

DEVELOPMENT AND EVALUATION OF MILLET-BASED YOGHURT

By

ASWIN P K (2020-06-001)

FATHIMA SHIMNA (2020-06-002)

NEEHARIKA (2020-06-011)

NAYANTHARA B (2020-06-013)



KERALA AGRICULTURAL UNIVERSITY

DEPARTMENT OF PROCESSING AND FOOD ENGINEERING

KELAPPAJI COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY

TAVANUR - 679573 , MALAPPURAM

KERALA, INDIA

2023-2024

DEVELOPMENT AND EVALUATION OF MILLET-BASED YOGHURT

By

ASWIN P K (2020-06-001)

FATHIMA SHIMNA (2020-06-002)

NEEHARIKA (2020-06-011)

NAYANTHARA B (2020-06-013)

PROJECT REPORT

Submitted in partial fulfilment of the requirement of

BACHELOR OF TECHNOLOGY

IN

FOOD TECHNOLOGY

Faculty of Agricultural Engineering and Technology

Kerala Agricultural University



DEPARTMENT OF PROCESSING AND FOOD ENGINEERING

KELAPPAJI COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY

TAVANUR - 679573, MALAPPURAM

KERALA, INDIA

2023-2024

DECLARATION

We hereby declare that this project report entitled “**DEVELOPMENT AND EVALUATION OF MILLET-BASED YOGHURT**” is a bonafide record of project work done by us during the course of the project and that the report has not previously formed the basis for the award to us of any degree, diploma, associateship, fellowship of other similar title of any other university or society.

Place: Tavanur

Date: 24-01-2024

ASWIN P K (2020-06-001)

FATHIMA SHIMNA (2020-06-002)

NEEHARIKA (2020-06-011)

NAYANTHARA B (2020-06-013)

CERTIFICATE

Certified that this project report entitled “**DEVELOPMENT AND EVALUATION OF MILLET-BASED YOGHURT**” is a record of project work done jointly by ASWIN P K (2020-06-001), FATHIMA SHIMNA (2020-06-002), NEEHARIKA (2020-06-011) and NAYANTHARA B (2020-06-013) under my guidance and supervision and that it has not previously performed the basis for award us of any degree, diploma, associateship, fellowship to them.

Place: Tavanur

Date: 24-01-2024

Dr. RAJESH G K

Assistant Professor

Dept. of PFE

K.C.A.E.T, Tavanur

ACKNOWLEDGEMENT

Words are not substitute for thoughts and feelings. It is more so when we the project team wish to express our sincere thanks to our project guide, **Dr. Rajesh G K**, Assistant Professor and **Er. Anjali M G** our co-guide, for their valuable guidance, encouragement and advice throughout the project work.

We express our scrupulous gratitude to **Dr. Prince M V**, Head of the Department, Department of Processing and Food Engineering, KCAET, Tavanur. We also extend our sincere thanks and gratitude to **Smt. Sreeja R**, Assistant Professor, Department of Processing and Food Engineering for her support and encouragement.

We would like to thank **Dr. Senthilkumar R**, Assistant Professor and **Dr. Sruthy P S**, Assistant Professor for their valuable guidance and support.

Also we thankfully remember the services **Smt. Sandhya** (Skilled Assistant), **Smt. Geetha T R** (Lab Assistant), KCAET Tavanur, for their immense help and their continuous support throughout the project.

We would also like to thank our **parents** for their love and support throughout the project. We also extend our thanks to all the **faculties of library**, KCAET, for their help and support. Last but not the least we thank our sincere friends for their ever-willing help, support and cooperation that helped us to complete the project successfully. Above all, we bow our head before **The Almighty**, whose grace and blessings have empowered us to complete this toil.

LIST OF CONTENTS

Chapter No.	Title	Page No.
	LIST OF TABLES	
	LIST OF FIGURES	
	LIST OF PLATES	
	SYMBOLS AND ABBREVIATIONS	
I	INDRODUCTION	1
II	REVIEW OF LITERATURE	7
III	MATERIALS AND METHODS	27
IV	RESULTS AND DISCUSSION	48
V	SUMMARY AND CONCLUSION	60
	REFERENCE	62
	APPENDICES	
	ABSTRACT	

LIST OF TABLES

Table No	Title	Page No
2.1	Nutritional composition of different varieties of yoghurt	16
3.1	Yoghurt sample proportions	33
3.2	Nine-point hedonic scale	36
4.1	Physico -chemical properties of cow milk and millets	48
4.2	Sensory evaluation	51
4.3	Physico-chemical composition of optimized yoghurt samples	54
4.4	Proximate analysis of optimized yoghurt sample	57
4.5	Total yeast and mould count	58

LIST OF FIGURES

Figure No	Title	Page No
3.1	Flow chart –Preparation of finger millet-based yoghurt	29
3.2	Flow chart –Preparation of proso millet-based yoghurt	31
3.3	Flow chart –Preparation of foxtail millet-based yoghurt	32
3.4	Sensory evaluation of developed yoghurt samples	37
4.1	Radar chart of sensory evaluation	52
4.2	Day 14 th YGC agar plate	58

LIST OF PLATES

Figure No	Title	Page No
3.1	pH meter	38
3.2	Refractometer	39
3.3	Spectrophotometer	41
3.4	Soxhlet Apparatus	42
3.5	Crude fibre estimator	43
3.6	Infrared moisture meter	44

SYMBOLS AND ABBREVIATIONS

<i>et al.</i>	:	and others
%	:	per cent
&	:	and
/	:	per
<	:	less than
>	:	greater than
±	:	plus or minus sign
°	:	degree
°B	:	degree brix
°C	:	degree celsius
Cfu ml ⁻¹		colony forming units per millilitre
etc.	:	etcetera
Fig.	:	figure
g	:	gram
g ml ⁻¹	:	gram per millilitres
NaOH	:	sodium hydroxide
NaCl	:	sodium chloride
KCAET and Technology	:	Kelappaji College of Agricultural Engineering and Technology
Kg	:	kilogram
mg	:	milli gram
min	:	minute
ml	:	millilitre
No.	:	number
pH	:	percentage of H ⁺ ions

INTRODUCTION

CHAPTER I

INTRODUCTION

Growing interest in healthy eating has given rise to a new range of food products on the market that also provides nourishment, improve health by increasing well-being and reducing the risk of certain diseases. The present importance of functional foods on the market is variable and difficult to determine, but it is clear that they have a high growth potential. Among the different product sectors, the dairy sector is the one which has undergone greatest change, with the introduction of new products claiming healthy characteristics. In recent years, traditional products like skimmed dairy products or those with probiotic characteristics like yoghurt have expanded to incorporate an ample range of fermented milk of prebiotic or probiotic nature, and yogurt and milk with different active ingredients that offer the consumer an alternative to conventional dairy products.

Dairy products are considered as a key component to an overall healthy, balanced and nutritious diet. This indicates that consumers are inclined to continue consuming dairy, at a time when they are prioritizing nutrition and health. As a result of this, consumers are making greater efforts to seek out multi-functional benefits.

Though milk is a nutrient rich and well-balanced food, it is very susceptible to microbial growth results in spoilage. To prevent this, various shelf stable dairy products have been developed *viz.* cheese, milk powder, concentrated milk, fermented milk, butter, ice cream etc.

Fermentation is an ancient and inexpensive food preservation method. It improves the nutritional value and digestibility of raw products enhances sensory characteristics, and improves the functional qualities available to local communities (Blandino *et al.*, 2012). Traditionally, fermented foods and beverages (sour porridges, beverages, fermented vegetables, fruits, milk, meat, alcoholic and non-alcoholic

beverages) represent a major dietary component in countries (Nout *et al.*, 1997). The microorganisms may be indigenous to the food, or may be added as a starter culture after pre-treating or cooking the product (Mokoena *et al.*, 2016). The use of lactic acid bacteria (LAB) increases the acidity and decreases the pH of the substrate, thereby inhibiting many pathogens.

Fermented milks have been developed throughout the world as a means of preserving milk against spoilage. Fermented milks are popular in view of organoleptic and other properties such as the characteristic flavour, refreshing taste and improved digestibility. The composition of fermented milks can be easily tailored to meet various dietary requirements especially in the production of low-calorie fermented milks.

Yoghurt is a fermented milk product from an anaerobic fermentation of milk and milk products by the lactic acid fermentation through the action of majorly *Streptococcus thermophilus* and *Lactobacillus bulgaricus*. Other microorganisms like *Lactobacillus acidophilus*, *Lactobacillus subsp. casei*, and *bifidobacteria* are also used in yoghurt fermentation (Priyanka *et al.*, 2012). Yoghurt is formed when milk is coagulated, or form curds by the work of lactic acid or its degree present or introduced into milk enough to coagulate it (Priyanka *et al.*, 2012). Yoghurt and similar fermented milk products in particular are thought to originate from the Middle East. The first production of fermented milk products derived from the requirement to prolong the shelf life of milk instead of being disposed. Yoghurt manufacture was in the beginning based on knowledge and empirical processes without standard procedures or investigation of the steps that occur during the entire process. Only after the late 20th century, when yoghurt became a profitable commercial good, its manufacture became industrialized and the processes were standardized. During the last 20 years, interest in yoghurt manufacture has increased tremendously for scientific and commercial reasons. Scientific findings have suggested that new fermented dairy products produced by inoculating probiotic cultures and fortification with bioactive

compounds promote human health with improved textural and rheological characteristics. Thus, there has been an increased consumer demand for yoghurt and similar fermented dairy products (Tamime and Robinson, 2007). Yoghurt is one of the most popular fermented milk products worldwide and has gained widespread consumer acceptance as a healthy food. Yoghurt is a product of the lactic acid fermentation of milk by addition of a starter culture containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* sp. *Bulgaricus* (Tamime and Robinson, 2007).

Yoghurt is said to be a dense food and is rich in protein, carbohydrates, amino acids, minerals (Calcium and Phosphorus) and vitamins (thiamin-B1, riboflavin-B2, niacin-B3, folate-B9, cobalamin-B12, and vitamin C), but is lacking in iron. The fat content of yoghurt depends on the fat content of the mixture and the type of milk used (Weerathilake *et al.*, 2014). According to the NDBsr26, a 100g 2 serving of plain low-fat yoghurt contains 183 milligrams of calcium, 17 milligrams of magnesium, 234 milligrams of potassium, 144 milligrams of phosphorus, and 0.9 milligrams of zinc (El-Abbadi *et al.*, 2014).

Zhang and Mahoney (1989) mention that, animal milk products are commonly consumed all over the world, with high or sufficient proportions of proteins, vitamins, and minerals except iron. The lack of iron in dairy products decreases the iron density of diets because dairy products are consumed daily. The fortification of dairy product with staple available products is essential for iron improvement daily by diet diversification.

Millet is important cereal and nutritious food rich in minerals and phytochemicals. Millets are grown in marginal areas but they are relatively rich in proteins and minerals and are superior in amino acid balance in comparison to conventional cereal crops besides vitamins and fibre (Singh, 2012). Millets are also rich sources of phytochemicals and micronutrients and they have good balance of amino acids (Mal *et al.*, 2010). The main drawback in millet nutrition is its

bioavailability due to very high antinutritional factors which can be reduced by various processing techniques. Soaking is one of the methods used to improve the nutritional value of millet that leads to the breakdown several components into simpler compounds which alter the texture, flavor, aroma and taste (Parveen *et al.*, 2003). Soaking followed by germination can improve the nutrition of millets as it helps in reducing starch component, induces hydrolytic enzymes for phytate reduction and some flavanoid components. Sprouted seeds contain high protein, low unsaturated fatty acids, low carbohydrate and vitamins compared to ungerminated seeds. Mineral content such as phosphorus, calcium, zinc and copper were higher in sprouts as the hydrolysis of phytic acid gets activated during germination. Hence, soaking and germination of millet is important processing method in developing food product with low viscosity and high energy (Inyang *et al.*, 2008). And there is meagre information on soaking and germination time of millets to extract the millet milk and developing millet milk-based value additional products with bio available nutrients. This study mainly focuses on standardizing soaking and germination time to obtain improved millet milk yield and to evaluate curd based on millet milk. Processing and utilization of millets are largely confined to home scale that renders many of the valuable nutrients unavailable to human beings on wider scale. Millets are nutritionally, especially in micronutrient content superior to the commonly consumed cereals. Luteolin, a flavone present in sorghum and millets is reported to have antioxidant, anti-inflammatory, cancer preventive and anti-arrhythmic properties (NAAS, 2012). Value-addition and improving health benefits of millets by combining with milk and by applying advanced technologies for their processing and preservation opens new avenues for the product diversification.

Hence an effort has been made to develop millet-based yoghurt with the following objectives:

1. To formulate and optimize millet-based yoghurt with incorporation of fingermillet, proso millet and foxtail millet.
2. To evaluate the consumer acceptability of the developed yoghurt.
3. To conduct the shelf-life studies of the optimized yoghurt.

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

This chapter deals with the literature reviews on formulation of millet-based yoghurt, its quality evaluation, sensory analysis and storage studies.

2.1 YOGHURT

The term "yoghurt" comes from the Turkish word "jugurt," which was used to designate acidic fermented dairy dishes and drinks (Priyanka *et al.*, 2012). *Streptococcus thermophilus* and *Lactobacillus delbrueckii spp. bulgaricus* ferment milk to make yogurt. It contains some digested lactose, and is richer in some essential nutrients like protein, calcium, phosphorus, riboflavin, thiamine, vitamin B12, folate, niacin, magnesium and zinc compared to milk (Hadi *et al.*, 2015, Ademosun *et al.*, 2019). Yoghurt is formed when milk is coagulated, or form curds by the work of lactic acid or its degree present or introduced into milk enough to coagulate it (Priyanka *et al.*, 2012).

