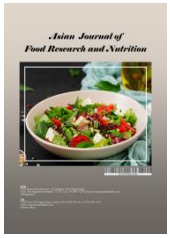

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Physicochemical and Morphological Analysis of Millet Yoghurt Powder: Characterization and Quality Evaluation

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Aim: To fulfil the growing demand for alternatives to animal-derived products, novel plant-based (PB) foods and beverages have been developed and made available to the market in recent years. People who eat too much cholesterol, have lactose intolerance or malabsorption problems, or are allergic to milk proteins demand for alternative options. Therefore this study utilizes millet milk to develop yoghurt powder and the physicochemical and morphological structures of the developed yoghurt powder was evaluated.

Study Design: The present study was conducted in three phases –

1. Millet milk extraction
2. Inoculation of starter culture
3. Dehydration

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Place and Duration of the Study: The present study was conducted in the Department of Food & Nutrition of Babasaheb Bhimrao Ambedkar University Lucknow. The duration of the study is 5 months i.e. from January - May, 2023.
Methodology: The developed millet yoghurt was dehydrated to form yoghurt powder. In the present study, we have analysed various physicochemical properties such as total protein, total fat, pH etc. The methods used were according to AOAC methods. Morphological analysis was conducted using scanning electron microscope (SEM) and the elemental composition was studied by analysing the Energy dispersive X-ray (EDX) graphs.

Keywords: Millets; finger millet; protein; SEM; EDX.

1. INTRODUCTION

Novel plant-based (PB) foods and beverages have been devised and made accessible to the market in recent years to meet the growing demand for alternatives to animal-derived products. Milk and dairy products have long been regarded as a food class containing necessary chemicals for human nutrition that are rarely found in the same proportion in other foods. People who consume too much cholesterol in their diets, have lactose intolerance or malabsorption issues, or have an allergy to milk proteins should, nevertheless, consume alternatives. Additionally, the growing popularity of vegetarianism, increased consumer knowledge of how food choices affect the environment and their health, and the restricted use of dairy products in some regions are all driving up demand for plant based products [1].

Millets are superfoods. With India's growing malnutrition problem, both under-nutrition (vitamin, mineral, and protein deficiencies) and over-nutrition (obesity, metabolic syndrome, and lifestyle diseases), there is a growing awareness of the need to move to healthier, more accessible, and inexpensive diets that include millets [2].

Physicochemical analysis helps ensure the quality and consistency of millet yogurt powder. By assessing various parameters, such as pH, acidity, and moisture content, manufacturers can monitor and maintain the desired quality standards throughout the production process. It also provides information about its nutritional composition [3-5]. Parameters like protein content, fat content, and total solids help determine the nutritional value of the product, which is important for consumers seeking specific dietary requirements. It helps both manufacturers and consumers in making informed decisions about the product [6]. Surface study of a food product is critical in handling, manufacturing, and processing in a food industry. As a result, SEM was used to examine the developed millet yoghurt powder for roughness, morphology, and geometric shape [7].

This paper aims in analysing the physicochemical and morphological characteristics of the developed yoghurt powder in comparison with a market cow milk yoghurt powder.

2. MATERIALS AND METHODS

2.1 Materials

The two millet varieties, finger millet and kodo millet, were purchased from the local market. The beginning culture sachet was also acquired online from a reputable supplier.

2.2 Methodology

2.2.1 Preparation of millet milk

Whole grain millet varieties include finger millet and kodo millet. The contaminants were removed, weighed individually, and the weights were recorded. After that, the grains were steeped for 12 hours. The water was drained, and the grains were pulverised for milk extraction with a small amount of water. The extracted milk was then filtered through a muslin cloth and pasteurised for 30 minutes at 63 degrees Celsius. The milk was then allowed to cool to normal temperature before being used to make yoghurt.

2.2.2 Selection of bacterial strain

The strains used in the study were *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. These bacteria can create lactic acid, which contributes in the production of yoghurt production of bacterial strain.

