMP preparation which was altered the nutritive value.

Significant variations (p> 0.05) were observed in the mineral contents (Ca, P) between the control and beverage mix. The mineral content such as calcium and phosphorous content is 583 and 506.4 mg/100 g. Tripathi et al. [30] reported that calcium content of millet beverage powder is 96.5 mg/100 g. Rathour [31] reported that the calcium and phosphorus content in the ragi grain from different varieties ranged from 398.63 to 501.14 mg/100 g and 303.34 to 322.42 mg/100 g. The variation is due to use of RMP instead of millet flour.

The beverage mix had lower polyphenol and tannin content that is 84.3 mg/100 g and 59.51 mg/ 100 g which is due to processing steps involved in RMP preparation. The tannin and polyphenols content of beverage mix had significantly reduced due to various processing treatments of ragi such as soaking, milk extraction (dehulling) and roasting which causes either decomposition or alteration of the molecular structure of phenolic compounds leading to a reduced chemical reactivity or extractability due to a certain degree of polymerization. Singh et al. [7] reported the total phenolic content of unroasted and roasted samples was observed as 314.24 and 223.31 mg/100 g, respectively. A decrease of total and

free phenolics was also found in previous study [20]. This variation may be attributed to further processing steps during beverage mix preparation. But the bioavailability of mineral content was increased due to reduced tannin content in the beverage mix.

4. CONCLUSION

The instant beverage mix was developed with improved nutrition and presence of functional compounds increase the availability and bioavailability of calcium. The calcium rich formulas available in the market are dairy based and this is not accepted by lactose intolerance persons, this instant beverage mix used as a substitute for non dairy consumers. Thus the developed beverage mix could provide health benefits beyond its basic nutrition.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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