Reeta *et al.* (2015) mentioned that the nutritional composition of yoghurt varies depending on the strains of starter culture used in the fermentation, the type of milk used (whole, semi skimmed, or skimmed milk), the species from which the milk is obtained (bovine, goat, or sheep), the type of milk solids, solid non-fat, sweeteners, and fruits added before fermentation, and the length of the fermentation process.

2.1.1 History and Origin of Yoghurt

Fermentation is a food processing technique that has been used for thousands of years to preserve food. Acidifying bacteria are beneficial microorganism that helps in milk

preservation and in the improvement of the shelf life of milk by preventing the growth of undesirable microorganisms (Françoise, 2017).

According to Weerathilake *et al.* (2014), yoghurt over the centuries has been recognized as the most popular fermented food product and it has a wide range acceptance worldwide. It is known to have tremendous nutritional and health benefits. Yoghurt origin is dated back to the 6000 B.C. in central Asia when the Neolithic people began food producers by milking their cows and storing them in sheep-skin. This accidentally led to the discovery of fermented milk products which includes yoghurt. Over centuries, yoghurt has evolved into a commercial making/production which has further improved into the production and availability of varieties with a range of flavours, forms, and textures (Weerathilake *et al.*, 2014).

Yoghurt was found by nomadic peoples in the Middle East approximately 5,000 B.C (Francoise, 2017). It has been eaten by various cultures for thousands of years. Yoghurt gets its name from the Turkish term yogurmak, which implies thickening, coagulating, or curdling (Moreno *et al.*, 2013). Yogurt is also known as kатык (Armenia), dahi (India), zabadi (Egypt), mast (Iran), lebenraib (Saudi Arabia), laban (Iraq and Lebanon), roba (Sudan), iogurte (Brazil), cuajada (Spain), coalhada (Portugal), dovga (Azerbaijan), and matsoni in many cultures and nations (Georgia, Russia, and Japan) (Ramandeep *et al.*, 2017, Fisberg and Machado ,2015).

In recent times, yogurt is typically milk that has been fermented and acidified with viable and well-defined bacteria, creating a thickened, often flavoured, product with an extended shelf life. It contains essential nutrients and is a medium for fortification (were other health improving and nutrient modifying probiotics, fibers, vitamins, and minerals are added). Yoghurts also represent functional food and can be modified with sweeteners, fruits, and flavours to affect the nutritional and health benefit, consistency, and aroma. Yoghurt is recently produced from other animal milk

like goat, sheep, buffalo, camel, etc. and plant sources like rice, soy, and nuts (Fisberg and Machado, 2015).

2.1.2 Types of Yoghurt

Weerathilake *et al.*, (2014)) and Ramandeep *et al.*, (2017)) mentioned that different types of yoghurt are available in different varieties and forms based on the various factors associated with their production. The numerous categories of yoghurt are:

2.1.2.1 Based on the chemical composition of the milk:

Milk is the major ingredient in yoghurt production, the different variety of nutrient composition is based on the nutrient of the milk used. Due to diet diversification, milk type production and dairy diet preference, yoghurt production can come in forms of regular yogurt or full-fat, low-fat yogurt and non-fat yogurt. Low-fat yogurt and non-fat yogurt are produced from low-fat milk or partially-skim milk, and skim milk respectively.

2.1.2.2 Based on the physical nature of the product:

- a. Set yoghurt: Set yoghurt is also known as solid yoghurt. It is majorly characterized as incubated and cooled in final packaging during production.
- b. Stirred yoghurt: Stirred yoghurt are known as semi-solid yoghurt. During production the mixture is incubated, after fermentation breaking is done by stirring before cooling and packaging.

c. Drinking yoghurt: Drinking yogurt are in fluid state. In production it usually undergoes the process of homogenization to reduce the particle size which assured the hydro colloidal distribution and stabilization of protein suspension.

2.1.2.3 Based on the flavour of the product:

a. Plain/Natural yoghurt: It is made to be unsweetened and is a naturally fermented milk product containing no added colour or any other additives. It is closer to the nutritional value of milk, provides the nutritional benefits associated with fermentation and is low in calories. Plain/natural yoghurt has the richest calcium content amongst other yogurt products.

b. Flavoured yoghurt: Yoghurt comes in different flavours due to different consumer preference, needs and demands. Flavours are added during production stage based on the need for a wide array of tastes and to increase the sweetness of the product.

2.1.2.4 Based on the manufacturing processes:

a. Pasteurized and UHT yoghurt: Pasteurized yogurt are prepared after fermentation by heat treatment with different time-temperature combinations in order to prolong the shelf life and to reduce the natural tartness of the yogurt.

b. Frozen yoghurt: The Pennsylvania Code defines frozen yogurt as a food which is prepared by freezing while stirring a pasteurized mix. It is inoculated and incubated to get the fermented yoghurt product before it is frozen. It is produced to have the same consistency as ice-cream.

c. Dried yoghurt/yoghurt powder: It is produced by fermenting a non-fat milk with standard starter culture/microorganism to the right pH and consistency, the freeze

drying it to get yoghurt powder. Yoghurt powders are used in the production of confectioneries and in baked products.

d. Concentrated yoghurt: After fermentation, the coagulum is broken down then the yogurt is concentrated by boiling off some water under vacuum conditions. Heating of low pH Yogurt leads to denaturation of protein which produces a rough and gritty texture.

2.1.2.5 Based on their production method or Origin:

a. Balkan-style Yogurt: Balkan-style yogurt is also known as set-style yogurt which is produced to have a characteristic thick texture and made in small and individual batches. It is incubated for over 12 hours or more until the desired thickness, flavour and creaminess is attained. It can be used as a substitute for sour cream, salad dressings or topping. It can also be consumed regularly, either unsweetened, sweetened or with addition of fruits, cereals or anything of choice.

b. Greek-style Yogurt: It is also known as Mediterranean-style yogurt, it is manufactured with partially condensed milk or by straining whey from plain yogurt to make it thicker and creamier. It is available in full-fat and low-fat yogurt.

c. European-style Yogurt/Stirred Curd Yogurt: The European-style yogurt is a type of stirred yogurt with a characteristic creamy and smooth texture. It is manufactured by fermenting the yogurt mixture in a large vat instead of individual cups, then cooling and stirring in order to obtain the creamy texture. It is mostly produced with added fruits (like blueberries, strawberries, mango, and peach) and flavours.

d. French-style Yogurt: This style of yogurt is also known as custard-style yogurt. It is made by direct culturing in a pot according to a French culture, its final product is in a pudding-like texture. Sometimes French-style yogurts are flavoured with fruit pieces

which is stirred into the mixture. It is known to be a good source of iron, protein and vitamin A.

e. Fruit Yogurt: Fruit yoghurt can either be produced by setting the fruits at the bottom of the packaging (sundae-style yogurt) or the uniform distribution of fruit within the yogurt itself (Swiss-style yogurt). Fruit pieces or pulp are added at production stage, it produces variety of tastes and increases the consumer appeal and sweetness of yoghurt.

f. Herbal Yogurt: this is produced by the addition of herbal substances, spices and seeds like cinnamon, fenugreek, Moringa, Ugu etc. during the production process before fermentation or after fermentation of the yoghurt.

2.1.3 Production of Yoghurt

Yogurt is made with a variety of ingredients including milk, sweeteners, stabilizers, fruits, flavours, and bacterial cultures (Weerathilake *et al.*, 2014, Corrieu and Be'al,2016). Production processes of yoghurt are as follows:

2.1.3.1 Milk Standardization:

In the production of yoghurt, milk standardization is important for the mixing of solid fat (SF) and solid not fat (SNF) (Ramandeep *et al.*, 2017). According the Codex Alimentarius Commission, yogurt should have a minimum protein content of 2.7% and a maximum fat content of 15% in order to achieve this, the FAO/WHO standards stipulate that milk should be standardized with the minimum SNF and milk fat content of 8.2% and 3%. The average composition of bovine milk comprised of 4.5% lactose, 3.3% protein, 3.5% of fat and 0.7% mineral matter, to attain the desired SNF content the milk mixture is fortified with milk powder. (Weerathilake *et al.*, 2014) Stabilizers such as pectin and gelatin are added to the yogurt mix in order to attain the

characteristic properties of yogurt namely, texture, mouth feel, appearance, viscosity and 10 to inhibit the whey separation, the use of thickeners and stabilizers (gelatin, pectin, xanthan gum, carrageenan, starch, etc.) at concentrations varying from 5% to 10% is allowed by FAO/WHO to improve the yogurt texture (Corrieu and Be'al2016).

2.1.3.2 Homogenization:

Homogenization is a size reduction process; it involves the breaking down of fat globules into smaller size in order to get uniformity and a size of 1µm throughout the yoghurt product (Ramandeep *et al.*, 2017) Homogenization is a very important stage and process in yoghurt production because it prevents the separation of fat from whey and also make the creaminess of milk uniform in order to attain a good end product. Homogenizers and Viscolizer are used in this process. Milk homogenization is accomplished by forcing the liquid milk through a small opening at a high speed to break down the fat globules with shearing force (Weerathilake *et al.*, 2014) Homogenization pressures often range between 10 and 20 MPa for 10 -17 minutes. Since the efficiency of homogenization is much better when the fat phase is in a liquid state, the process is usually carried out at high temperatures (55°C to 80°C) (Lange *et al.*, 2020). Serra *et al.*, (2009) identified that recently, ultra-high-pressure homogenization are introduced into commercial yogurt production. This has brought the increase in yogurt firmness and water holding capacity in comparison to the conventional homogenization process (Ramandeep *et al.*, 2017).

2.1.3.3 Pasteurization (Heat treatment):

The aim of pasteurization in yoghurt production is to rid the milk of all pathogen, to substantially reduce the total bacterial count for improved quality and to destroy lipase

and other milk enzymes. Pasteurization of milk is done using plate heat exchanger at industrial yogurt manufacturing. The mix is heated at 90 or 95° C for 3–7 min (5min) before cooling down to fermentation temperature. The heat exchanger can either be for batch process (Holding method or low temperature long time (LTLT) method) or a continuous process (High temperature short time (HTST) method) (Corrieu and Be'al, 2016).

2.1.3.4 Inoculation and Fermentation:

After pasteurization, the temperature of the mix is allowed to reduce to 43-46 °C. 2% (v/v) of starter culture is added in ratio with the yogurt mix (Weerathilake *et al.*, 2014). A typical standard starter culture consists of *Staphylococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* in 1:1 ratio. In bacterial fermentation, lactose is converted to lactic acid which reduces the pH of the milk from 11 6.7 to ≤ 4.6 which causes the formation of a gel/coagulation of the protein casein. This process is known as milk acidification (Lange *et al.*, 2020). During milk acidification, volatile compounds are produced which gives the yoghurt a characteristic flavour and aroma. (Ramandeep *et al.*, 2017)

2.1.3.5 Cooling and Packaging:

Packaging and cooling of yoghurt is based on their physical nature, whether it is a stirred type or a set type of yoghurt. A stirred type yoghurt is produced fermenting the yoghurt mix in a tank followed by breaking and stirring prior to packaging and cooling. While a set type yoghurt is produced by filling the mix into the packaging material and fermenting/incubating in the packaging material, then cooling. According to the USDA Specifications, after the final steps in manufacturing and/or packaging, the yogurt

should be cooled and maintained at temperatures less than 7.2 °C (Weerathilake *et al.*, 2014)

2.1.4 Nutritional and Health Benefits of Yoghurt

Aryan and Olson, (2017) identified in history during the 1500s, that King Francis I of France was cured of a severe diarrhea by consuming prescribed yoghurt. Yoghurt is known to contain the same nutritional profile as milk. Consumption of yoghurt may lead to enhancement in bone and muscle health of both developing children and young adults and also the elderly. Calcium, protein and vitamin D are essential for good muscle and bone growth, development and maintenance. Due to the rich micro flora (probiotics) in yoghurt which are viable cells, consumption of yoghurt may lead to the enhancement of immune response and reduction in risk of infectious diseases associated with gut, stomach and the small intestine. Studies shows that deficiency of vitamins and minerals leads to immune impairment, micronutrients like zinc and vitamin B6, beneficial bacteria and protein are essential for the improvement of human and animal immunity (El-Abadi *et al.*, 2014).

Weerathilake *et al.*, (2014) stated that the consumption of probiotics are beneficial in maintaining excellent health and restoring bodily vitality. They also discovered that probiotics in yoghurt are therapeutic and important in the treatment of intestinal disorders, as well as the prevention of urogenital infection, constipation relief, diarrhea prevention, infantile diarrhea prevention, hypercholesterolemia prevention, colon/bladder cancer prevention, and osteoporosis prevention. Yogurt consumption is also reported to be effective in cytokine production, T-cell function and natural killer-cell activity, and thereby result an overall immunological enhancement, also provides preventive effects on the relapse of ulcerative colitis, maintenance of normal intestinal flora, enhancement of the immune system, reduction of the lactose- intolerance, serum cholesterol levels, and the enhance anti carcinogenic activity.

