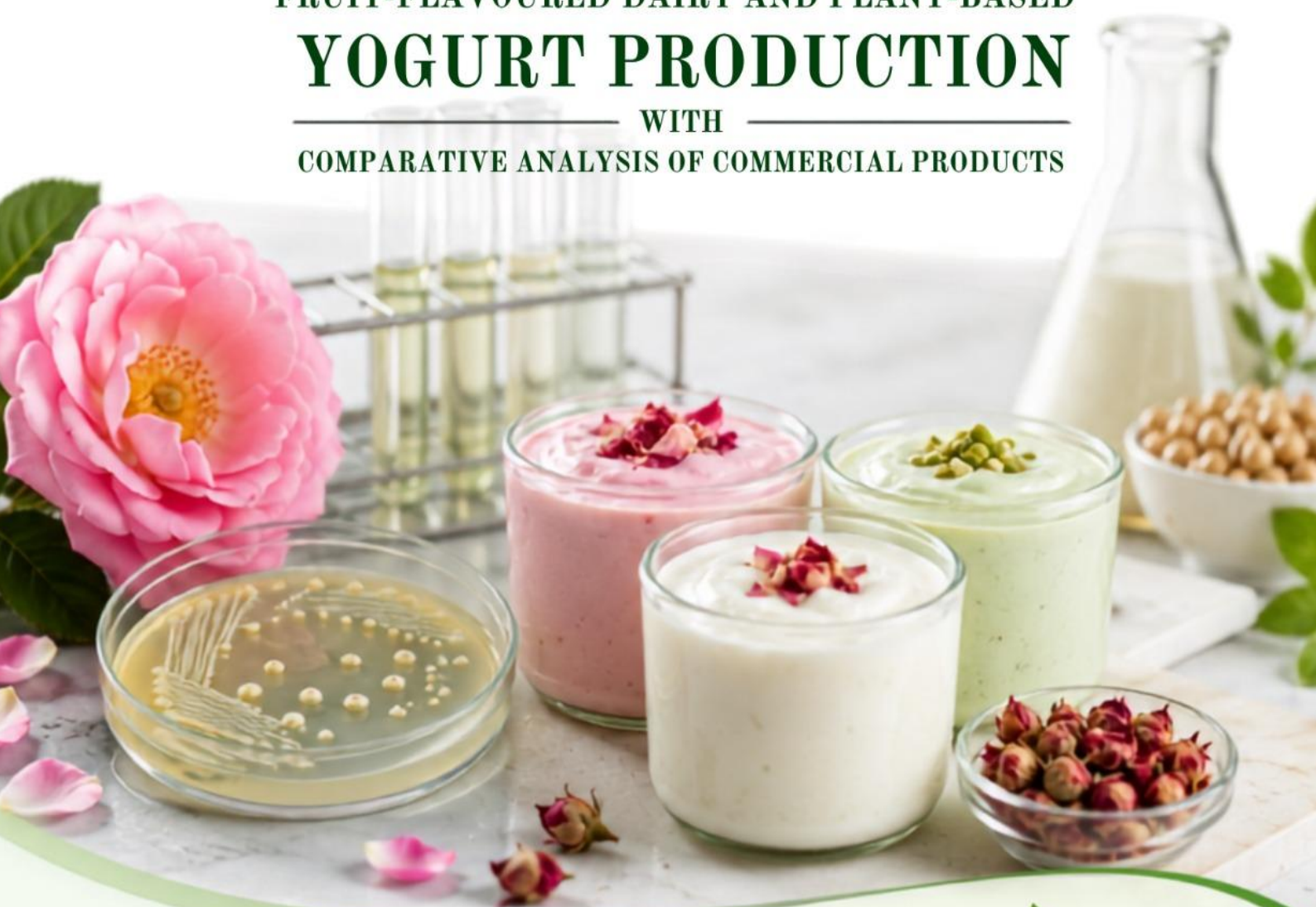


ISOLATION AND CHARACTERIZATION OF  
**MICROBIAL FLORA**  
FROM  
**ROSA INDICA**

AND EVALUATION OF ITS POTENTIAL ROLE IN  
FRUIT-FLAVOURED DAIRY AND PLANT-BASED  
**YOGURT PRODUCTION**  
WITH  
COMPARATIVE ANALYSIS OF COMMERCIAL PRODUCTS



Isolation



Characterization



Yogurt  
Production



Comparative  
Analysis



Exploring Natural  
Microbial Potential for  
Healthy & Sustainable  
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# ISOLATION AND CHARACTERIZATION OF MICROBIAL FLORA FROM *ROSA INDICA* AND EVALUATION OF ITS POTENTIAL ROLE IN FRUIT-FLAVOURED DAIRY AND PLANT-BASED YOGURT PRODUCTION WITH COMPARATIVE ANALYSIS OF COMMERCIAL PRODUCTS

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## ABSTRACT

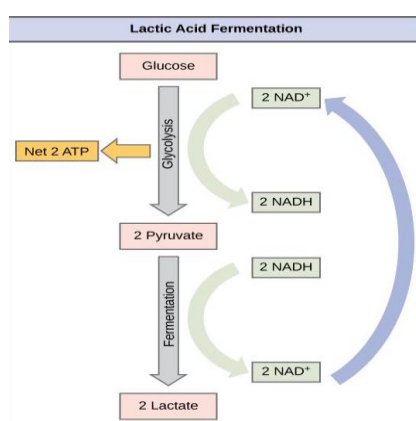
The present study focuses on the isolation, identification, and characterization of microbial flora from *Rosa indica* flowers and their application in the preparation of functional yogurt products. The isolated microorganisms were evaluated for their fermentation potential in two yogurt systems: fruit-flavored dairy yogurt (pasteurized milk with mango and apple) and plant-based soy yogurt. Biochemical analyses were conducted to determine the nutritional and functional properties of the developed products. Furthermore, a comparative assessment with commercially available yogurts was performed. The results demonstrated that microbial cultures from *Rosa indica* possess promising fermentation abilities and contribute to enhanced bioactive properties in yogurt formulations.