2.2.3 Preparation of yoghurt

Lactobacillus bulgaricus and *Streptococcus thermophilus* were used in a 1:1 ratio for inoculation. The starter culture was then added to the pasteurised millet milk, which was constantly mixed to avoid the formation of lumps. The mixture was then incubated at 42 degrees Celsius for 6 hours to allow the yoghurt to solidify and form. The resulting yoghurt was allowed to cool before being refrigerated.

2.2.4 Dehydration

The freeze drying technique was utilised in this investigation to create yoghurt powder. It entails drying the product via sublimation at low temperatures and under vacuum. Drying the product extends its shelf life by decreasing its water activity. The yoghurt was allowed to freeze dry for 72 hours at -30 C. It was then finely powdered and sieved.

2.2.5 Packaging and storage

The obtained yoghurt powder were packaged in air tight glass bottle and stored at low temperature.

2.2.6 Physico-chemical analysis

Physico chemical properties of the product were analysed and a comparison was made with the cow milk yoghurt powder. Physical properties such a bulk density, tapped density, hausner's ratio and flowability were determined and chemical properties such as pH, total soluble solids(TSS), moisture content (%), titrable acidity (%), total fat (%) and total protein(%) were also analysed.

2.2.6.1 Determination of Ph

The millet yoghurt powder was subjected to the physical analysis such as, Ph - Digital pH meter was used (ATC model no:6032), 10 mL of sample was dissolved in distilled water in a clean sterile beaker and under sterile environment the pH was measured.

2.2.6.2 Determination of acidity

Titrable acidity is a term used to describe the acidity level of a substance, which can be measured through a titration process.(AOAC 1990) The titrable acidity of the developed millet yoghurt powder is determined by dissolving the sample in water to reconstitute it into a liquid form. The endpoint is usually marked by a colour change in the indicator solution, indicating that all the acid in the yogurt powder has been neutralized [8].

2.2.6.3 Determination of total soluble solids

Total soluble solids content of a solution was determined by the index of refraction.(AOAC 2005) [9] This method tests the solids concentration of a sucrose-containing solution. Handheld refractometer is an analogue instrument used for measuring a liquid's refractive index. The refractometer was calibrated with distilled water. A small drop of sample was placed on the glass and closed. The reading was observed using magnifying eyepiece, reading is expressed as °Brix.

2.2.6.4 Determination of moisture content

Moisture contents of millet yoghurt powder were determined gravimetrically by oven drying at 102 °C for 2 h (AOAC 1990)(1) Moisture content was measured conventionally by loss in mass and expressed as percentage by mass (grams per 100 grams).

The moisture content, W, as percentage by mass of the sample (grams per 100 grams), is equal to:

Where,

M0 is the mass, in grams, of the dish and lid.
M1 is the mass, in grams, of the dish and lid, and the test portion before drying.
M1 is the mass, in grams, of the dish and lid, and the test portion after drying.

2.2.6.5 Determination of protein content

The AOAC (2005) [10] method was used to determine the chemical analysis, which included total protein. The total protein was calculated using the Kjeldhal technique. To determine the amount of nitrogen it was calculated by following formula,

$$\textbf{Nitrogen}\% = \frac{\{\textbf{sample titre} - \textbf{Blank titre}\} * \textbf{Normality of HCl} * 14 * \textbf{Vol}}{\textbf{madeup of the digest} * 100}$$

$$\frac{\textbf{A liquot of the digest taken} * \textbf{Weight of sample taken} * 100}{0}$$

(Protein % = Nitrogen % ×6.25)*

2.2.6.6 Determination of fat content

The "Soxhlet" method used for the study is recognized by the *Association of Official Analytical Chemists (AOAC)* [2] as the standard method for crude fat analysis.