The nutritional composition of yoghurt is said to be similar to that of milk. Yoghurt contains protein, amino acids, carbohydrates, calcium, phosphorus, vitamins and minerals (Table 2.1). The nutritional composition of yoghurt varies according to the variety or type of Yoghurt. He also identified that yoghurt is a rich source of riboflavin (Vitamin B2), thiamin (Vitamin B1), vitamin B12, folate, niacin, magnesium and zinc. The carbohydrate present in yoghurt is lactose. In total, raw milk contains about 4.6% lactose content, the amount of lactose is normally lowered by 20- 30% during the fermentation process by the conversion of lactose into a simpler form of glucose and galactose due to the metabolic activity of lactic acid bacteria (Ramandeep *et al.*, 2017

Table 2.1 Nutritional composition of different varieties of yoghurt (per 100 g)

Component	Whole milk yoghurt	Low fat yoghurt	Non-fat yoghurt	Greek style yoghurt	Drinking yoghurt
Energy (kcal)	79	56	54	133	62
Carbohydrate (g)	7.8	7.4	8.2	4.8	13.1
Fat (g)	3.0	1.0	0.2	10.2	Trace
Thiamin(mg)	0.06	0.12	0.04	0.12	0.03
Riboflavin(mg)	0.27	0.22	0.29	0.13	0.16
Niacin (mg)	0.2	0.1	0.1	0.1	0.1
VitaminB6(mg)	0.10	0.01	0.07	0.01	0.05
Vitamin B12(mg)	0.2	0.3	0.2	0.2	0.2
Folate (µg)	18	18	8	6	12
Carotene (µg)	21	Trace	Trace	Trace	Trace
Vitamin D	0	0.1	Trace	0.1	Trace
Potassium(mg)	280	228	247	184	130

Calcium (mg)	200	162	160	126	100
Phosphorus (mg)	170	143	151	138	81

Source: *Weerathilake et al.*, (2014)

2.1.5 Raw Materials for Yoghurt Manufacture

Tamime and Robinson (1999) reported that yoghurt is produced using a variety of ingredients like milk, sweeteners, stabilizers, fruits, flavors, and bacterial cultures. Milk is the main ingredient employed in yoghurt production. The type of milk to be used depends on the variety or type of the yoghurt that will be prepared. Whole milk is used for full fat/regular yoghurt, partially skimmed milk is used for low fat yoghurt and skimmed milk is used for non-fat yoghurt. Cream or butter fat is used to regulate the fat content however skim milk powder, whey protein concentrate are used to raise the total solid content of the yoghurt. Stabilizers are normally added to the mix to build up the body and texture leading to an increase in firmness, avoids whey separation, and helps in uniform distribution of constituents. Moreover, sweeteners are used to enhance the flavour and consumer appeal.

2.1.6 Starter Cultures for Yoghurt Production

2.1.6.1 Lactobacillus bulgaricus

Lactobacillus bulgaricus grows at reasonably low pH values (below pH 5.0) and has most favorable growing temperature of around 37°C. There are various nutritional benefits from these lactic acid bacteria, such as better absorption of lactose and control of intestinal infections, control of cancer cells, lowering of serum cholesterol levels etc (Gilliland, 1990).

Sanders and Klaenhammer (2001) reported that *Lactobacillus bulgaricus* is a probiotic strain of human origin available commonly in usual foods such as milk, yoghurts, juice and dietary supplements. It is assumed that the strain is functionally essential to human because of their health favorable effects which have been built up to ease, prevent several diseases like colon cancer, diabetes, hay-fever, and lactose intolerance. *Lactobacillus bulgaricus* is a homo fermentative species, fermenting sugars into lactic acid and known as Gram positive non-spore-forming rods with rounded ends that occur one by one and in short chain (Gopal, 2011). Traditional yoghurt is produced from symbiotic growth of starter bacteria *Streptococcus thermophilus* and *Lactobacillus bulgaricus*. To get better health benefits, the current trend is to add *Lactobacillus acidophilus* to yoghurt (Ashraf and Shah, 2011).

2.1.6.2 *Streptococcus thermophilus*

Rasic and Kurmann (1978) stated that the growth temperature for *S. thermophilus* varied from 20°C to 50°C with an optimum of 40°C to 45°C. According to Carper (1998), yoghurt containing *S. thermophilus* and *L. bulgaricus* reduced the rate of lung cancer in mice. It is a thermophilic lactic acid bacterium (LAB) commonly used in the production of yoghurt products and can be considered as the important industrial dairy starter after *L. lactis* (Hols *et.al.*, 2005). Combinations of probiotic commodities containing *S. thermophilus* helped to increase gastrointestinal function, for example prevention of rota viral diarrhea in infant and reduce the severity of Necrotizing Enterocolitis in neonates (Saavedra *et. al.*, 1994; Bin-Nun *et.al.*, 2005).

2.2 MILLETS

Millet is an annual small-seeded cereal crop grown all over the world for food, feed, forage and fuel. There are about 20 different species of millets. Commonly cultivated species include proso millet (*Panicum miliaceum* L.), pearl millet (*Pennisetum glaucum* (L.) Gaertn.), finger millet (*Eleusine coracana* Gaertn.), kodo millet (*Paspalum scrobiculatum* L.), foxtail millet (*Setaria italica* (L.) P. Beauvois), little millet (*Panicum sumatrense* Roth ex Roem. & Schult.) and barnyard millet (*Echinochloa esculenta* (A. Braun) H. Scholz). Though they belong to the Poaceae family, a significant morphogenetic diversity exists at species, genus and subfamily levels. They differ at their genome size, ploidy levels and breeding systems. Millet is the world's sixth most important cereal grain supporting as a major source of energy and protein for millions of people in India, Africa, and China, and especially for the people living in arid and semiarid regions. Millets are cultivated globally with major contributions from India, Nigeria, Niger, China, Mali and Burkina Faso. Asia (48%) and Africa (48%) dominate the production of millet compared to Europe (3%) and America (1%). Short duration and wide adaptability under different environmental conditions make millet one of the most suitable crop for sustainable agriculture and future food security. Millets can give significantly higher yields on marginal lands with low fertility and low input agricultural systems compared to many other crops. Millet can serve as a saviour for the world's rapidly increasing population with the potential to avert food shortage and famine.

2.2.1 Nutritional Values of Millets

Millet grains are nutritionally comparable and even superior to major cereals with respect to protein, energy, vitamins, and minerals (Sehgal and Kawatra, 2003). Millets are the rich source of minerals, nutraceuticals, and higher dietary fibers than rice or wheat and contains 9-14% protein, 70-80% carbohydrates (Hadimani and Malleshi, 1993). These are rich sources of phytochemicals and micronutrients (Singh

et al., 2012b). The nutritional status of a community has therefore been recognized as an important indicator of national development (Singh and Raghuvanshi, 2012). In the face of increasing population and stagnant wheat and rice productions, millets can be a promising alternative in solving the problem of food insecurity and malnutrition. The quality of protein is mainly a function of its essential amino acids. Finger millet contains 44.7% essential amino acids (Mbithi *et al.*, 2000) of the total amino acids, which is higher than the 33.9% essential amino acids in FAO reference protein (FAO, 1991). The characterization of the proteins of millet grains shows that prolamin fraction constitutes the major storage protein of the grain and lysine is the most limiting amino acid followed by cystine but millets are relatively high in methionine (Monteiro *et al.*, 1987; Sudharshana *et al.*, 1987; Kumar and Parmeswaran, 1999). The true digestibility and biological value of these millets ranges between 95.0 to 99.3 and 48.3 to 56.5 respectively (Geervani and Eggum, 1989b). Among the millets, pearl millet (Bajra) has the highest content of macronutrients, and micronutrients such as iron, zinc, Mg, P, folic acid and riboflavin, significantly rich in resistant starch, soluble and insoluble dietary fibres (Antony *et al.* 1996; Ragaee *et al.*, 2006). Finger millet seed coat is an edible material and contains good proportion of dietary fibre, minerals and phytochemicals. The seed coat matter (SCM) forms a by-product of millet milling, malting and decortication industries and can be utilized as composite flour in biscuit preparation (Krishnan *et al.*, 2011). Finger millet (ragi) is an extraordinary source of calcium. Kodo millet and little millet are also reported to have 37% to 38% of dietary fiber, which is the highest among the cereals and though low in fat - Jaybhaye *et al*

It is also rich in essential amino acids, like lysine, threonine, valine, sulphur containing amino acids and the ratio of leucine to isoleucine is about 2 (Ravindran, 1992; Antony *et al.*, 1996). Kodo millet has the highest free radical (DPPH) quenching activity followed by great millet (sorghum) and finger millet (Hegde and Chandra, 2005). Sorghum is exceptionally high in antioxidant activities followed by millets. Millets are valued for their high content of vitamin B, folic acid, phosphorus, iron and potassium. Finger millet contains 16 times more calcium than

maize. The niacin content in pearl millet is higher than all other cereals. In addition, millets are gluten-free, easy to digest and are a great source of antioxidants and might have anti-carcinogenic properties (Dykes and Rooney, 2006). The lipid content is generally high (3–6%) for pearl millet, higher than for sorghum and most other common cereals. About 75% of the fatty acids in pearl millet are unsaturated and linoleic acid is particularly high (46.3%). For this reason, energy content of millet is greater than sorghum and nearly equal to that of brown rice. Finger millet are good source of dietary calcium and magnesium and iron content is significant.

2.2.2 Traditional Uses of Millets

Millet grains are important as food for poor families to battle mal-nutrition and a chief source of income. In traditional systems, indigenous knowledge plays a central role in disease diagnosis and health care practices. Celiac disorder is an immune-mediated enteropathy which is triggered by intake of gluten rich food (Becker, Damiani, Melo, Borges, & de Barros Vilas Boas, 2014). Millets are gluten-free food and can be a substitute for celiac diseases and gluten sensitive patients (Annor, Marcone, Corredig, Bertoft, & Seetharaman, 2015; Saleh *et al.*, 2013). Polyphenols of millets exhibit inhibitory activity against its malt amylase, aldose reductase of cataract eye lenses and snake venom phospholipases (Mathanghi & Sudha, 2012). Phenolics of finger millet seed coat were reported to decrease hyperglycemia by blocking the α -amylase and α -glucosidase enzyme activity (Shobana, Sreerama, & Malleshi, 2009). Seed coat phenolics of finger millet are also reported as an inhibitor of cataractogenesis in human eye lenses (Chethan, 2008). Food processing techniques, such as soaking, germination, fermentation, and puffing of millets enhance the nutritional quality of millets, improve digestibility with reducing antinutrient content in their cereals (Handa, Kumar, Panghal, Suri, & Kaur, 2017; Jaybhaye & Srivastav, 2015). The presence of antinutrient in finger millet was reported to lower glycemic effect, reduced starch digestibility, and absorption (Kumari & Sumathi, 2002). Pearl millets were traditionally consumed for the treatment of celiac disorder, constipation and several

non-communicable diseases (Jnawali, Kumar, & Tanwar, 2016). In China, foxtail millet is highly appreciated for its high nutritional value, easily digestible and non-allergic properties and also it plays a significant role in human health. The fermented products of millets are consumed as probiotics and recommended for the treatment of diarrhea in young children (Manisseri & Gudipati, 2012; Nduti *et al.*, 2016).

2.2.3 Types of Millets

2.2.3.1 Finger Millet

Finger millet, one of the oldest crops in India is referred as “nrtta-kondaka” in the ancient Indian Sanskrit literature, which means “Dancing grain,” was also addressed as “rajika” or “markataka” (Achaya, 2009). Earliest report of finger millet comes from Hallur in Karnataka of India dating approximately 2300 BC (Singh, 2008). There is some debate as to the origin of finger millet, and there are theories that finger millet might have travelled to India by sea from Arabia or South Africa or across the Indian Ocean in both directions (Achaya, 2009).

Nutritionally, FM is considered a good source of essential nutrients. FM is substantially rich in minerals, thus being declared more micro-nutrient dense than rice and wheat. Table 1 depicts the nutritional composition attributed to FM grain. The millet contains about 65%–75% carbohydrates, 5%–8% protein, 15%–20% dietary fiber, 2.5%–3.5% minerals, and 1%–2% ether extractives (a portion of dry matter extracted with ether) (Devi *et al.*, 2014). The major portion of carbohydrates in FM includes starch (60%), pentosans (6.2%–7.2%), cellulose (1.4%–1.8%), and lignins (0.04%–0.6%) (Hassan *et al.*, 2021). Furthermore, FM also contains a higher amount of total dietary fiber (17.10%–18.90%) and insoluble dietary fiber (13%–wheat and rice. FMs' dietary fibers mainly include non-starch poly-saccharides, non- α -glucan oligosaccharides, resistance starches, some polyols, and modified starches (Lafiandra *et al.*, 2014). Non-starchy polysaccharides are primarily made up of “arabinoxylans”

(principal non-starch compound), with a small number of β -d-glucans, contributing the main component of the fraction of soluble dietary fiber in grain (Udeh *et al.*, 2017). It contains less protein content than other millets, it is enriched with a balanced amino profile and higher sulfur-containing amino acids. It is estimated that 44.7% of the amino acids in FM are essential amino acids, which is higher than wheat, rice, and the FAO reference protein (33.9%). There are abundant amounts of lysine and methionine in FM, which are lacking in other plant diets (Abioye *et al.*, 2022). In relation to nutritional value, the protein quality score of FMs is 52 higher than that of sorghum (37) and the protein efficiency ratio is 0.95, comparable to that of control casein (1.9) (Shobana *et al.*, 2013). The low-fat content in FM (1.3%–1.8%) makes it suitable for a good shelf life. The amounts of free, bound, and structural lipids in FM are 2.2%, 2.4%, and 0.6%, respectively. Regarding fatty acids, they comprise two beneficial polyunsaturated fatty acids, linoleic acid and α -linolenic acid, which play a vital role in metabolism and the normal development of the central nervous system. The major fatty acids found in decorticated FM, oleic acid (50.43%), palmitic acid (26.18%), and linoleic acid (20.26%), are the most abundant, while stearic acid (0.12%) and linolenic acid (2.60%) are present in relatively low concentrations (Mitharwal *et al.*, 2021). It is also the richest source of calcium (344 mg/100 g of FM) and potassium (408 mg/100 g FM) among all other cereal crops like brown rice, wheat, or maize (Gull *et al.*, 2014). It is also reported to contain an appropriate amount of iron (3.9 mg/100 g). Both water-soluble and fat-soluble vitamins, vitamin B1 (thiamin), vitamin B2 (riboflavin), vitamin B3 (niacin), and vitamin E (α -tocopherols), are present in significant amounts (Kumar *et al.*, 2016). It was reported that FM levels of vitamin B1, B2, and B3 were 0.38 mg/100 g, 0.14 mg/100 g, and 1.1 mg/100 g, respectively, which are equivalent to 35%, 12%, and 8% of the recommended daily allowance for healthy adults (Taylor & Kruger, 2016).