## Keywords

*Rosa indica*, Microbial isolation, Plant-based yogurt, Soy yogurt, Fruit-flavored yogurt, Phytochemical analysis, Functional food

## 1.0 INTRODUCTION

Fermented foods are among the earliest forms of food processing, traditionally developed to extend shelf life by inhibiting spoilage microorganisms while simultaneously enhancing sensory properties such as flavour, aroma, and texture. Over time, fermentation has gained scientific importance due to its role in improving nutritional quality and producing bioactive compounds beneficial to human health. These foods are now widely recognized as functional foods with global dietary relevance [1]. Among them, yogurt remains one of the most popular fermented products due to its nutritional composition and probiotic potential [2].



**Figure 1. Schematic representation of lactic acid fermentation in food systems.**

Source: Adapted from Handbook of Indigenous Fermented Foods.[1]

Milk and dairy products serve as important dietary sources of high-quality protein, energy, and essential minerals, including calcium, magnesium, potassium, zinc, and phosphorus, which exhibit high bioavailability [3][4]. These nutrients are fundamental for growth, development, and the maintenance of physiological functions. Calcium plays a critical role in skeletal development, bone mineral density, and the prevention of osteoporosis. Adequate intake during early life is essential for achieving peak bone mass, while continued consumption in later stages helps reduce fracture risk [3]. Furthermore, dairy consumption has been associated with improved oral health and may contribute to the regulation of cholesterol levels, body weight, and blood pressure, although the long-term health implications of milk fat remain inconclusive [5].

Yogurt is a widely consumed fermented dairy product valued for its nutritional, sensory, and functional attributes. It is typically characterized by a smooth, semi-solid gel structure, mild acidity, and a desirable flavour profile. Consumer acceptance is strongly influenced by sensory parameters such as taste, texture, and appearance. The development of yogurt flavour is primarily attributed to the metabolic activity of lactic acid bacteria, which produce a range of volatile compounds including organic acids, alcohols, aldehydes, and esters ([6][7]. -label products.

Two fruit-flavoured yoghurt formulations were developed in this study, one using dairy milk as the base and the other using plant-based soy milk as an alternative matrix.

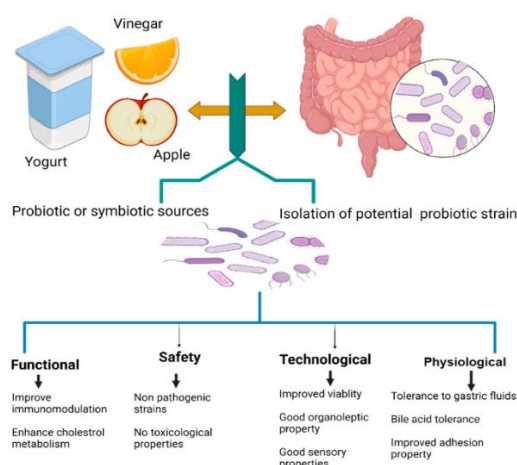
Fruits used in yogurt production are carefully selected to ensure desirable flavour, texture, and product stability. Suitable cultivars are chosen based on their compatibility with processing conditions and their ability to maintain quality during storage. Fruit quality is influenced by variety, cultivation practices, and environmental factors such as soil and climate [8].

Mango (*Mangifera indica*) is valued for its flavour and bioactive compounds. Its peel, about 20% of the fruit, is rich in polyphenols, carotenoids, and vitamins with antioxidant and antimicrobial properties, making it a potential functional ingredient [9][10].

Apple and its derivatives, including pulp, powder, and pomace, are widely used in yogurt [11]. Apple mainly affects physicochemical properties and improves texture by enhancing gel structure and firmness, particularly when used at optimal levels [12]. However, limited studies exist on the effects of apple pulp on yogurt texture and aroma [13].

Increasing consumer demand for lactose-free and plant-based alternatives has led to the development of non-dairy yogurt products. Soy-based yogurt, in particular, has gained considerable attention due to its high protein content and nutritional value. Produced through the fermentation of soy milk using lactic acid bacteria, it offers probiotic benefits comparable to conventional yogurt and is suitable for lactose-intolerant and vegan populations [14]. The incorporation of fruit components such as pulp, juice, or extracts further enhances yogurt by improving both sensory quality and nutritional value. Fruits such as mango and apple are rich in vitamins, phenolic compounds, and antioxidants, which contribute to improved functional and sensory characteristics [10][11].

Probiotics are defined as live microorganisms that confer health benefits when administered in adequate amounts, primarily by improving the balance of intestinal microbiota. Dairy products, particularly yogurt, are considered effective carriers for probiotics due to their ability to support microbial viability during storage and gastrointestinal transit [14][15]. Common probiotic strains include species of *Lactobacillus* and *Bifidobacterium*, which contribute to improved gut health, enhanced immune function, and protection against pathogenic microorganisms [16]. The efficacy of probiotic products depends on several factors, including microbial strain selection, product formulation, processing conditions, and storage environment [17].



**Figure 2. Schematic representation of probiotic sources, isolation of beneficial microbial strains, and their functional, safety, technological, and physiological roles in gut health.**

Source: Adapted from the consensus guidelines on the definition and appropriate use of probiotics established by the International Scientific Association for Probiotics and Prebiotics [32].

