

**DETECTION OF ENTERIC PATHOGENS IN DIARY PRODUCTS SOLD IN BENIN-  
CITY, EDO STATE**

**BY**

**IMOUBE BRIGHT OSHUARE  
LSC2103951**

**DEPARTMENT OF MICROBIOLOGY,  
FACULTY OF LIFE SCIENCE,  
UNIVERSITY OF BENIN,  
BENIN CITY.**

**NOVEMBER 2025**

**DETECTION OF ENTERIC PATHOGENS IN DIARY PRODUCTS SOLD IN BENIN-  
CITY, EDO STATE**

**BY**

**IMOUBE BRIGHT OSHUARE  
LSC2103951**

**A PROJECT REPORT WRITTEN AND SUBMITTED TO THE DEPARTMENT OF  
MICROBIOLOGY, IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR  
THE AWARD OF BACHELOR OF SCIENCE (HONOURS) DEGREE (B.Sc) IN  
MICROBIOLOGY.**

**NOVEMBER 2025**

## **CERTIFICATION**

This is to certify that this project work was carried out by Bright Oshuare IMOUBE in the department of Microbiology, Faculty of Life Sciences, University of Benin-City under the supervision PROF. S.E. OMONIGHO.

---

PROF. S.E. OMONIGHO

---

Date

## **DEDICATION**

This project is dedicated to the Almighty GOD, the Author and Finisher of my faith, the one who made all things beautiful and perfect in his time for his grace and mercies throughout my period of study.

## ACKNOWLEDGMENT

I would like to express my sincere gratitude to God Almighty for wisdom and understanding for the successful completion of my project.

I would like to thank my supervisor PROF. S.E Omonigho for his patience and understanding towards me and the success of this project.

I would like to thank Dr. Mrs. Imoni for her love, care, patience and support during this project, her assistance really proved that she really want the best for me.

I would like to specially appreciate my parents Mr and Mrs. Omoh IMOOBE for their unwavering support, love, care, finance and prayers and also my siblings (Aliyu Rachel, IMOOBE Emmanuel, IMOOBE Salvation, Imoobe Success, Imoobe Wisdom and Imoobe Prosper) for their motivation to keep going.

I would like to appericiate my sugar daddy, odogwu paranran, big daddy (PROF. T.O.T IMOOBE) for his support financially, academically, morally and mentally

I would also like to appreciate my friends who turned family ( Ojezuabi Victoria, Godfrey Grace, Osamede Covenant, Abiode Francis and Freedom Heritage ) for their support and fun moments we had together.

A special shout out to all my roommates ( my bunkieeeeeee – COMRADE Stephen Marvellous, Aladejare Gift, Mohammed Favour, Eze Chisom, Thomas Debbie, Emmanuel Blessing, Vin-ighalo Ose, Imoobe Bernice, Imoobe Peace, Imaru Osato, Ara Francisca, Momoh Sharon) for their love in their own special ways and making hostel life sweet and accommodating. I love you all.

## TABLE OF CONTENTS

CERTIFICATION	iii
DEDICATION	iv
ACKNOWLEDGMENT	v
TABLE OF CONTENTS	vi
LIST OF TABLE	x
ABSTRACT	xi
CHAPTER ONE	1
INTRODUCTION	1
1.1 Aim and objectives	4
CHAPTER 2	5
LITERATURE REVIEW	5
2.1 Types of Dairy Products and Their Nutritional Value	9
2.1.1. Milk (Fresh, Pasteurized, UHT)	9
2.1.2. Fermented Dairy Products	11
2.1.3. Cheese	13
2.1.4. Butter and Cream	15
2.1.5. Evaporated, Condensed, and Powdered Milk	16
2.1.6. Frozen Dairy Desserts (Ice Cream, Gelato, Kulfi)	18
2.1.7. Traditional and Regional Dairy Products (Labneh, Skyr, Kumis, Qurt)	20
2.1.7.1 Fluid milk	21
2.1.8 Nutritional Roles and Health consideration of dairy products	22

2.2 Importance and Significance of Dairy Products	22
2.2.1 Contribution to Human Nutrition and Growth	23
2.2.2 Yoghurt as a Functional Dairy Product	23
2.2.3 Gut Microbiome and Immunity	24
2.2.4 Cardiometabolic Benefits	25
2.2.5 Bone Health and Osteoporosis Prevention	26
2.2.6 Role in Weight Management	26
2.2.7 Public Health and Food Security	27
2.2.8 Muscle Growth and Maintenance	27
2.2.9 Digestive and Immune Benefits	28
2.3 Safety and Hygiene	28
2.4 Hygiene and food-safety challenges in dairy products	29
2.5 Key points of failure in the dairy-to-yogurt chain	31
2.6 Routes and Risk Factors of Contamination	32
2.6.1.2 Common Pathogens in Milk and Yoghurt	33
2.6.1.3 Pathogenic Bacteria	34
2.6.1.4 Microbial Contamination of Yoghurt	34
2.6.1.5 Opportunistic Spoilage Microorganisms	35
2.7 Common Microbial Contaminants of Yoghurt	36
2.7.1 Yeasts and Moulds	36
2.7.2 Coliforms and Gram-Negative Bacteria	38
2.8. Public-Health Implications	40
2.8.1 Outbreaks and Clinical Impact	42