In Soxhlet method crude fat content of the food sample is determined by solvent extracting method, then determining the weight of the fat recovered. The total fat percentage is calculated using the formula,

- Weight of empty flask (g) = W1
- Weight of flask and extracted fat (g) = W2
- Weight of sample = S

$$\% \text{ Crude fat} = (W2 - W1) \times 100/S$$

2.3 Morphological Analysis of the Developed Millet Yoghurt Powder

2.3.1 Scanning electron microscopy

The morphological structures were analysed using scanning electron microscopy (SEM) and the micrographs were obtained. The dried samples from each drying step were placed in an airtight desiccator containing silica gel to entirely remove moisture. The absence of moisture was established by measuring the sample weight on a regular basis until it reached a constant weight. The dried yoghurt was first glued to an iron stub and then made electrically conductive by covering it with a tiny layer of gold in a vacuum chamber for 40 seconds. The photographs were shot at a voltage of 10 kv, at different magnifications of X330, X1000, X2000, X2500 [6].

2.3.2 Energy dispersive X-ray (EDX) analysis

Scanning electron microscopy (SEM) in conjunction with energy dispersive X-ray spectrometry (EDS) is powerful to determine the distribution of elements in a matrix. The EDS analysis generates a qualitative spectrum that identifies the components present in the sample.

2.4 Statistical Analysis

All the findings of this study are presented as the average of replicates. Microsoft excel was used to determine the mean and standard deviation for evaluation.

3. RESULTS AND DISCUSSION

3.1 Physicochemical Analysis

Physical parameters such as bulk density, tapped density, and flowability of the product were analysed and the results are as shown in Table 1.

3.2 Morphological Analysis

The SEM images were obtained for the developed product as shown in the Figs. 1 a, b, c and d shows the morphological structure of the developed millet yoghurt powder.

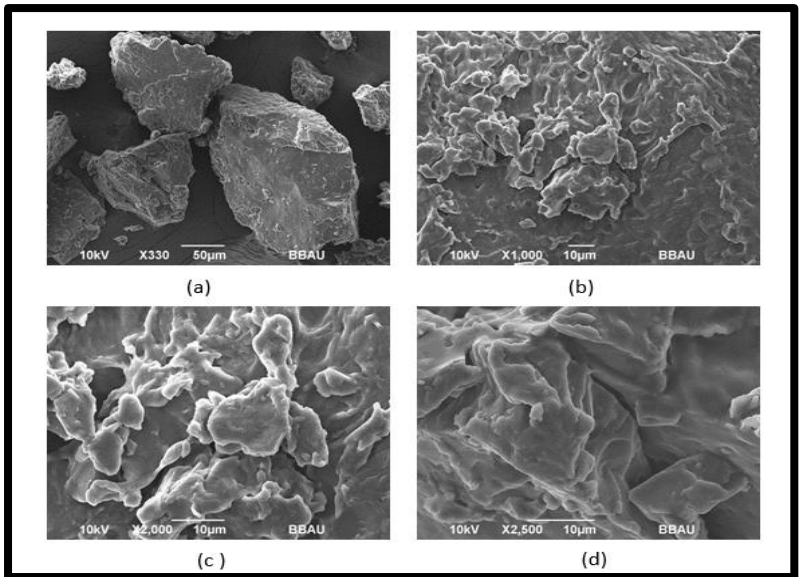

Fig. 1. [SEM images of sample at different magnification (a) x330 (b) x2000 (c)x1000 (d) x2,500]

Table 1. Physical parameters of millet yoghurt powder

Physical parameter	Millet yoghurt powder	Cow milk yoghurt powder
Bulk density (kg/m^3)	760kg/m^3	538 ± 10.05
Tapped density (kg/m^3)	1050	746 ± 6.22
Flowabilty (%)	27.6	27.93

3.3 EDX Analysis

Edx analysis was used to determine the different elements present in the sample. Fig. 2 depicts the EDX graphs and the elements by weight (%).