2.2.3.2 Foxtail Millet

Foxtail millet (*Setaria italica*) is one of the most important food crops of the semiarid tropics, beginning in China, continuing through India and extending over most of Africa and parts of Southern U.S.A. It plays a very important role in the agriculture and food of many developing countries because of its sustainability to grow under adverse heat and limited rainfall conditions. This cereal is also known as Italian millet. In India, this millet is grown primarily in the hot drought – prone arid and semiarid zones – and used mostly for food purposes especially by people of economically weaker sections. This millet contains 12.3% crude protein and 3.3% minerals (Gopalan et al. 1987). It could be milled to remove the husk, cooked and puffed for use in the breakfast and specialty foods and malted for use as an adjunct in brewing and also in the development of high-calorie density of weaning foods (Malleshi and Desikachar 1985). However, because the grains are coarse, the edible portion of this millet is only about 79%, an indication of the presence of a high amount of fiber. The fiber is associated with polyphenols and phytate, which reduce the availability of proteins and important minerals especially iron and zinc. The removal of these by simple processing techniques such as dehulling, soaking and cooking provides an opportunity to test this millet for protein and phytate and tannin contents.

2.2.3.3 Proso Millet

Proso millet was likely domesticated in China sometime around 10,000 BP. Current archeological theorists believe that proso millet domestication took place around the beginning of the Holocene as global temperatures became warmer and hunter-gatherers were exposed to new plants and environments (Bettinger *et al.*, 2007, 2010a,b). A wild ancestor for proso millet has yet to be identified (Miller *et al.*, 2016); however, weedy forms of millet, which may include a wild progenitor, are found across Eurasia (Zohary *et al.*, 2012). Chromosomal *in situ* hybridization with genomic DNA and phylogenetic data provide evidence of the allotetraploid origin of proso millet, with *Panicum capillare* or a close relative, and *Panicum repens* as ancestors (Hunt *et al.*, 2014).

The protein content in proso millet is around 11% (dry basis) (Kalinova and Moudry, 2006). They reported that the proso millet protein is richer in essential amino acids (leucine, isoleucine, methionine), compared to wheat. Hence, the protein quality of proso (Essential Amino Acid Index) was higher (51%) compared to wheat. (Lorenz and Dilsaver, 1980) conducted a thorough study on the milling characteristics and proximate composition and nutritive value of the proso-millet flours. Compared with wheat, millet flours were high in ash and crude fat and were higher in protein content.

2.2.4 Millet Milk

Millets have the potential to be a good source of dairy substitutes when these aspects are taken into account. It fills up the gap in other sources due to its high protein content, lower starch level, moderate flavour, and few calories. Millets are an excellent crop for use as a dairy substitute through value addition due to their widespread cultivation and reputation for thriving with little maintenance. Millet milk is primarily selected due to its better nutritional value compared to other plant milk sources. This makes it an excellent dairy substitute especially in the current trend toward high nutritional value and low-calorie diets (Raajeswari & Nithya, 2018). It was reported that millet milk retains its nutritional stability at both high and low processing temperatures (U. K. *et al.*, 2020).

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

This chapter deals with the materials used and methodology followed for the development and quality evaluation of millet-based yoghurt and are discussed under the following sections.

3.1 RAW MATERIAL

Pasteurized homogenized milk (Milma) were procured from milk booth. The millets viz., ragi, proso and foxtail were purchased from the supermarket. Greek starter culture containing strain such as *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, were purchased from M/S Tempeh Chennai and it was preserved under refrigerated condition till the conduct of experiment.

3.2 DETERMINATION OF PHYSIO-CHEMICAL PROPERTIES OF COW MILK AND MILLETS

By using the standard methods, physio-chemical properties of cow milk and millets (ragi, proso and foxtail) viz. colour, moisture content, ash, crude fat, crude fibre, crude protein and carbohydrate were determined.

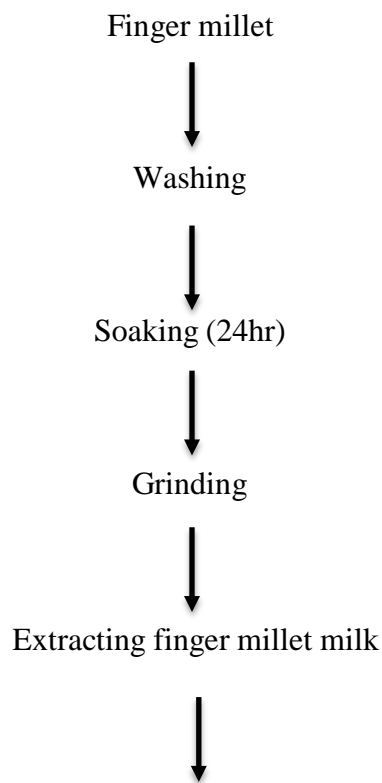
3.3 PREPARATION OF MILLET- BASED YOGHURT

3.3.1 Preparation of finger millet-based yoghurt

The finger millet about 500g was weighed and cleaned thoroughly with water. It was then soaked in water for 24 hr. After soaking period, the water was drained out and

millet was ground with water in the ratio 1:2 (water to millet ratio). The finger millet milk was extracted using muslin cloth.

Cow milk and the prepared finger millet milk were prepared and then thoroughly mixed for homogeneity. The mixed milk was then pasteurized in a cooking pot for 15min at 85°C. This was done to achieve a sterile and conducive environment for the starter culture and to denature and coagulate whey proteins to enhance the viscosity and texture of the product. The pasteurized millet milk was allowed to cool to a temperature of 43-45°C before it was inoculated with the starter culture for 6 hours at 40-45°C. The finger millet-based yoghurt was done. The flow chart for millet yoghurt production is shown below in fig 3.1



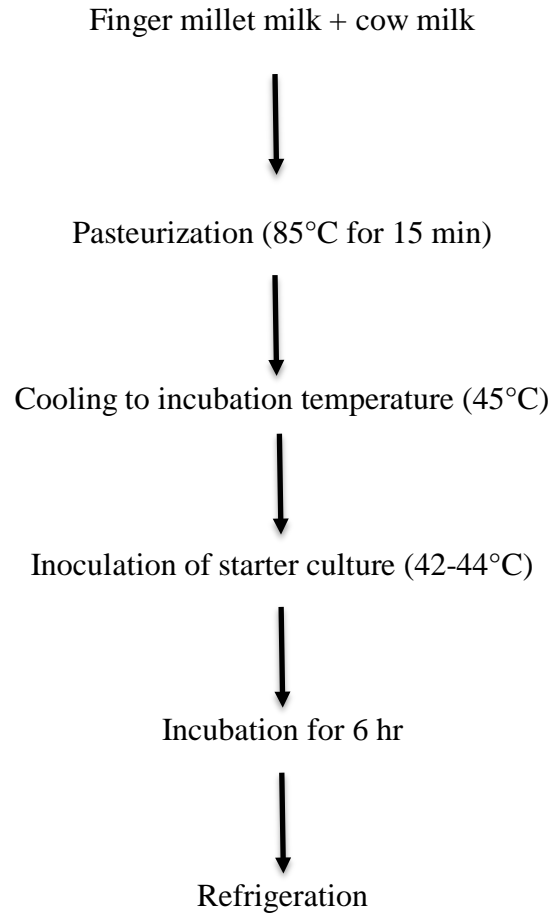
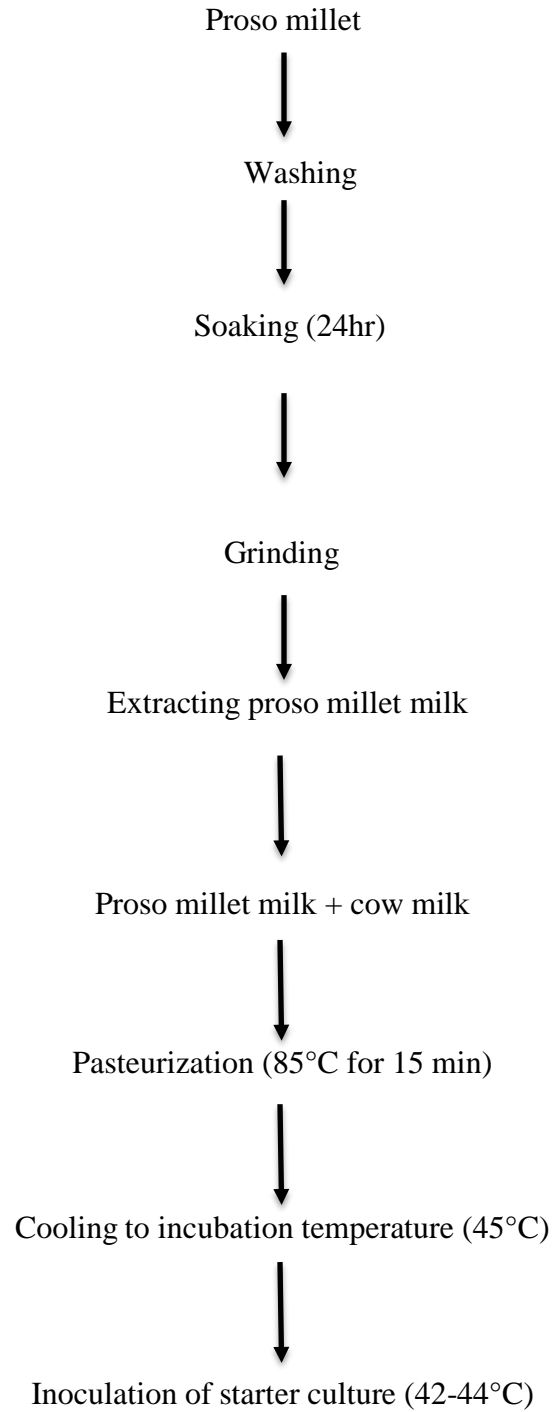


Fig 3.1 Preparation of finger millet-based yoghurt

3.3.2 Preparation of proso millet-based yoghurt

The proso millet-based yoghurt were prepared as per the procedure discussed in section 3.2.1 The flow chart for the production of proso millet yoghurt is given below in fig 3.2



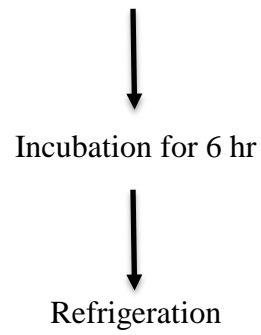
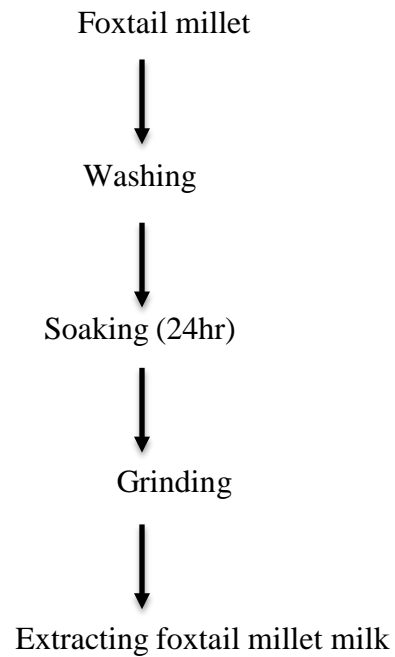


Fig.3.2 Preparation of proso millet-based yoghurt

3.3.3 Preparation of foxtail millet-based yoghurt

The foxtail millet-based yoghurt was prepared as per the procedure discussed in section 3.2.1. The flow chart for the production of proso millet yoghurt is given below in fig 3.3



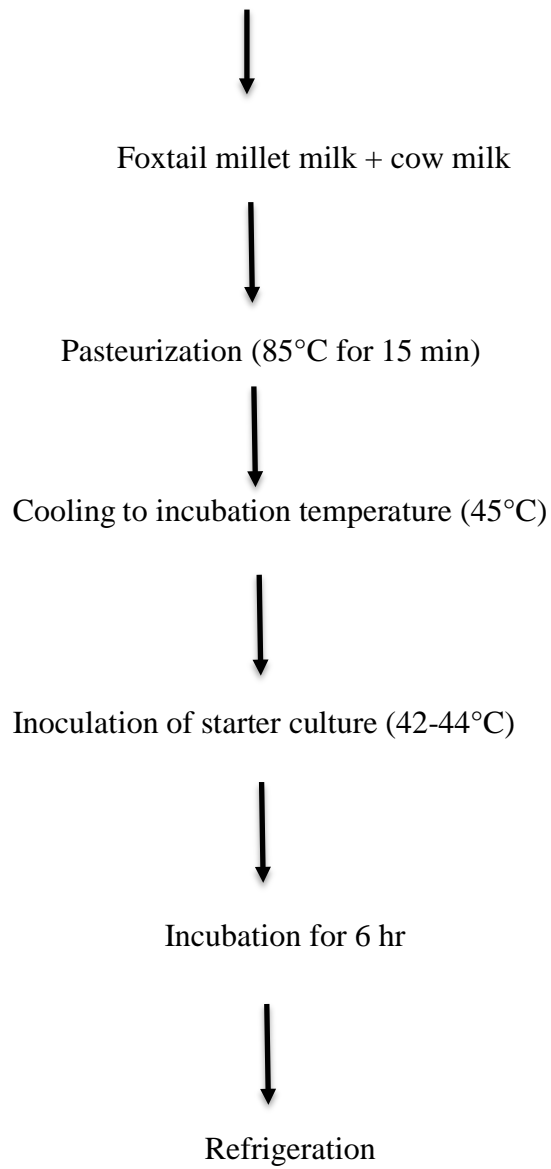


Fig 3.3 Preparation of foxtail millet-based yoghurt

Table 3.1: Yoghurt Samples Proportion

Yoghurt samples	Cow milk %	Millet milk %
Finger Millet (Ragi)		
R1	90	10
R2	80	20
R3	70	30
Foxtail Millet		
F1	90	10
F2	80	20
F3	70	30
Proso Millet		
P1	90	10
P2	80	20
P3	70	30
CONTROL	100	0

*R=Ragi, F=Foxtail Millet, P=Proso Millet, CONTROL=Control

3.4 EXPERIMENTAL DESIGN

Based on the detailed review of literature and the preliminary studies conducted, the independent variables were chosen as types of millet, millet and milk ratio. The sensory qualities and nutritional qualities were the dependent variables.