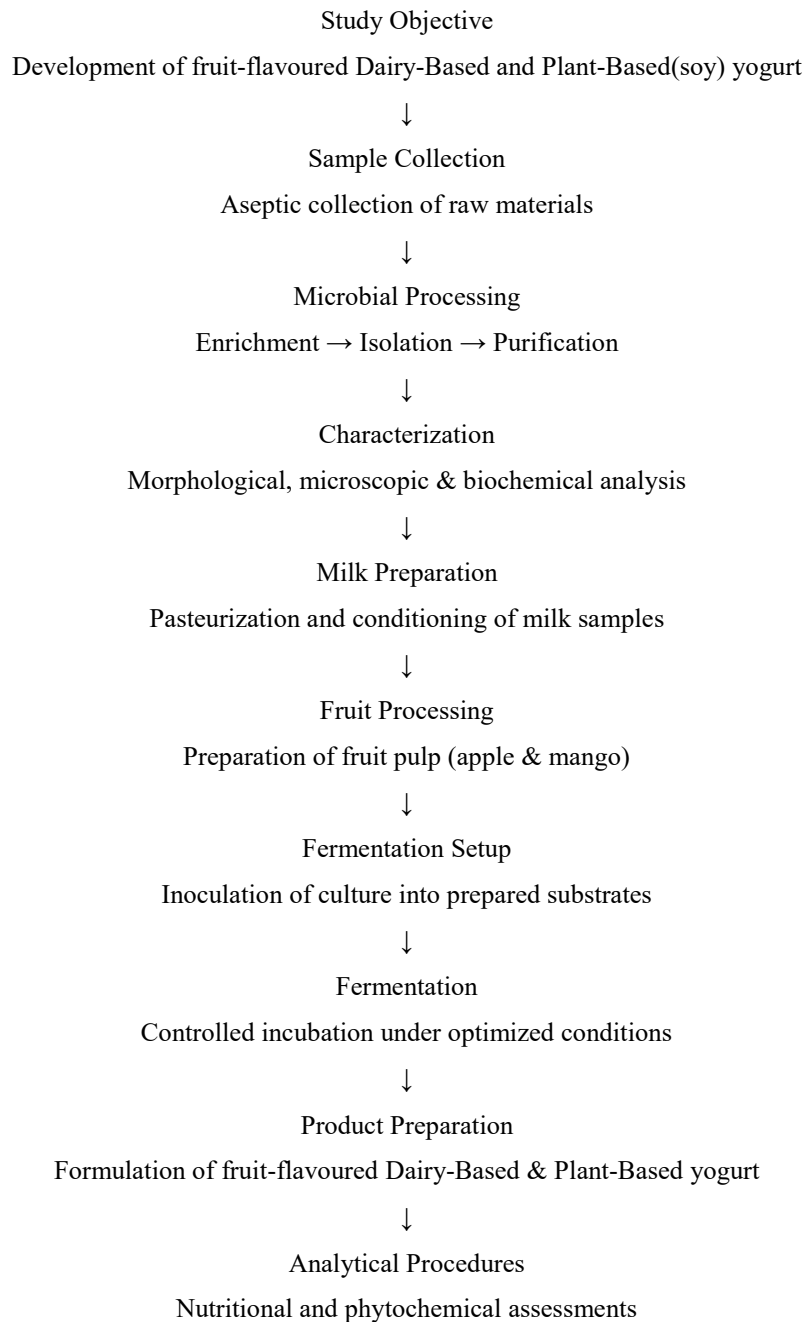
Lactose intolerance is a metabolic condition characterized by reduced activity of the lactase enzyme, resulting in the inability to properly digest lactose. The fermentation of undigested lactose in the colon can lead to gastrointestinal discomfort. Fermented dairy products such as yogurt typically contain reduced lactose levels and are therefore more easily tolerated. Additionally, plant-based alternatives such as soy yogurt provide suitable options for individuals with lactose intolerance while maintaining adequate nutritional intake [18][19].

Floral surfaces, including rose petals, serve as natural reservoirs for diverse microbial populations, including species of *Bacillus*. These environments provide nutrient-rich conditions conducive to microbial growth, making them suitable sources for isolating beneficial strains [20]. *Bacillus subtilis* is particularly noteworthy for its ability to produce extracellular enzymes and antimicrobial compounds, which contribute to food safety and quality [21]. Although it is not suitable as a primary starter culture in yogurt production, it can function as an adjunct culture alongside lactic acid bacteria to enhance fermentation efficiency and improve product. Its enzymatic activity facilitates protein hydrolysis, improving digestibility and flavour development. The probiotic potential of *Bacillus subtilis* is further supported by its ability to form heat-resistant spores, enabling survival during processing and passage through the gastrointestinal tract [22].

Therefore, the present study focuses on the development of fruit-flavoured dairy yogurt and plant-based yogurt (soy) using mango and apple, along with the isolation and application of beneficial bacterial strains from *Rosa indica*. This approach aims to enhance the nutritional, functional, and sensory properties of yogurt while exploring novel microbial sources for probiotic applications.

## 2. MATERIALS AND METHODS

### 2.1. Overview of Experimental Design



**Figure 3: Schematic representation of the experimental methodology for preparation of fruit-flavoured yogurt.**

**Table 1: Materials Used in the Study**

| Category      | Items                                                                                                                               |
|---------------|-------------------------------------------------------------------------------------------------------------------------------------|
| Raw materials | Rose petals ( <i>Rosa</i> spp.), raw milk, apple, mango, soybean, <i>Bacillus</i> spp                                               |
| Culture Media | Nutrient agar, starch agar, milk agar, MR-VP broth, urea broth, nitrate broth                                                       |
| Reagents      | Kovac's reagent, iodine, methyl red, VP reagents, DNS reagent, Folin–Ciocalteu reagent, Gram stain kit, BSA, phytochemical reagents |
| Equipment     | Autoclave, incubator, laminar airflow, microscope, spectrophotometer, pH meter, analytical balance                                  |

## 2.2. Isolation of Bacteria

Serial dilution ( $10^{-1}$ – $10^{-6}$ ), spread plate, and streak plate techniques were used for the isolation and purification of bacterial cultures. Incubation was performed at 37°C for 24 hours [23].