4. DISCUSSION

The above results indicate that the physical properties of millet yoghurt powder were close to that of the cow milk yoghurt powder. A flowability percentage <30° indicates 'excellent' flow whereas >56° indicates 'very poor' flow. Chemical parameters such as pH, acidity, total soluble solids (TSS), protein, and fat were determined for the developed product and compared with cow milk yoghurt. The results are as show in Table 2. The results indicate that the developed product can be a nutritious option in comparison with cow milk yoghurt powder as it has a high protein content and low in fat. The pH value of the developed yoghurt was also close to the standard.

The findings from SEM images are as follows: Fig. 1 (a), at x330 magnification, depicts the uneven geometry of particles which might be due to the amorphous nature of the sample whereas in Fig d, at x2500 magnification, the structure clearly shows smooth surfaces.The EDX Table depicts that the element present in the highest percentage is carbon (49.76%) and the lowest elemental percentage for calcium (0.20%).

Table 2. Chemical parameters of millet yoghurt powder

Parameter	Millet yoghurt powder	Cow milk yoghurt powder
pH	4.6	4.48
Total soluble solids (TSS; ^{0}brix)	10.8	13.80
Moisture content	1.10	1.35
Titrable acidity (%)	0.12	0.19
Protein (%)	4.8	3.45
Fat (%)	2.9	4.63

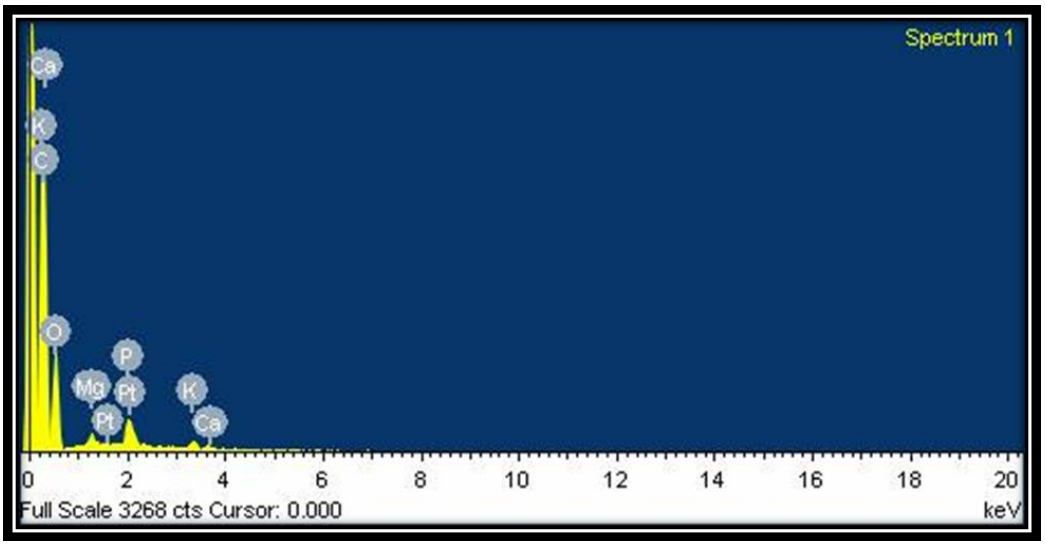

Fig. 2. EDX graph of the product

Table 3. Elemental composition of the millet yoghurt powder

Element	Weight%	Atomic%
C K	49.76	58.50
O K	45.45	40.11
Mg K	0.56	0.32
P K	1.21	0.55
K K	0.82	0.30
Ca K	0.20	0.07
Pt M	2.00	0.14
Totals	100.00	

5. CONCLUSION

The study on millet yogurt powder highlights its potential as a nutritious and sustainable alternative to traditional dairy yogurt. The findings indicate that millet yogurt powder can cater to the growing demand for healthier, plant-based, and allergen-friendly food options. From the results obtained from this study, it was clearly observed that the developed product can be a highly nutritious alternative option to animal milk based yoghurt especially for vegans and anyone seeking a lactose-free or gluten-free product..

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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