3.4.1 Development of Millet Based Yoghurt

a. Independent Variables

Types of Millet

- i. R : Finger Millet (Ragi)
- ii. F : Foxtail Millet
- iii. P : Proso Millet

Millet and Cow Milk Ratio (millet milk: cow milk)

- i. R1 : 10% Ragi milk and 90% cow milk
- ii. R2 : 20% Ragi milk and 80% cow milk
- iii. R3 : 30% Ragi milk and 70% cow milk
- iv. F1 : 10% foxtail milk and 10% cow milk
- v. F2 : 20% Foxtail milk and 80% cow milk
- vi. F3 : 30% Foxtail milk and 70% cow milk
- vii. P1 : 10% Proso milk and 90% cow milk
- viii. P2 : 20% Proso milk and 80% cow milk
- ix. P3 : 30% Proso milk and 70% cow milk

b. Dependant Variables

- pH
- TSS
- SNF
- Titratable Acidity
- Carbohydrate
- Crude protein
- Fat
- Ash
- Moisture content
- Sensory Attribute

3.5 SENSORY ANALYSIS OF YOGHURT SAMPLES

Samples of the product were evaluated using nine-point hedonic scale method and overall acceptability by 15 panelists drawn from students and staff of Kelappaji College of Agricultural Engineering and Technology, Tavanur, Malappuram.

They were served coded samples of the product and were asked to compare for appearance/colour, flavour, odour, taste, texture, mouthfeel, and overall acceptability using a 9-point Hedonic scale test with 9 indicating extreme acceptance and 1 indicating severe dissatisfaction as shown in the table below.

Table 3.2 9-point hedonic scale

9-Point Hedonic Scale	
Dislike Extremely	1
Dislike Very Much	2
Dislike Moderately	3
Dislike Slightly	4
Neither Like nor Dislike	5
Like Slightly	6
Like Moderately	7
Like Very Much	8
Like Extremely	9

The products were analysed by 15 untrained consumers. Nine samples were kept for sensory analysis including a control of fresh yoghurt purely made only from cow milk. To unify the conditions of the evaluation, all samples were prepared in disposable cups coded. Evaluation of these samples were done by each panellist in a monadic order, following a balanced incomplete box design (Stone *et al.*, 2020)

The samples were served altogether in one session and sample's identities were kept hidden. During the test, the panellist was asked to pause between the samples and cleanse their palates with prepared tap water at room temperature.

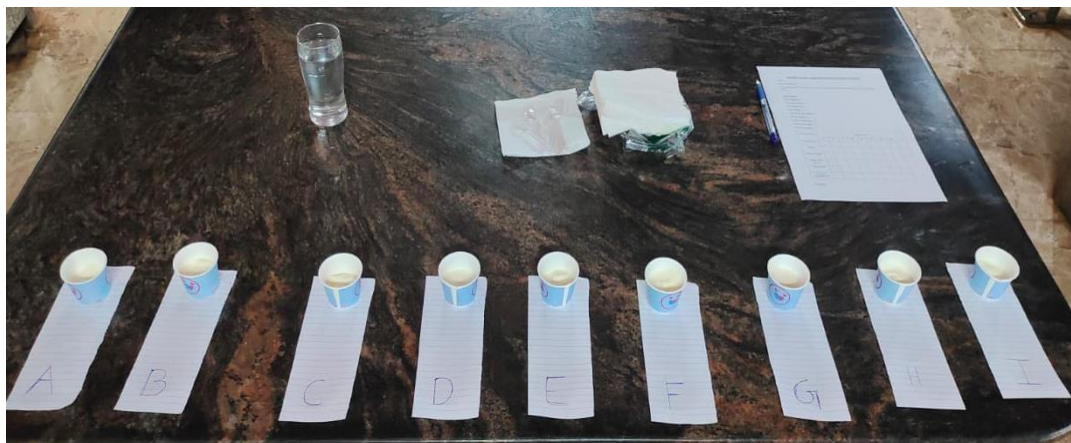


Fig.3.4 Sensory evaluation of the developed yoghurt samples

3.6 OPTIMIZATION OF PROCESS VARIABLE FOR THE PREPARATION OF YOGHURT SAMPLES

Nine treatments were selected in this study. The optimization of process parameters was done based on the sensory evaluation of the developed samples.

3.7 DETERMINATION OF PHYSIO - CHEMICAL PROPERTIES OF OPTIMIZED MILLET BASED YOGHURT SAMPLES

The physio-chemical properties of the optimized yoghurt samples were conducted and are discussed under subsequent sections.

3.7.1 pH measurement

The pH of the freshly prepared yoghurt samples was determined using a digital pH meter (M/s. Systronics; Model MK VI) shown in Plate 3.1. Initially, the pH meter was standardised with distilled water of pH 7.0 and standards of pH

4.0, 7.0 and 9.0. Samples was taken in a beaker and the electrode of pH meter was immersed in the sample. The reading was directly recorded from the pH meter. This procedure was repeated thrice for precision and the average value was noted (AOAC, 1990).



Plate 3.1. pH Meter

3.7.2 Total Soluble Solids

Total soluble solid (TSS) of freshly prepared yoghurt was measured using a hand refractometer (Erma inc, Tokyo) as shown in plate 3.2. One or two drops of the prepared sample were placed on the hand refractometer for TSS measurement. It was expressed in degree Brix (AOAC, 1990).



Plate 3.2. Refractometer

3.7.3. Total Titrable Acidity

Acidity of powder sample was determined by titration method (AOAC, 2005). 10 ml of diluted powder sample (one gram powder in 10 ml distilled water) was taken in a conical flask and two to three drops of phenolphthalein indicator were added to it. This solution was titrated against 0.1N NaOH until the point that light pink end point showed up. Volume of 0.1N NaOH used was recorded. This procedure was carried out three times and the average value was considered as acidity of the sample. The acidity of yoghurt sample was calculated using Eq (1).

% Acid

$$= \frac{\text{Eq. weight of acid} \times \text{titre value} \times \text{normality of NaOH} \times \text{volume made up} \times 100}{1000 \times \text{volume of aliquot taken for titration} \times \text{volume of sample}}$$

...Eq (1)

3.7.4 Proximate Analysis of Yoghurt Samples

3.7.4.1 Carbohydrates

Total Carbohydrate content of the yoghurt samples was determined by anthrone method. Carbohydrates are first hydrolysed into simple sugars using dilute hydrochloric acid. In hot acidic medium glucose is dehydrated to hydroxymethyl furfural. This compound forms with anthrone a green-coloured product with an absorption maximum at 630 nm.

In the boiling tube 100 mg of yoghurt samples was weighed using weighing balance as shown in plate 3.3 and hydrolysed for three hours with 5 ml of 2.5 N HCL by using boiling water bath and cooled to room temperature. Sodium carbonate is added to the above solution to neutralize it. Then the solution is made up to 100 ml and centrifuged to collect the supernatant. Then 0.5 ml and 0.1 ml of the supernatant solution was taken as test solution. The standards were prepared by taking 0, 0.2, 0.4, 0.6, 0.8 and 1 ml of the glucose working standard. In the test tube volume is made up to 1 ml in all tubes with distilled water. Then add 4 ml anthrone reagent to all tubes and heat for 8 minutes. Cool rapidly and read at 630 nm using spectrophotometer as shown in plate 3.4. Draw a standard graph by plotting concentration of the standard against absorbance and calculate the amount of carbohydrate in the yoghurt samples using the graph.

$$\begin{aligned} & \text{Amount of carbohydrate present in 100 mg of sample} \\ &= \frac{\text{mg of glucose} \times 100}{\text{volume of test sample}} \end{aligned}$$

...Eq (2)



Plate 3.3. Spectrophotometer

3.7.4.2 Protein

Protein estimation by Lowry's method involves various materials and reagents, including 2% Sodium Carbonate in 0.1 N Sodium Hydroxide (Reagent A), 0.5% Copper Sulphate in 1% potassium sodium tartrate (Reagent B), alkaline copper solution (Reagent C), and Folin-Ciocalteu reagent (Reagent D). The principle of the method relies on the development of a blue color resulting from the reduction of phosphomolybdic-phosphotungstic components in the Folin-Ciocalteu reagent by amino acids like tyrosine and tryptophan present in the protein. Additionally, the color developed by the biuret reaction of the protein with alkaline cupric tartrate is measured in Lowry's method.

The procedure involves the extraction of protein from the sample, typically carried out with buffers used for enzyme assays. The sample is weighed, ground, and centrifuged, and the supernatant is used for protein estimation. In the estimation step, working standards and sample extracts are pipetted into test tubes, and reagents C and D are added successively. After incubation, the developed blue color is measured at 660 nm, and a standard graph is drawn for protein quantification.

For calculation, the amount of protein is expressed in mg/g or 100 g of the sample.

3.7.4.2 Crude fat

The fat content of the sample was estimated by the method described by (Cohen, 1917). The lipid in the sample was extracted with petroleum ether (60-80°C) in Soxhlet apparatus for four hours as shown in plate 3.5. Then the solvent was evaporated and the remaining residue was weighed. The fat content was expressed as percentage.

$$\text{Crude fat \%} = \frac{(w_3 - w_2) \times 100}{w_1} \quad \dots \text{Eq (3)}$$

W_1 = Weight of sample used

W_2 = Weight of flask

W_3 = Weight of flask with fat residue

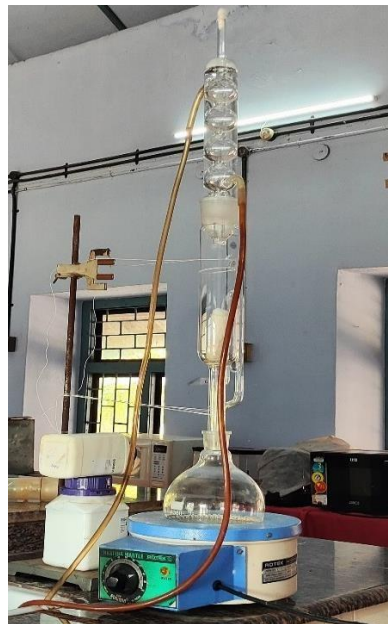


Plate 3.4 Soxhlet Apparatus

3.7.4.3 Crude fibre

Crude fibre content was determined as per the method described by (Maynard, 1970). The dried sample was taken in the pre weighed glass crucible (W₁) it was placed in crucible holder with the glass extractor. 150 ml of pre heated 1.25% H₂SO₄ was added in the extractor and the contents are boiled for 30 mins at 500°C and 30 mins for 400°C. The acid residue was drained out from the extractor through fibre flow system. The residue was washed with distilled water. Then 150 ml of pre heated 1.25% NaOH added and digested for 30 mins at 500°C and 30 mins at 400°C. Then the residue was washed with distilled water and dried for two to four hours at 100°C, cooled and weighted.

$$\text{Crude fat \%} = \frac{(w_3 - w_2) \times 100}{w_1} \quad \dots \text{Eq (4)}$$

W₁ = Weight of sample used

W₂ = Weight of crucible

W₃ = Weight of residue with crucible



Plate 3.5 Crude Fibre Apparatus

3.7.4.4 Moisture content

The moisture content of the yoghurt sample was determined using infrared moisture meter.



Plate 3.6. Infrared moisture meter

3.7.4.5 Ash Content

The ash content was determined by the direct heating method as contain in AOAC (2005). In this method, 2g each of the samples was measured into a crucible of known weight, the sample was burnt to ash in a muffle furnace for 3h at 5500C. It was then cooled in a desiccator and the weight of the ash was finally determined. The % Ash content was calculated as;

$$\% \text{ Ash} = \frac{w_1 - w_2}{w_1} \times 100 \quad \dots \text{Eq (5)}$$

Where;

W_1 = Initial weight of the sample
 W_2 =

Weight of the dried sample

3.7.4.6 Total Solids-Non-Fat

The total solids-not-fat was determined as described by AOAC (2005). It was obtained by taking the difference between % Total Solids and % Fat content.