Figure 4.1



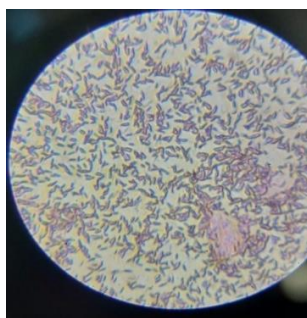
Figure 4.2

4.1. Enrichment of microorganisms from *Rosa indica* petals using dip method

4.2. Isolation of pure colonies using streak plate method

## 2.3. Gram Staining

The isolate was identified as Gram-positive, rod-shaped *Bacillus* spp., indicating a thick peptidoglycan cell wall structure [24].



**Figure 5: Identification of an isolate by using Gram Staining**

## 2.4. Biochemical Characterization

**Table 2: Biochemical Tests of Isolated Bacteria**

| Test                | Principle                                  | Positive Result                  |
|---------------------|--------------------------------------------|----------------------------------|
| Catalase test       | Breakdown of H <sub>2</sub> O <sub>2</sub> | Bubble formation                 |
| Indole test         | Tryptophan degradation                     | Red ring                         |
| MR test             | Mixed acid fermentation                    | Red colour                       |
| VP test             | acetone production                         | Pink/red color                   |
| Stach hydrolysis    | Amylase activity                           | Clear zone                       |
| Citrate utilization | citrate as carbon source                   | Blue color                       |
| Carbohydrate test   | Fermentation of sugars producing acid/gas  | Yellow colour (acid), gas bubble |
| Casein              | Breakdown of casein protein by protease    | Clear zone around colony         |

Source: Adapted and modified from Microbiology laboratory manual. Pearson Education. [23].

## 2.5. Milk Preparation and Biochemical Analysis

Raw milk (dairy & soy milk) was pasteurised by controlled heating and cooling cycles to eliminate unwanted microorganisms while preserving nutritional quality [25].

Protein content was determined using the Lowry method, based on the formation of a copper-protein complex and reduction of Folin–Ciocalteu reagent, measured at 660 nm [26].

Carbohydrate content was estimated using the DNS method, which quantifies reducing sugars by measuring absorbance at 540 nm [27].

## 2.6. Preparation of Fruit Pulp and Phytochemical Screening

Fresh apple and mango were processed aseptically to obtain pulp. Phytochemical screening of apple pulp was carried out using standard qualitative methods to detect alkaloids, tannins, flavonoids, phenols, terpenoids, steroids, and saponins based on characteristic color reactions [28][29].

**Table 3: Phytochemical Screening of Sample**

| Compounds  | Test used                   | principle                                                                     | Observations          |
|------------|-----------------------------|-------------------------------------------------------------------------------|-----------------------|
| Alkaloids  | Mayer's/Wagner's            | Alkaloids react with reagents to form insoluble complexes                     | Precipitate formation |
| Tannins    | Ferric chloride             | Tannins form complexes with Fe <sup>3+</sup> ions producing colored compounds | Blue/green color      |
| Flavonoids | Alkaline/ lead acetate test | Flavonoids form colored complexes in alkaline conditions or with metal ions   | Yellow color          |
| Steroids   | Salkowski test              | Steroids react with sulfuric acid forming colored complexes                   | Green/blue ring       |
| Terpenoids | Salkowski test              | Terpenoids react with chloroform and acid to form characteristic layers       | Reddish-brown layer   |
| Saponins   | Froth test                  | Saponins reduce surface tension leading to foam formation                     | Stable foam           |
| phenols    | Ferric chloride             | Formation of colored complexes with iron ions                                 | Blue/green color      |

Source: Adapted and modified from Pharmacognosy [29].

## 2.7. Yogurt Preparation and Fermentation

Pasteurized dairy and plant-based milk were inoculated with the isolated bacterial culture and supplemented with fruit pulp. Fermentation was carried out at approximately 37°C under aseptic conditions. During fermentation, microbial metabolism led to acid production, resulting in coagulation of milk proteins in dairy samples and development of yogurt-like consistency in plant-based samples ([30][31].

## 2.8. Nutritional and Phytochemical Analysis of Yogurt

The protein and carbohydrate content of the yoghurt samples were estimated using the Lowry and DNS methods, respectively [26][27].

## 3. RESULTS

**Table 4: Biochemical Characterization**

| Sr. no | Biochemical tests | Result |
|--------|-------------------|--------|
| 1.     | Citrate           | -      |
| 5.     | MR                | -      |
| 6.     | MR-VP             | -      |
| 7.     | carbohydrate      | +      |
| 8.     | Catalase          | +      |
| 9.     | Indole            | +      |
| 10.    | Casien            | +      |
| 11.    | Amylase           | +      |

These findings indicate high metabolic versatility typical of *Bacillus* species

### 3.1 Preparation of Milk Substrates

Both dairy and soy milk were successfully pasteurized, showing no contamination post-treatment



Figure 6.1: Pasteurization of milk

Figure 6.2: pasteurized vegan milk

### 3.2. Biochemical Analysis of Milk

Protein and carbohydrate estimations confirmed the presence of macronutrients in both milk types. Optical density (OD) increased with concentration, indicating reliable detection

Table 5: Protein OD values (dairy and soy milk)

| Dairy milk |          | Soy milk   |         |
|------------|----------|------------|---------|
| Absorbance | Readings | Absorbance | Reading |
| Blank      | 0        | Blank      |         |
| 30         | 0.206    | 30         | 0.261   |
| 60         | 0.732    | 60         | 0.772   |
| 120        | 1.047    | 120        | 1.538   |
| 240        | 1.850    | 240        | 1.820   |

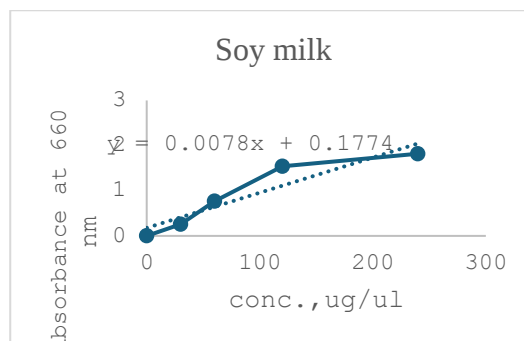
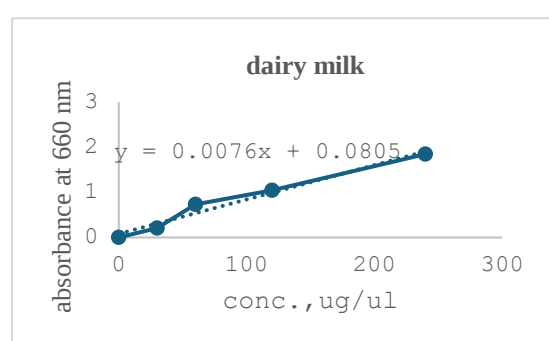


Figure 7: Protein standard curve

The protein conc. in dairy milk is 260.80ug/ul

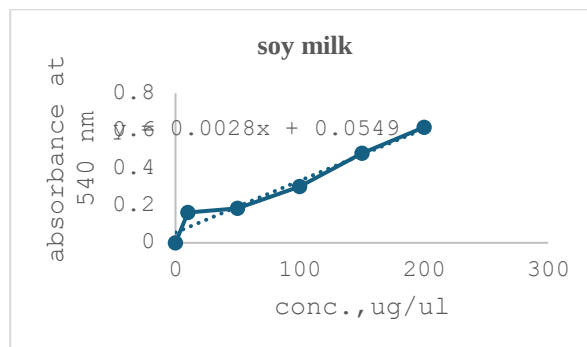
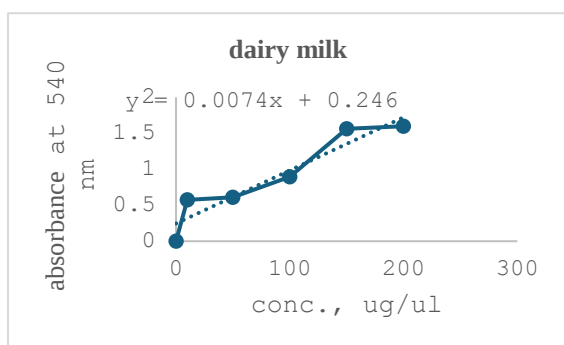
The protein conc. in soy milk is 307.51ug/ul

Table 6: Carbohydrate OD values (Dairy and soy milk)

| Dairy milk |         | Soy milk   |         |
|------------|---------|------------|---------|
| Absorbance | Reading | Absorbance | Reading |
| Blank      |         |            |         |
| 10         | 0.573   |            |         |
| 50         | 0.612   |            |         |

|     |       |
|-----|-------|
| 100 | 0.894 |
| 150 | 1.561 |
| 200 | 1.597 |

| Absorbance | Reading |
|------------|---------|
| Blank      |         |
| 10         | 0.162   |
| 50         | 0.185   |
| 100        | 0.300   |
| 150        | 0.478   |
| 200        | 0.618   |



**Figure 8: Carbohydrate standard curve**

The carb. conc. in dairy milk is 245.54ug/ul

The carb. conc. in soy milk is 518.96ug/ul

Soy milk showed strong protein reactivity due to plant storage proteins, while carbohydrates were attributed to oligosaccharides.

### 3.3. Fruit Sample Preparation and Phytochemical Analysis

Apple (*Malus domestica*) and mango (*Mangifera indica*) were processed into extracts under sterile conditions.

Phytochemical screening of apple pulp revealed the presence of flavonoids, phenolics, tannins, saponins, and terpenoids, indicating antioxidant potential (Pandey & Rizvi, 2009).

**Table 7: Phytochemical Analysis of Apple Pulp**

| Sr.no. | Phytochemical test | Result |
|--------|--------------------|--------|
| 1.     | Alkaloids          | (-)    |
| 2      | Tannis             | (+)    |
| 3      | Flavonoids         | (+)    |

|   |            |     |
|---|------------|-----|
| 4 | Steroids   | (+) |
| 5 | Terpenoids | (+) |
| 6 | Saponins   | (+) |
| 7 | Phenolics  | (+) |

### 3.4. Yogurt Production

Fermentation using the isolated *Bacillus* strain resulted in yogurt formation in both dairy and soy milk within 24 hours



**Figure 9.1: Dairy Yogurt formation after 24 hours of fermentation**

**Figure.9.2: Plant-based yogurt**

The addition of fruit extracts enhanced sensory attributes such as color and aroma. Mango-based yogurt showed greater visual and aromatic intensity compared to apple variants.

### 3.5. Biochemical Analysis of Yogurt

Protein and carbohydrate contents were quantified to evaluate nutritional quality.

Protein levels were slightly higher in mango yogurt samples.

**Table 8: Protein values of Dairy and soya yogurt**

| Absorbance | Readings |
|------------|----------|
|------------|----------|

|       |       |
|-------|-------|
| Blank | 0     |
| 30    | 0.823 |
| 60    | 0.917 |
| 120   | 1.067 |
| 240   | 1.176 |

**Apple sample (Dairy)**

| Absorbance | Readings |
|------------|----------|
| Blank      | 0        |
| 30         | 0.723    |
| 60         | 0.918    |
| 120        | 1.076    |
| 240        | 1.196    |