That is:

$$\% \text{ Solids-Not-Fat} = \% \text{ Total Solids} - \% \text{ Fat content} \quad \dots \text{Eq (6)}$$

3.8 MICROBIAL ANALYSIS AND SHELF- L I F E STUDIES OF OPTIMIZED MILLET BASED YOGHURT SAMPLES

The analysis was done for the determination of total yeast and mould count. YGC agar (Yeast Extract Chloramphenicol Agar) were used for the microbial analysis. First is the sterilisation process in which agar media and glass wares were autoclaved at 121°C for 15 min to make them sterile.

To get 10^{-1} dilution, one gram of the powder sample was taken and added to 10 ml of sterile water blank. Suspension was shaken well for 10 to 15 min to get homogenised suspension of microorganisms and this gave a dilution of 10^{-1} . One ml from (10^{-1}) this dilution was transferred to 9 ml of sterile water blank with a sterile one ml pipette, which gave a dilution of 10^{-2} .

The process was repeated up to 10^{-7} dilutions with the sterile water blank. One ml aliquots from all dilutions were relocated to the sterile Petri dishes with respective agar media for the enumeration

of microbes. About 15-20 ml of growth media were poured to plates at temperature (45-50°C) and the plates were rotated clockwise and anticlockwise directions on a flat surface to have a uniform distribution. Plates were kept undisturbed until the agar gets solidify. After solidification, the plates were inverted and incubated at 42°C for 72 h. The colonies were counted after the incubation period and the number of colonies forming units per millilitre of sample and survival rates were calculated by applying the following formulas:

$$\frac{CFU}{ml} = \frac{\text{mean no. of CFUs} \times \text{Dilution Factor}}{\text{Volume of sample plated (ml)}} \quad \dots \text{Eq (7)}$$

RESULTS AND
DISCUSSIONS

CHAPTER IV

RESULTS AND DISCUSSIONS

This chapter presents the details of the development of millet-based yoghurt as well as the results of the observations and analysis carried out in the study. The physio-chemical analysis of cow milk, millets and millet-based yoghurts, optimization for process parameter of yoghurt and shelf-life studies of the optimized yoghurt are elaborated in this chapter.

4.1. PHYSIO -CHEMICAL PROPERTIES OF COW MILK AND MILLETS

The physio-chemical properties of cow milk, finger millet, proso millet and foxtail millet are given below.

Table 4.1 Physio-chemical properties of cow milk, finger millet, proso millet and foxtail millet.

PROPERTIES	RAW MATERIALS			
	Cow Milk	Finger Millet	Proso Millet	Foxtail Millet
Colour	L*=81.0±8.1 a*=-1.5±3 b*=7.5±4.4	L*=19.23±0.42 a*=18.28±0.81 b*=19.38±0.15	L*=76.14±0.75 a*=56±0.05 b*=1.57±0.19	L*=61.38±2.21 a*=3.91±0.05 b*=16.2±0.65
Moisture content (%)	87.2	7.8	8.34	10.46
Ash (%)	0.72	2.7	1.66	0.47
Crude fat(g)	3.7	1.8	4.9	2.38

Crude fibre(g)	0	3.5	0.7	8
Crude Protein(g)	3.5	8.2	11.58	11.5
Carbohydrate(g)	4.9	83.3	80.1	75.2

4.2 DEVELOPMENT OF MILLET-BASED YOGHURT

Millet-based yoghurt samples was developed from three types of millets viz. finger millet, foxtail millet, proso millet. For the optimization of millet-basedyoghurt nine types of treatments were made by selecting difference in concentration of millet milk and cow milk as a process parameter. The nine millet-based yoghurt samples developed are listed in table 3.1. These nine millets- based yoghurt samples were then kept for sensory analysis for optimization.

4.3 SENSORY ANALYSIS OF DEVELOPED YOGHURT SAMPLES

The developed millet-based yoghurt along with control was subjected to sensory evaluation by a panel of 15 untrained members by nine point-hedonic scale.

Following is the 9 developed millet-based yoghurt samples

- i. R1 : 10% Ragi milk and 90% cow milk
 - ii. R2 : 20% Ragi milk and 80% cow milk
 - iii. R3 : 30% Ragi milk and 70% cow milk

- iv. F1 : 10% Foxtail milk and 10% cow milk
- v. F2 : 20% Foxtail milk and 80% cow milk
- vi. F3 : 30% Foxtail milk and 70% cow milk
- vii. P1 : 10% Proso milk and 90% cow milk
- viii. P2 : 20% Proso milk and 80% cow milk
- ix. P3 : 30% Proso milk and 70% cow milk

The sensory evaluation was carried out on the basis of appearance, flavour, odour, texture, taste and overall acceptability of the developed product. The sensory evaluation of the millet enriched product revealed that there were significant differences among the treatments for the organoleptic qualities. Considering the results of sensory evaluation, the best product was selected. The selected product was the one which was of good taste and overall acceptability. and were given in Table 4.2. P1 and R2 are the two product that finally selected after sensory analysis for further methods and determination.

The sample that had the highest appearance acceptability was CONTROL and the sample with the lowest appearance acceptability was F1. There was no significant difference among the appearance in F2, F3 and P3 samples. There was no significant difference among P1 and R3 samples in appearance. The sample CONTROL had a significant difference in appearance from the other samples. The samples enriched with finger millet and proso millet had the best appearance acceptability than the other enrichment sources.

The samples enriched with R2 and P1 has the most acceptable taste than those enriched with other millet sources. CONTROL had the highest taste quality and F3 had the least taste quality. There was no significant difference between the value of F2 and P3 samples in taste.

CONTROL had the highest texture parameter value. There was no significant difference between F2 and P3 samples and also R2, F3, F1 and P2 samples. R3 had the lowest aroma property value and P1 and P3 had the highest. The samples enriched with ragi millet had the least aroma acceptability and proso millet having the best among the millet enriched sources.

CONTROL had the highest mouthfeel value and then R1 and P1 had the highest mouthfeel value. There was no significant difference among P1 and R1 samples in mouthfeel. The sample with the highest overall/general acceptability was recorded as CONTROL and F1 had the lowest. The sample enriched with proso millet had the highest overall/general acceptability than the other enrichment sources. There were no significant difference between P3 and R3 samples in general acceptability.

Among the enriched yoghurt samples, P1 was the most preferred after control and then R2 was the most preferred as shown in fig4.1. P1 and R2 were the two products that was finally selected for further studies.

Table 4.2 Sensory evaluation

SAMPLE	R1	F2	P3	R2	F3	P1	R3	F1	P2	CONTROL
CODE	A	B	C	D	E	F	G	H	I	J
APPEARANCE	7	7.6	7.6	7.1	7.5	6.9	6.8	7.2	7.5	8
ODOUR	6.7	6.9	7	6.6	6.5	7	6.2	6.3	6.8	7.5
FLAVOUR	6.5	6.2	6.2	7.1	5.5	7	6.7	5.6	6	7.8
TEXTURE	6.9	7.2	7.2	6.7	6.7	7.3	6.9	6.7	6.7	8

MOUTHFEEL	7.2	6	6.3	7	6.1	7.2	6.5	6.1	6.3	7.9
OVERALL ACCEPTABILITY	7.1	6.5	7	7.1	6.1	7.5	7	5.8	6.8	8

*R = Ragi milk-based yoghurt, F = Finger milk-based yoghurt, P = Proso milk-based yoghurt, CONTROL = Control

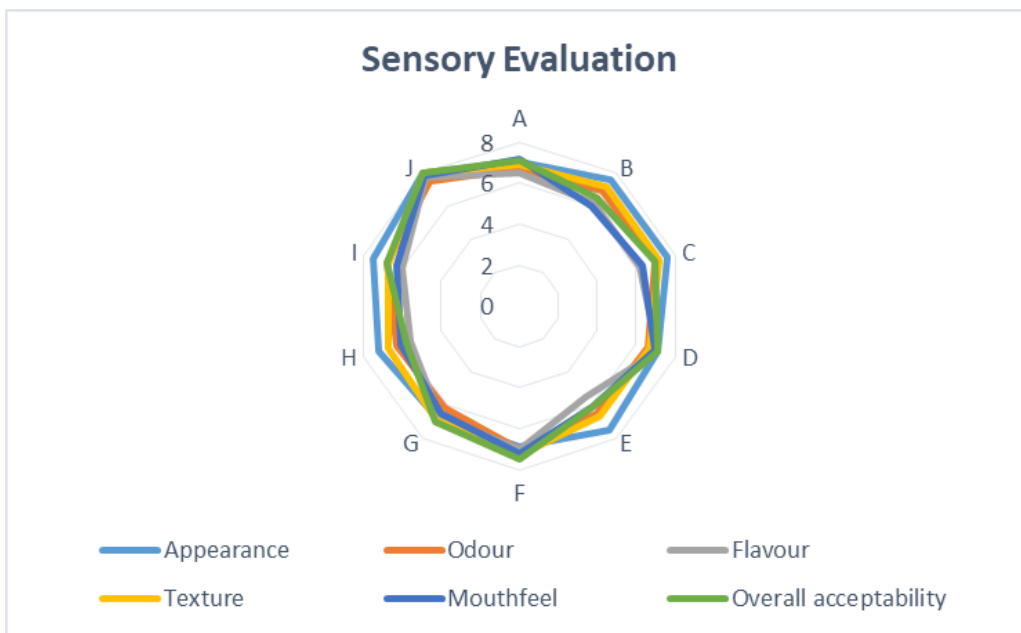


Fig 4.1 Radar chart of sensory evaluation

4.4 OPTIMIZATION OF PROCESS VARIABLE FOR THE PREPARATION OF YOGHURT SAMPLES

Based on the sensory evaluation of the developed nine treatments P1 and R2 was selected for further studies. P1 contains 10% proso milk and 90% cow milk and R2 contains 20% ragi milk and 80% cow milk.

4.5 PHYSIO- CHEMICAL COMPOSITION OF OPTIMIZED YOGHURT SAMPLE

4.5.1.1 pH

The pH content of the samples R2, P1 and CONTROL was recorded as 4.43,4.53, and 4.6 respectively. The pH of the samples enriched with ragi and prosomillet shows a slight difference than the control sample. This suggests that therate of acid generation by the starter culture was affected by the addition of millet milk.

Ademosun *et al.*, (2019) enriched yoghurt with tomato juice and reported pH level which ranged 4.14 to 4.25. Ezeonu *et al.*, (2016) conducted analysis on coconut, tiger nut and fresh cow milk yoghurt, and was reported the pH to range from 4.21 to 51 4.52. %. Mbaeyi-Nwaohaet *et al.*, (2017) analyzed flavoured yoghurt enriched African bush mango and recorded the pH to rangefrom 4.69 to 5.01. The result of the pH levels recorded above shows that the pH of plant-based yoghurt enrichments is along the same pH levels of acidity. According to FDA standards for yoghurt pH (4.6 or below), the pH findings for yoghurt are within specifications and within the Food Health 9(1),43-60 (2023).

4.5.2 TSS

TSS of yoghurt samples was tested by using a hand refractometer. The TSS of the samples R2, P1 and CONTROL was recorded as 9.3°B, 10°B and 8.5 °B. Result shows that there is an increase in TSS with the addition of millet milk. This might be due to the increased solid content in millet milk. Similar result was observed by Saranya (2015) in the case of microencapsulated banana pseudo stem juice powder.

4.5.2.1 Titrable acidity

The total titratable acidity of the samples was 0.99%, 0.93%, 0.9% for R2, P1, CONTROL respectively. The samples enriched with ragi increased in total titratable acidity when compared with proso and control. The data analyzed showed that yoghurt samples enriched with plant-based sources had more titratable acid than yoghurt. This could be due to the acid content of the individual plant products. Ezeonu *et al.*, (2016) conducted analysis on coconut, tiger nut and fresh cow milk yoghurt, and reported the titratable acid of the yoghurt samples to have ranged from 0.60 to 0.81%.

Table 4.3 Physio-chemical composition of optimized yoghurt samples

Samples	pH	Total Soluble Solids(°B)	Titrable Acidity (%)
R2	4.43	9.3	0.99
P1	4.53	10	0.93
CONTROL	4.7	8.5	0.9

4.5.3 Proximate Analysis of optimized yoghurt sample

4.5.3.1 Moisture Content

The moisture content of the samples was 85%, 82.01% and 84.3% for CONTROL, P1 and R2. This value corresponded with the report by Ahmad, (1994) who stated that the maximum moisture content of yoghurt should be 84% , as much water in yoghurt makes it less viscous thereby affecting texture and mouth feel.

4.5.3.2 Ash Content

The ash content of the samples was 0.52%, 0.61% and 0.79 % for CONTROL, P1 and R2 respectively. The results showed significant difference between all the samples in their ash content. The ash value is an index of mineral content, which is needed for bone development, teeth formation and body functions (Trachoo and. better source of minerals among the samples.

4.5.3.3 Carbohydrate

The carbohydrate content of the samples was 4.63 g, 5.71g and 6.21g for R2, P1 and CONTROL respectively. The low carbohydrate value is attributed to the process of fermentation which converts carbohydrate basically lactose to lactic acid. This makes yoghurt an ideal food for lactose intolerance individuals (Ehirim and Ndimantang, 2004). The reduction in the carbohydrate content of enriched yoghurt samples reduced due to higher availability of other proximate parameters like protein, ash, moisture and due to the conversion of lactose into lactic acid.

4.5.3.4 Crude Protein

The crude protein content of the samples was 3.74 g, 3.226g and 3.051 g for R2, P1 and CONTROL respectively. The result shows that yoghurt samples enriched with millets has more protein due to the higher protein content in plant-based sources that contribute to the enhancement of protein of yoghurt. The protein obtained for enriched yoghurt sample agreed with the findings of Kale et al. (2011) who reported protein content of date yoghurt was in the range of 3.45g to 3.47g and Gupta *et al.*, (2022) for commercially available dairy and plant-based yogurts.