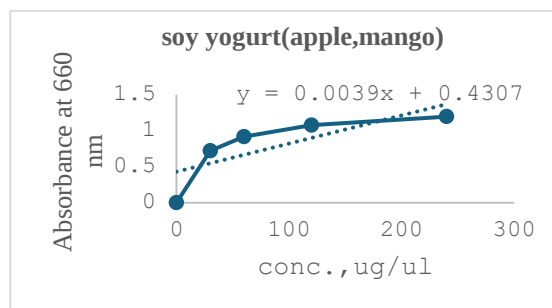
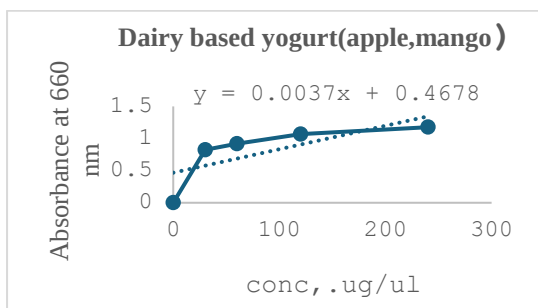
**Mango sample (dairy)**

| Absorbance | Readings |
|------------|----------|
| Blank      | 0        |
| 30         | 0.723    |
| 60         | 0.918    |
| 120        | 1.076    |
| 240        | 1.196    |

**Apple (soy yogurt)**

| Absorbance | Readings |
|------------|----------|
| Blank      | 0        |
| 30         | 0.823    |
| 60         | 0.917    |
| 120        | 1.067    |
| 240        | 1.176    |

**Mango (soy yogurt)**



**Figure 10: Protein standard curve**

The protein conc. in Apple is 27.22  $\mu\text{g/ul}$

The protein conc. in Apple is 288.0  $\mu\text{g/ul}$

The protein conc. in Mango is 33.92  $\mu\text{g/ul}$

The protein conc. in Mango is 1.492  $\mu\text{g/ul}$

Carbohydrates were detected in both samples, confirming sugar availability after fermentation.

**Table 9: Carbohydrate OD values dairy and plant-based yogurt**

| Absorbance | Reading |
|------------|---------|
| Blank      |         |
| 10         | 0.305   |

|     |       |
|-----|-------|
| 50  | 0.487 |
| 100 | 0.571 |
| 150 | 0.625 |
| 200 | 0.689 |

| Absorbance | Reading |
|------------|---------|
| Blank      |         |
| 10         | 0.305   |
| 50         | 0.487   |
| 100        | 0.571   |
| 150        | 0.625   |
| 200        | 0.689   |

Apple sample (dairy based yogurt)

Mango sample (dairy based yogurt )

| Absorbance | Reading |
|------------|---------|
| Blank      |         |
| 10         | 0.305   |
| 50         | 0.487   |
| 100        | 0.571   |
| 150        | 0.625   |
| 200        | 0.689   |

| Absorbance | Reading |
|------------|---------|
| Blank      |         |
| 10         | 0.305   |
| 50         | 0.487   |
| 100        | 0.571   |
| 150        | 0.625   |
| 200        | 0.689   |

Apple sample (soy yogurt)

Mango sample (soy yogurt)

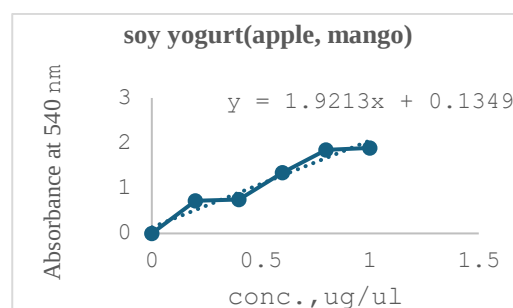
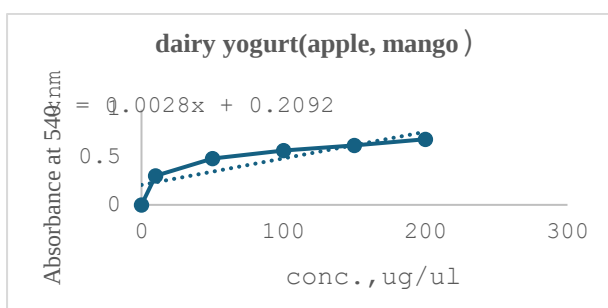


Figure 11: Carbohydrate standard curve

- The carb conc. of apple is  $180.28\mu\text{g}/\text{ul}$
- The carb conc. of mango is  $178.85\mu\text{g}/\text{ul}$

The carb conc. of apple is  $165.95\mu\text{g}/\text{ul}$

The carb conc. of apple is  $194.84\mu\text{g}/\text{ul}$

### 3.6. Direct comparison with commercial yogurt (g/100g)

Table 10: Comparison with Commercial Yogurt

| Sample (Fruit Type) | Protein (g/100 g) | Carbohydrate (g/100 g) | Comparison with Market (Fruit-Flavoured Yogurt) |
|---------------------|-------------------|------------------------|-------------------------------------------------|
|---------------------|-------------------|------------------------|-------------------------------------------------|

|                                  |             |              |                                                              |
|----------------------------------|-------------|--------------|--------------------------------------------------------------|
| <b>Dairy Apple Yogurt</b>        | <b>5.00</b> | <b>10.0</b>  | <b>Comparable to Amul &amp; Nestle</b>                       |
| <b>Dairy Mango yogurt</b>        | <b>0.62</b> | <b>9.9</b>   | <b>Much lower protein than all brands; carbs similar</b>     |
| <b>Vegan Apple yogurt</b>        | <b>5.30</b> | <b>9.2</b>   | <b>Slightly higher protein than Amul; carbs within range</b> |
| <b>Vegan Mango Yogurt</b>        | <b>7.04</b> | <b>10.8</b>  | <b>Higher protein; comparable to Epigamia</b>                |
| <b>Amul ( Strawberry/ Mango)</b> | <b>3-5</b>  | <b>10-15</b> | <b>Standard fruit yogurt</b>                                 |
| <b>Nestle( mixed fruit)</b>      | <b>3-6</b>  | <b>8-14</b>  | <b>Balanced fruit yogurt</b>                                 |

Comparison with fruit-flavoured commercial yogurts showed that the developed vegan variants exhibited higher protein content, while carbohydrate levels remained within the typical range influenced by fruit sugars.

#### 4. DISCUSSION

The present study explored the potential of microorganisms isolated from *Rosa indica* petals for application in yogurt fermentation, along with a comparative evaluation of dairy and plant-based formulations. The microbial isolate showed characteristics typical of *Bacillus* species, including positive enzymatic activities such as catalase

and starch hydrolysis. These traits suggest that the organism is metabolically active and capable of contributing to fermentation processes, which is consistent with earlier findings on *Bacillus* spp. used in food microbiology

During fermentation, noticeable differences were observed between dairy and plant-based yogurt systems. The dairy samples developed a firmer and more cohesive texture, which can be attributed to the coagulation of casein proteins under acidic conditions. This mechanism is well established in conventional yogurt production. In contrast, the plant-based (soy) yogurt exhibited a comparatively softer consistency, likely due to the absence of casein and differences in plant protein structure.

One of the most significant findings of this study was the variation in protein content between the samples. While dairy products are generally considered rich in protein due to casein and whey fractions, the experimental results revealed that the plant-based yogurt, particularly the mango-enriched variant, showed higher protein levels. This may be explained by the presence of concentrated storage proteins in soybeans and possible structural modifications during fermentation that improve protein availability. These findings indicate that plant-based yogurt can serve as a nutritionally competitive, and in some cases superior, alternative to dairy products.

Carbohydrate estimation confirmed the presence of sufficient fermentable sugars in both dairy and plant-based samples, which supported microbial growth and acid production during fermentation. The carbohydrate values obtained were comparable to those reported in commercial yogurt products, suggesting that the developed formulations meet general nutritional expectations.

The incorporation of fruit pulp, specifically mango and apple, contributed to improvements in sensory attributes such as flavour, colour, and texture. In addition, phytochemical analysis of the fruit pulp revealed the presence of bioactive compounds such as flavonoids and phenolic constituents. These compounds are naturally occurring in fruits and are known for their antioxidant. Their presence highlights the potential of fruit-enriched yogurt formulations to provide added functional benefits, although the analysis was limited to the fruit pulp used in the preparation.

When compared with commercially available fruit yogurts, the prepared samples demonstrated comparable carbohydrate content and, in some cases, higher protein levels, particularly in plant-based variants. This suggests that the use of naturally isolated microorganisms combined with fruit enrichment can result in products that are both nutritionally valuable and functionally relevant.

Overall, the study highlights the importance of exploring non-traditional microbial sources and plant-based substrates in the development of innovative fermented foods. Such approaches are especially relevant in the context of increasing demand for dairy alternatives and health-oriented food products.

## 5. Conclusion

The present study demonstrates that microorganisms isolated from *Rosa indica* petals can be effectively utilized in yogurt fermentation. The bacterial isolate exhibited suitable biochemical characteristics, supporting its potential application in fermented food production.

The developed yogurt samples showed satisfactory nutritional composition, with plant-based (soy) yogurt exhibiting higher protein content compared to certain dairy samples. This finding emphasizes the potential of plant-based yogurt as a strong alternative protein source and supports its increasing importance in modern dietary practices.

Carbohydrate levels in all samples were within acceptable ranges, confirming effective fermentation. The addition of fruit pulp, such as mango and apple, enhanced the sensory qualities of the yogurt and contributed natural bioactive compounds. Phytochemical analysis confirmed the presence of such compounds in the fruit pulp, indicating their potential contribution to the overall functional value of the final product.

Comparison with commercially available yogurt products showed that the developed formulations were nutritionally comparable, particularly in terms of carbohydrate content, and in some cases superior in protein content.

In conclusion, the combination of floral-derived microorganisms, plant-based substrates, and fruit enrichment represents a promising strategy for the development of innovative, nutritious, and functional yogurt products. Further research focusing on optimization, sensory evaluation, and shelf-life studies will support their potential for large-scale application.

## References

1. Steinkraus, K. H. (1994). Nutritional significance of fermented foods. *Food Research International*, 27(3), 259–267. [https://doi.org/10.1016/0963-9969\(94\)90165-8](https://doi.org/10.1016/0963-9969(94)90165-8)
2. Hesseltn, C. W. (1980). Fermented foods. In A. H. Rose (Ed.), *Economic microbiology* (Vol. 5, pp. 1–16). Academic Press.
3. Rizzoli, R. (2014). Dairy products, yogurts, and bone health. *American Journal of Clinical Nutrition*, 99(5), 1256S–1262S. <https://doi.org/10.3945/ajcn.113.073249>
4. Bailey, R. L. (2010). Dairy intake and bone health. *American Journal of Clinical Nutrition*, 91(5), 1317S–1322S. <https://doi.org/10.3945/ajcn.2010.28674F>
5. Bomjour, J. P. (2005). Dietary protein intake and bone health. *Journal of the American College of Nutrition*, 24(6), 526S–536S. (No DOI found in record provided)
6. Chen, H. (2010). Volatile flavor compounds in yogurt. *Food Chemistry*, 122(2), 489–496. <https://doi.org/10.1016/j.foodchem.2010.02.068>
7. Imhof, R., & Bosset, J. O. (1994). Relationships between micro-organisms and formation of aroma compounds in fermented dairy products. *Zeitschrift für Lebensmittel-Untersuchung und Forschung*, 198(3), 267–276. <https://doi.org/10.1007/BF01195355>
8. Chandan, R. C. (1993). Manufacturing yogurt and fermented milks. *Dairy Science and Technology*.
9. Palafox-Carlos, H., et al. (2012). Effect of mango (*Mangifera indica* L.) by-products on functional foods. *Food Research International*, 46(2), 573–580. <https://doi.org/10.1016/j.foodres.2011.11.013>
10. Jahurul, M. H. A., & Mohd Omar, A. K. (2015). Mango (*Mangifera indica* L.) by-products and their valuable components. *Food Chemistry*, 183, 173–180. <https://doi.org/10.1016/j.foodchem.2015.03.063>
11. Jovanovic, A. A., & Gorjanovic, S. Z. (2020). Apple polyphenols and their health benefits. *Journal of Food Composition and Analysis*, 88, 103430. <https://doi.org/10.1016/j.jfca.2020.103430>
12. Wang, X., et al. (2019). Effect of apple components on yogurt texture. *Journal of Food Processing and Preservation*, 43(5), e13945. <https://doi.org/10.1111/jfpp.13945>
13. Feng, C., & Cao, Y. (2019). Influence of fruit pulp on yogurt quality characteristics. *Food Science and Technology*. (Journal article—DOI not provided; please supply full citation or I can look it up)
14. Deshwal, G. K., & Kadyan, S. (2021). Probiotic potential of fermented dairy products. *Journal of Food Science and Technology*, 58(3), 789–798. <https://doi.org/10.1007/s13197-020-04629-6>
15. Fenster, K., et al. (2019). The production and delivery of probiotics: A review. *Food Science & Nutrition*, 7(3), 735–750. <https://doi.org/10.1002/fsn3.1125>