4.5.3.5 Fat

The fat content of the sample P1, R2 and CONTROL was recorded as 2.08g, 2.57g, 3.79g respectively. The yogurt typically has lower fat content than milk because some of the milk fat is removed during the yogurt-making process. The addition of beneficial bacteria cultures, such as *Lactobacillus bulgaricus* and *Streptococcus thermophilus*, to milk initiates the fermentation process. These bacteria consume lactose, the milk sugar, and convert it into lactic acid. This acidification leads to the denaturation of proteins in the milk, causing them to coagulate and trap some of the fat. Additionally, plant-based milk has no fat when compared to the animal-based milk. It is also due to the use of low-fat milk during the preparation of yogurt.

4.5.3.6 Crude Fibre

The fibre content of finger millet-based yoghurt samples was 0.234g and proso millet was 0.117g as compared to milk-yoghurt (0.02g). There was an increased fibre content for millet-based yoghurt as compared to milk yoghurt, this might be due to the

high fibre content of the millets. According to Schneeman (2002) the crude fibre contributes to the health of the gastrointestinal system and metabolic system in man.

4.5.3.7 SNF

The SNF of the prepared samples were 6.93, 7.92, and 4.71 for R2, P1 and CONTROL respectively.

Table 4.4 Proximate analysis of optimized yoghurt samples

Samples	R2	P1	CRL
Moisture Content(%)	84.3	82.01	85
Ash Content(%)	0.79	0.61	0.52
SNF(%)	6.93	7.92	4.71
Carbohydrate(g)	4.63	5.71	6.21
Crude Protein (g)	3.74	3.226	3.051
Crude fibre(g)	0.234	0.117	0.02
Fat(g)	2.57	2.08	3.79

*R2= 20% finger millet milk based yoghurt, P1=10% proso millet milk based yoghurt, CRL=Control

4.6 MICROBIAL ANALYSIS AND SHELF-LIFE STUDIES OF OPTIMIZED YOGHURT SAMPLES

The total yeast and mould count of the selected samples are given in the table 4.2.

It is seen that the yeast and mould count for all the selected samples at initial day was 0, however on 14th day the yeast and mould count of finger millet-based yoghurt sample was increased to 1×10^{-2} as shown in fig 4.2. There was no yeast and mould growth found in P1 sample

Table 4.5 Total yeast and mould count

Sample Code	Day	Yeast and Mould Count
R2	0 th	Nil
P1		Nil
CRL		Nil
R2	14 th	1×10^{-2}
P1		Nil
CRL		Nil

R2 = 20% ragi milk and 80% cow milk

P1 = 10% proso milk and 90% cow milk

CRL = 100 % cow milk



Fig 4.2. Day 14 th YGC Agar Plate

SUMMARY AND CONCLUSION

CHAPTER 5

SUMMARY AND CONCLUSION

Currently the relevance of diet on healthy wellbeing has increased, which results in the rising demand for food products that support health along with supply of adequate nutrients. One among these products is yoghurt which is made from milk. Yoghurts are prepared by fermentation of milk with bacterial cultures (mixture of *Streptococcus thermophilus* and *Lactobacillus bulgaricus*). Fermentation of milk is considered as one of the oldest methods to preserve milk with an extended shelf life. Yoghurt is effectively digestible and highly nutritious dairy product and also a rich source of more than ten essential nutrients. The nutritional composition of yoghurt relies on the strains of starter culture used for fermentation, type of milk used (whole, semi or skimmed milk), species that milk is obtained (bovine, goat, sheep), type of milk solids, solid non-fat, sweeteners and fruits added before fermentation and the duration of fermentation process. *Lactobacillus* is beneficial and important in the fermentation of yoghurt; it is a probiotic which provides essential health benefits in the stomach and Gastrointestinal Tracts of humans and aids bowel movements. It also converts lactose sugar to lactic acid in milk which brings about curd/yoghurt formation in milk.

The project work entitled Development and Evaluation of Millet-Based Yoghurt are summarized as follows: -

The physiochemical properties of raw milk, millets, yoghurt viz. were determined using the standard procedures. Based on the literature review and preliminary studies, nine millet-based yoghurt samples were selected in this study. The optimization of process parameters was conducted based on sensory analysis. The optimized samples only were considered for further studies. Shelf-life studies of the optimized yoghurt samples were conducted based on microbial studies. Samples were

stored at refrigerated temperature of 3°C to 4°C for 14 days. Yeast and mould growth studies were conducted every 3 days interval.

The result of the study is furnished below:

The moisture content, ash content, crude fat, crude fibre, crude protein, and carbohydrate of raw milk was found to be 87.2%, 0.72%, 3.7g, 0g, 3.5g, and 4.9g respectively and colour characteristics of cow milk were found to be $L^*=81.0\pm 8.1$, $a^*=-1.5\pm 3$, $b^*=7.5\pm 4.4$. The moisture content, ash content, crude fat, crude fibre, crude protein, and carbohydrate of finger millet was found to be 7.8%, 2.7%, 1.8g, 3.5g, 8.2g, and 83.3g respectively and the colour characteristics $L^*=19.23\pm 0.42$, $a^*=18.28\pm 0.81$, $b^*=19.38\pm 0.15$. The moisture content, ash content, crude fat, crude fibre, crude protein, and carbohydrate of proso millet was found to be 8.34%, 1.66%, 4.9g, 0.7g, 11.58g, and 80.1g respectively and colour characteristics of cow milk were found to be $L^*=76.14\pm 0.75$, $a^*=56\pm 0.05$, $b^*=1.57\pm 0.19$. The moisture content, ash content, crude fat, crude fibre, crude protein, and carbohydrate of foxtail millet was found to be 10.46%, 0.47%, 2.38g, 8g, 11.5g, and 75.2g respectively and colour characteristics of cow milk were found to be $L^*=61.38\pm 2.21$, $a^*=3.91\pm 0.05$, $b^*=16.2\pm 0.65$.

From the preliminary studies using the three types of millet viz. finger millet, proso millet and foxtail millet nine samples were developed and kept for sensory analysis. Based on the sensory analysis P1(10% proso millet milk and 90% cow milk) and R2(20% finger millet milk and 80% cow milk) were the two samples that were finally selected for further studies.

The results of physio-chemical properties of P1 viz. pH, TSS, titrable acidity, moisture content, ash content, carbohydrate, crude protein, fat, crude fibre, SNF are 4.53, 10 brix, 0.93%, 82.01%, 0.61%, 5.71g, 3.226g, 2.08g, 0.117g, 7.92g respectively. The results of physio-chemical properties of R2 viz. pH, TSS, titrable acidity, moisture content, ash content, carbohydrate, crude protein, fat, crude fibre, SNF are 4.43, 9.3 brix, 0.99%, 84.3%, 0.79%, 4.63g, 3.74g, 0.234g, 2.57g, 6.93 g respectively. The results of physio-chemical properties of milk yoghurt viz. pH, TSS, titrable acidity,

moisture content, ash content, carbohydrate, crude protein, fat, crude fibre, SNF are 4.7, 8.5 brix, 0.9%, 85%, 0.52%, 6.21g, 3.051g, 3.79g, 0.02g, 4.71g respectively. It was observed that the yeast and mould count for all the selected samples at initial day was 0, however on 14th day the yeast and mould count of finger millet-based yoghurt sample was increased to 1×10^{-2} but is within the permissible limit. There was no yeast and mould growth found in P1 sample. From the microbial studies it is concluded that millet-based yoghurt sample is safe for consumption.

Further studies may be done on using extracted milk from plant-based sources so as to improve the nutritional profile and sensory acceptability of millet enriched yoghurts. Further studies maybe done on the development of new food products with millets to improve the availability of essential minerals and vitamins like iron, vitamin c etc. This project is the first step towards the development of vegan based millet yogurt which can be referred for further studies in the future.

REFERENCE

- Achaya, K. T. (2009). *The illustrated foods of India: A-Z*. Oxford University Press.
- Ajeesh Krishna, T. P., Maharajan, T., Antony Ceasar, S., & Ignacimuthu, S. (2023). Zinc supply influences the root-specific traits with the expression of root architecture modulating genes in millets. *Journal of Soil Science and Plant Nutrition*, 1-15.
- Antony, U., & Chandra, T. S. (1998). Antinutrient reduction and enhancement in protein, starch, and mineral availability in fermented flour of finger millet (*Eleusine coracana*). *Journal of Agricultural and Food Chemistry*, 46(7), 2578-2582.
- AOAC (2005) Official method of Analysis. 18th Edition, Association of Officiating Analytical Chemists, Washington DC, Method 935.14 and 992.2
- Ashraf, R., & Shah, N. P. (2011). Selective and differential enumerations of *Lactobacillus delbrueckii* subsp. *bulgaricus*, *Streptococcus thermophilus*, *Lactobacillus acidophilus*, *Lactobacillus casei* and *Bifidobacterium* spp. in yoghurt—A review. *International journal of foodmicrobiology*, 149(3), 194-208.
- Autieri, S. M., Lins, J. J., Leatham, M. P., Laux, D. C., Conway, T., & Cohen, P. S. (2007). 1- Fucose stimulates utilization of d-ribose by *Escherichia coli*. *Infection and immunity*, 75(11), 5465-5475.
- Baryeh, E. A. (2002). Physical properties of millet. *Journal of Food Engineering*, 51(1), 39-46.
- Becker, F. S., Damiani, C., de Melo, A. A. M., Borges, P. R. S., & de Barros Vilas Boas, E. V. (2014). Incorporation of buriti endocarp flour in gluten-free whole cookies as potential source of dietary fiber. *Plant Foods for Human Nutrition*, 69, 344-350.

- Blandino, A., Al-Aseeri, M. E., Pandiella, S. S., Cantero, D., & Webb, C. (2003). Cereal-based fermented foods and beverages. *Food research international*, 36(6), 527-543.
- Boachie, R. T., Okagu, O. D., Abioye, R., Hüttmann, N., Oliviero, T., Capuano, E., ... & Udenigwe, C. C. (2022). Lentil protein and tannic acid interaction limits in vitro peptic hydrolysis and alters peptidomic profiles of the proteins. *Journal of Agricultural and Food Chemistry*, 70(21), 6519- 6529.
- Bora, P., Ragaee, S., & Marcone, M. (2019). Characterisation of several types of millets as functional food ingredients. *International journal of food sciences and nutrition*, 70(6), 714-724.
- Buratti, S., & Benedetti, S. (2016). Alcoholic fermentation using electronic nose and electronic tongue. In *Electronic noses and tongues in food science* (pp. 291-299). Academic Press.
- Chethan, S., Dharmesh, S. M., & Malleshi, N. G. (2008). Inhibition of aldose reductase from cataracted eye lenses by finger millet (*Eleusine coracana*) polyphenols. *Bioorganic & medicinal chemistry*, 16(23), 10085-10090
- Devi, M. C., Meera, N. S., Theja, P. C., & Gopal, D. V. R. S. (2011). Production and optimization of β -galactosidase enzyme from probiotic *Lactobacillus* sps. *BioTechnol Indian J*, 5(3), 153-159.
- Devi, P. B., Vijayabharathi, R., Sathyabama, S., Malleshi, N. G., & Priyadarisini, V. B. (2014). Health benefits of finger millet (*Eleusine coracana* L.) polyphenols and dietary fiber: a review. *Journal of food science and technology*, 51, 1021-1040.
- Dewi, E. N., & Purnamayati, L. (2021). Characterization of *Caulerpa racemosa* yogurt processed using *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. *Food Research*, 5(3), 54-61.

- Dykes, L., & Rooney, L. W. (2006). Sorghum and millet phenols and antioxidants. *Journal of cereal science*, 44(3), 236-251.
- El-Abbadi, N. H., Dao, M. C., & Meydani, S. N. (2014). Yogurt: role in healthy and active aging. *The American journal of clinical nutrition*, 99(5), 1263S-1270S.
- Ezeonu, C. S., & Ejikeme, C. M. (2016). Qualitative and quantitative determination of phytochemical contents of indigenous Nigerian softwoods. *New Journal of Science*, 2016, 1-9.
- Ezeonu, C. S., Tatah, V. S., Nwokwu, C. D., & Jackson, S. M. (2016). Quantification of physicochemical components in yoghurts from coconut, tiger nut and fresh cow milk. *Advances in Biotechnology and Microbiology*, 1(5), 555-573.
- Fisberg, M., & Machado, R. (2015). History of yogurt and current patterns of consumption. *Nutrition reviews*, 73(suppl_1), 4-7.
- Geervani, P., & Eggum, B. O. (1989). Nutrient composition and protein quality of minor millets. *Plant Foods for Human Nutrition*, 39, 201-208
- Gilliland, S. E. (1990). Health and nutritional benefits from lactic acid bacteria. *FEMS Microbiology reviews*, 7(1-2), 175-188.
- Gull, A., Jan, R., Nayik, G. A., Prasad, K., & Kumar, P. (2014). Significance of finger millet in nutrition, health and value-added products: a review. *Magnesium (mg)*, 130(32), 120.
- Hadimani, N. A., & Malleshi, N. G. (1993). Studies on milling, physico-chemical properties, nutrient composition and dietary fibre content of millets. *Journal of Food Science and Technology (India)*, 30(1), 17-20.
- Hegde, P. S., Rajasekaran, N. S., & Chandra, T. S. (2005). Effects of the antioxidant properties of millet species on oxidative stress and glycemic status in alloxan-induced rats. *Nutrition Research*, 25(12), 1109-1120.