16. Soccol, C. R., & Pandey, A. (2010). Advances in probiotic research. *Biotechnology Advances*, 28(6), 929–939. <https://doi.org/10.1016/j.biotechadv.2010.07.006>
17. Meybodi, N. M., et al. (2020). Factors affecting probiotic viability. *LWT - Food Science and Technology*, 124, 109178. <https://doi.org/10.1016/j.lwt.2020.109178>
18. Harrington, L. K., & Mayberry, J. F. (2008). The effects of lactose intolerance on dairy consumption. *Nutrition Bulletin*, 33(1), 27–30. <https://doi.org/10.1111/j.1467-3010.2007.00696.x>
19. Ibrahim, S. A. (2013). Soy yogurt: Nutritional and health benefits. *Food Research International*, 54(1), 42–49. <https://doi.org/10.1016/j.foodres.2013.07.011>
20. Singh, R., et al. (2019). Microbial diversity of floral surfaces and their applications. *Journal of Microbial Research*, 174, 45–52.
21. Patel, A., et al. (2021). *Bacillus subtilis*: A versatile organism for food and agriculture. *Frontiers in Microbiology*, 12, 1234. <https://doi.org/10.3389/fmicb.2021.01234>
22. Kumar, M., et al. (2020). Probiotic properties of *Bacillus subtilis* and its applications. *LWT - Food Science and Technology*, 128, 109451. <https://doi.org/10.1016/j.lwt.2020.109451>
23. Cappuccino, J. G., & Welsh, C. (2017). *Microbiology laboratory manual*. Pearson Education.
24. Beveridge, T. J. (2001). Use of the Gram stain in microbiology. *Biotechnic & Histochemistry*, 76(3), 111–118. <https://doi.org/10.1080/10520290109354419>
25. Walstra, P., Wouters, J. T. M., & Geurts, T. J. (2006). *Dairy science and technology* (2nd ed.). CRC Press.
26. Lowry, O. H., Rosebrough, N. J., Farr, A. L., & Randall, R. J. (1951). Protein measurement with the Folin–Ciocalteu reagent. *Journal of Biological Chemistry*, 193(1), 265–275.
27. Miller, G. L. (1959). Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Analytical Chemistry*, 31(3), 426–428. <https://doi.org/10.1021/ac60147a030>
28. Harborne, J. B. (1998). *Phytochemical methods: A guide to modern techniques of plant analysis* (3rd ed.). Springer. <https://doi.org/10.1007/978-1-4615-1921-7>
29. Trease, G. E., & Evans, W. C. (2009). *Pharmacognosy* (16th ed.). Saunders Elsevier.
30. Tamime, A. Y., & Robinson, R. K. (2007). *Yogurt: Science and technology* (3rd ed.). Woodhead Publishing. <https://doi.org/10.1016/B978-1-84569-234-0.X5000-4>
31. Granato, D., Branco, G. F., Nazzaro, F., Cruz, A. G., & Faria, J. A. F. (2010). Functional foods and nondairy probiotic food development. *Comprehensive Reviews in Food Science and Food Safety*, 9(3), 292–302. <https://doi.org/10.1111/j.1541-4337.2010.00114.x>
32. Hill, C., Guarner, F., Reid, G., Gibson, G. R., Merenstein, D. J., Pot, B., Morelli, L., Canani, R. B., Flint, H. J., Salminen, S., Calder, P. C., & Sanders, M. E. (2014). Expert consensus document: The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. *Nature Reviews Gastroenterology & Hepatology*, 11(8), 506–514. <https://doi.org/10.1038/nrgastro.2014.66>

## ABOUT THE BOOK

This book explores the microbial diversity of *Rosa indica* and its promising application in the development of fruit-flavoured dairy and plant-based yogurts. From isolation and characterization to technological performance evaluation and comparison with commercial products, this study provides valuable insights into natural cultures with potential probiotic and functional benefits.



## KEY HIGHLIGHTS



### Isolation of Microbial Flora from *Rosa indica*

Systematic isolation and screening of bacteria and yeasts from *Rosa indica* flowers.



### Characterization of Isolates

Morphological, biochemical and molecular characterization of potential starter cultures.



### Yogurt Production

Development of fruit-flavoured dairy and plant-based yogurts using selected microbial cultures.



### Evaluation of Functional Properties

Assessment of probiotic potential, acid tolerance, antimicrobial activity and sensory attributes.



### Comparative Analysis with Commercial Products

Comparative study of physicochemical, microbial and sensory properties with leading commercial yogurts.



### Natural, Healthy & Sustainable

Promoting the use of natural microbial resources for healthier and sustainable food systems.

“ Harnessing Nature’s Microbial Diversity for Innovative, Healthy and Sustainable Yogurt Solutions ”



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A comprehensive resource for microbiologists, food technologists, nutritionists and anyone interested in natural starter cultures and functional fermented foods.

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