Hunt, H. V., Badakshi, F., Romanova, O., Howe, C. J., Jones, M. K., & Heslop-Harrison, J. P. (2014). Reticulate evolution in *Panicum* (Poaceae): the origin of tetraploid broomcorn millet, *P. miliaceum*. *Journal of Experimental Botany*, *65*(12), 3165-3175.

Hols, P., Hancy, F., Fontaine, L., Grossiord, B., Prozzi, D., Leblond-Bourget, N., ... & Kleerebezem, M. (2005). New insights in the molecular biology and physiology of *Streptococcus thermophilus* revealed by comparative genomics. *FEMS microbiology reviews*, *29*(3), 435-463.

Inyang CU, Zakari UM. Effect of germination and fermentation of pearl millet on proximate, chemical and sensory properties of instant Fura-A Nigerian cereal food. *Pakistan Journal of Nutrition*. 2008; *7*(1):9-12.

Jaybhaye, R. V., Pardeshi, I. L., Vengaiah, P. C., & Srivastav, P. P. (2014). Processing and technology for millet-based food products: a review. *Journal of ready to eat food*, *1*(2), 32-48.

Jnawali, P., Kumar, V., & Tanwar, B. (2016). Celiac disease: Overview and considerations for development of gluten-free foods. *Food Science and Human Wellness*, *5*(4), 169-176.

Kalsi, R., Bhasin, J., Goksen, G., & Kashyap, P. (2023). Exploration of nutritional, pharmacological, and the processing trends for valorization of finger millet (*Eleusine coracana*): A review. *Food Science & Nutrition*, *11*(11), 6802-6819.

Kaur, R., Kaur, G., Mishra, S. K., Panwar, H., Mishra, K. K., & Brar, G. S. (2017). Yogurt: A nature's wonder for mankind. *International Journal of Fermented Foods*, *6*(1), 57-69.

Krishnan, R., Dharmaraj, U., & Malleshi, N. G. (2012). Influence of decortication, popping and malting on bioaccessibility of calcium, iron and zinc in finger millet. *LWT-Food Science and Technology*, *48*(2), 169-174

- Kumar, A., Tomer, V., Kaur, A., Kumar, V., & Gupta, K. (2018). Millets: a solution to agrarian and nutritional challenges. *Agriculture & food security*, 7(1), 1-15.
- Lafiandra, D., Riccardi, G., & Shewry, P. R. (2014). Improving cereal grain carbohydrates for diet and health. *Journal of cereal science*, 59(3), 312-326.
- Lakshmi Kumari, P., & Sumathi, S. (2002). Effect of consumption of finger millet on hyperglycemia in non-insulin dependent diabetes mellitus (NIDDM) subjects. *Plant Foods for Human Nutrition*, 57, 205-213
- Lange, I., Mleko, S., Tomczynska-Mleko, M., Polishchuk, G., Janas, P., & Ozimek, L. (2020). Technology and factors influencing Greek-style yogurt—a Review.
- Leite, A., Neto, G. C., Vettore, A. L., Yunes, J. A., & Arruda, P. (1999). The prolamins of sorghum, Coix and millets. *Seed proteins*, 141-157.
- Lorenz, K., Dilsaver, W., & Bates, L. (1980). Proso millets. Milling characteristics, proximate compositions, nutritive value of flours. *Cereal Chem*, 57(1), 16-20.
- Mal, B., Padulosi, S., & Bala Ravi, S. (2010). Minor millets in South Asia: learnings from IFAD-NUS Project in India and Nepal.
- Malleshi, N. G., & Desikachar, H. S. R. (1985). Milling, popping and malting characteristics of some minor millets. *Journal of Food Science and Technology, India*, 22(6), 400-403.
- Maringe, C., Spicer, J., Morris, M., Purushotham, A., Nolte, E., Sullivan, R., & Aggarwal, K. K., & Athmaselvi, K. A. Millet Milk as a substitute for Dairy Milk.
- Mathanghi, S. K., Kanchana, S., & Perasiriyana, V. (2020). Pinnacles of Proso millet (*Panicum miliaceum* L.): A nutri millet. *Trop Plant Res*, 7(1), 238-244.
- Maynard Smith, J. (1970). Natural selection and the concept of a protein space. *Nature*, 225(5232), 563-564.

- Mbaeyi-Nwaoha, I. E., Nnagbo, C. L., Obodoechi, C. M., Nweze, B. C., & Okonkwo, T. M. (2017). Production and evaluation of yoghurt contained local stabilizers- *Brachystegia eurycoma* ('Achi') and *Detarium microcarpum* ('Ofo'). *International Journal of Biotechnology and Food Science*, 5(2), 23-31.
- Mbithi-Mwikya, S., Ooghe, W., Van Camp, J., Ngundi, D., & Huyghebaert, A. (2000). Amino acid profiles after sprouting, autoclaving, and lactic acid fermentation of finger millet (*Eleusine coracana*) and kidney beans (*Phaseolus vulgaris* L.). *Journal of agricultural and food chemistry*, 48(8), 3081-3085
- Mbithi-Mwikya, S., Van Camp, J., Yiru, Y., & Huyghebaert, A. (2000). Nutrient and antinutrient changes in finger millet (*Eleusine coracana*) during sprouting. *LWT-Food Science and Technology*, 33(1), 9-14.
- Miller, N. F., Spengler, R. N., & Frachetti, M. (2016). Millet cultivation across Eurasia: Origins, spread, and the influence of seasonal climate. *The Holocene*, 26(10), 1566-1575.
- Mitharwal, S., Kumar, S., & Chauhan, K. (2021). Nutritional, polyphenolic composition and in vitro digestibility of finger millet (*Eleusine coracana* L.) with its potential food applications: A review. *Food Bioscience*, 44, 101382
- Nair U. K., A., Hema, V., Sinija, V. R., & Hariharan, S. (2020). Millet milk: A comparative study on the changes in nutritional quality of dairy and nondairy milks during processing and malting. *Journal of Food Process Engineering*, 43(3), 1–7.
- Nithiyantham, S., Kalaiselvi, P., Mahomoodally, M. F., Zengin, G., Abirami, A., & Srinivasan, G. (2019). Nutritional and functional roles of millets—A review. *Journal of food biochemistry*, 43(7), e12859.

Omola, E. M., Kawo, A. H., & Shamsudden, U. (2014). Physico-chemical, sensory and microbiological qualities of yoghurt brands sold in Kano metropolis, Nigeria. *Bayero Journal of pure and applied sciences*, 7(2), 26-30.

Parveen S, Hafiz F. Fermented cereal from indigenous raw materials. *Pakistan Journal of Nutrition*.2003; 2(5):289-291.

Pragya, S., & Rita, S. R. (2012). Finger millet for food and nutritional security. *African Journal of Food Science*, 6(4), 77-84.

Priyanka, K., & Hasan, S. A. A. (2012). Preparation and evaluation of montelukast sodium loaded solid lipid nanoparticles. *Journal of Young Pharmacists*, 4(3), 129-137.

Raajeswari, P. A., & Nithya, M. (2018). Standardization of Value-Added Recipes with Millet Milk Powder. *Research & Reviews: Journal of Food Science and Technology*, 5(2), 10–17.

Ramandeep, K., Nahid, A., Neelabh, C., & Navneet, K. (2017). Phytochemical screening of *Phyllanthus niruri* collected from Kerala region and its antioxidant and antimicrobial potentials. *Journal of Pharmaceutical Sciences and Research*, 9(8), 1312.

Rasic, J. L., & Kurmann, J. A. (1978). Yoghurt. Scientific grounds, technology, manufacture and preparations. *Yoghurt. Scientific grounds, technology, manufacture and preparations*

Rul, F. (2017). Yogurt: Microbiology, organoleptic properties and probiotic potential.

Sambucetti, M. E., & Zuleta, A. (1996). Resistant starch in dietary fiber values measured by the AOAC method in different cereals. *Cereal Chemistry*, 73(6), 759-761.

Sanchez-Vega, M. M. (2013). *Influence of various health beneficial spices on some characteristics of yogurt culture bacteria Lactobacillus acidophilus, and sensor acceptability of spicy probiotic yogurt*. Louisiana State University and Agricultural &

Mechanical College.

Sanders, M. E., & Klaenhammer, T. R. (2001). Invited review: the scientific basis of *Lactobacillus acidophilus* NCFM functionality as a probiotic. *Journal of dairy science*, *84*(2), 319-331.

Saranya, S., Sudheer, K. P., Ranasalva, N., & Nithya, C. (2016). Effect of process parameters on physical properties of spray dried banana pseudostem juice powder. *Advances in Life Sci*, *5*(17), 6768-6773.

Sehgal, S., Kawatra, A., & Singh, G. (2003). Recent technologies in pearl millet and sorghum processing and food product development. *Alternative Uses of Sorghum and Pearl Millet in Asia*, *60*.

Serra, M., Trujillo, A. J., Guamis, B., & Ferragut, V. (2009). Flavour profiles and survival of starter cultures of yoghurt produced from high-pressure homogenized milk. *International Dairy Journal*, *19*(2), 100-106.

Shobana, S., Krishnaswamy, K., Sudha, V., Malleshi, N. G., Anjana, R. M., Palaniappan, L., & Mohan, V. (2013). Finger millet (*Ragi*, *Eleusine coracana* L.): a review of its nutritional properties, processing, and plausible health benefits. *Advances in food and nutrition research*, *69*, 1-39.

Shobana, S., Sreerama, Y. N., & Malleshi, N. G. (2009). Composition and enzyme inhibitory properties of finger millet (*Eleusine coracana* L.) seed coat phenolics: Mode of inhibition of α -glucosidase and pancreatic amylase. *Food chemistry*, *115*(4), 1268-1273.

Singh, P., & Raghuvanshi, R. S. (2012). Finger millet for food and nutritional security. *African Journal of Food Science*, *6*(4), 77-84.

Singh, A. K. (2023). Early presence/introduction of African and East Asian millets in India: integral to traditional agriculture. *The Nucleus*, *66*(3), 261-271.

Stone, H., Bleibaum, R. N., & Thomas, H. A. (2020). *Sensory evaluation practices*. Academic press

Tamime, A. Y., & Robinson, R. K. (2007). *Tamime and Robinson's yoghurt: science and technology*. Elsevier.

Tyl, C., Marti, A., Hayek, J., Anderson, J., & Ismail, B. P. (2018). Effect of growing location and variety on nutritional and functional properties of proso millet (*Panicum miliaceum*) grown as a double crop. *Cereal Chemistry*, 95(2), 288-301

Udeh, H. O., Duodu, K. G., & Jideani, A. I. O. (2017). Finger Millet Bioactive Compounds, Bioaccessibility, and Potential Health Effects--a Review. *Czech Journal of Food Sciences*, 35(1).

Vanderhoof, J., & Mitmesser, S. (2010). Probiotics in the management of children with allergy and other disorders of intestinal inflammation. *Beneficial Microbes*, 1(4), 351-356.

Vilakati, N., Taylor, J. R., MacIntyre, U., & Kruger, J. (2016). Effects of processing and addition of a cowpea leaf relish on the iron and zinc nutritive value of a ready-to-eat sorghum-cowpea porridge aimed at young children. *LWT*, 73, 467-472.

Weerathilake, W. A. D. V., Rasika, D. M. D., Ruwanmali, J. K. U., & Munasinghe, M. A. D. D. (2014). The evolution, processing, varieties and health benefits of yogurt. *International Journal of Scientific and Research Publications*, 4(4), 1-10

APPENDIX

SENSORY SCORE CARD FOR MILLET FORTIFIED YOGHURT

Date:

Name of panelist:

You are requested to assess the product in terms of general acceptability on a 9-point hedonic scale.

Score System

Like extremely - 9

Like very much - 8

Like moderately - 7

Like slightly - 6

Neither like nor dislike - 5

Dislike slightly - 4

Dislike moderately - 3

Dislike very much - 2

Dislike extremely - 1

Characteristics	Sample code								
	A	B	C	D	E	F	G	H	I
Appearance									
Odour									
Flavour / taste									
Body and texture									
Mouth feel									
Overall acceptability									

Signature:

DEVELOPMENT AND EVALUATION OF MILLET-BASED YOGHURT

By

ASWIN P K (2020-06-001)

FATHIMA SHIMNA (2020-06-002)

NEEHARIKA (2020-06-011)

NAYANTHARA B (2020-06-013)

ABSTRACT

Submitted in partial fulfilment of the requirement of

BACHELOR OF TECHNOLOGY

IN

FOOD TECHNOLOGY

Faculty of Agricultural Engineering and Technology

Kerala Agricultural University



DEPARTMENT OF PROCESSING AND FOOD ENGINEERING

KELAPPAJI COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY

TAVANUR -679573 MALAPPURAM

KERALA, INDIA

2023-2024

ABSTRACT

Yoghurt is a cultured milk product obtained by lactic acid fermentation through the action of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* and resulting in reduction of pH with coagulation. Minor millets are cited as important addendum for major cereals. Millets are nutritionally superior to major cereal crops with respect of energy value, proteins, fat and minerals. Blending of millet grains or their milling fractions combined with other treatments is one of the most convenient techniques to produce food products with high nutritional value and functionality and to promote their utilization in a large range of food products. Hence the present study was done to formulate millet-based yoghurt from finger and proso millet milk. Millet concentrations at 10%, 20% and 30%, respectively were taken as independent variables. Optimization of process variables was done based on sensory evaluation of the developed yoghurt samples. Sensory evaluation revealed that yoghurt prepared from 10% proso millet milk extract and 20% finger millet milk extract secured high scores in organoleptic evaluation. Physicochemical analysis of the optimized yoghurt samples was conducted using the standard procedures. From the microbial analysis, it was understood that millet-based yoghurt could be safely stored for a period of 14 days under refrigerated storage.