2.9 Antimicrobial resistance (AMR) and dairy	43
2.9.1 Surveillance, outbreaks and lessons learned	44
2.10 Control and Mitigation Strategies	45
2.10.2 Challenges and Future Perspectives	48
CHAPTER 3	49
METHODOLOGY	49
3.1 Sample Collection	49
3.2 Sterilization of materials	49
3.3 Preparation of media	49
3.3.1 Nutrient agar	49
3.3.2 Preparation of Salmonella Shigella agar	50
3.3.3 Preparation of Eosin Methylene Blue	50
3.3.4 MacConkey Agar	50
3.3.5 Mueller Hinton Agar	50
3.4 Isolation and characterization of bacteria	51
3.5 Gram Staining	51
3.6 Morphological Analysis	51
3.7 Biochemical test	52
3.7.1 Catalase test	52
3.7.2 Citrate Utilization Test	52
3.7.3 Urease Test	52
3.7.4 Hydrogen Sulphide (H <sub>2</sub> S) Test	52
3.7.5 Indole Test	53

3.7.6 Coagulase test	53
3.7.7 Sugar Fermentation Test	54
3.8 Antibiotics Susceptibility Test	54
CHAPTER FOUR	56
RESULTS	56
CHATER FIVE	66
DISCUSSIONS	66
Conclusion	70
REFERENCES	70

## LIST OF TABLE

Table 4.1 Total Heterothrophic Bacteria Count (Cfu/MI) Of Dairy Products Sold In Benin City.	57
Table 4.2 Total Bacteria Counts (Cfu/MI) Of Dairy Samples Sold In Benin City On Macconkey Agar.	58
Table4.3 Total Bacteria Count (Cfu/MI) Of Dairy Samples Sold In Benin City On <i>Salmonella Shigella</i> Agar	59
Table 4.4 Total Bacteria Counts (Cfu/MI) Of Dairy Samples Sold In Benin City On Eosin Methlyene Blue Agar.	60
Table 4.5 Cultural, Morphological And Biochemical Characteristics Of Bacteria Isolates From Dairy Products Sold In Benin City.	61
Table 4.7: Antibiotics Sensitivity Pattern Of Gram-Negative Isolates From Dairy Products	63
Table 8: Ph Values Of Dairy Samples Sold In Benin City.	64
Table 4.9: Total Titratable Acidity (Mg/L) Values Of Dairy Samples Sold In Benin City.	65

## ABSTRACT

Food safety remains a critical public health concern globally, with dairy products repeatedly implicated in food-borne illness outbreaks. This study aimed to detect enteric pathogens in dairy products sold in Benin City, Edo State. Dairy product samples comprising of branded (Hollandia, Nana, Cedaa, and Super Yogo) and locally processed (Nono milk, Kindoromo, Wara, and Maishanu) products were purchased for this study. Enumeration and isolation of bacteria was done using serial dilution and pour plate techniques on selective agar. Bacterial isolates were identified based on cultural, morphological and biochemical characteristics. The pH and total titratable acidity of samples were determined using a pH meter and acid/base titration method respectively, while antibiotic susceptibility testing was performed using the Kirby-Bauer disc diffusion method. The total heterotrophic bacteria count of the samples ranged from  $4.8 \pm 0.26$  to  $8.6 \pm 0.26 \times 10^8$  CFU/ml, while colony counts on MacConkey agar, *Salmonella-Shigella* agar, and Eosin methylene blue agar ranged from  $0.80 \pm 0.485$  to  $6.67 \pm 1.53 \times 10^7$  CFU/ml;  $1.13 \pm 0.31$  to  $6.87 \pm 0.61 \times 10^4$  CFU/ml and  $5.07 \pm 0.64$  to  $7.0 \pm 0.40 \times 10^3$  CFU/ml respectively. Six bacterial isolates were identified in this study, which include: *Escherichia coli*<sup>1</sup>, *Escherichia coli*<sup>2</sup>, *Salmonella* sp.<sup>1</sup>, *Salmonella* sp.<sup>2</sup>, *Enterobacter aerogenes* and *Serratia marcescens* with *Escherichia coli*<sup>1</sup>, *Escherichia coli*<sup>2</sup>, *Salmonella* sp.<sup>1</sup> and *Salmonella* sp.<sup>2</sup> having the highest frequency of occurrence of 33.33% each. The pH values of dairy samples ranged from  $3.82 \pm 0.01$  to  $6.96 \pm 0.03$ , while total titratable acidity ranged from  $0.17 \pm 0.02$  to  $1.86 \pm 0.02$  mg/l. Most isolates were resistant to antibiotics used, with resistance indices ranging from 0.1 to 0.8, with *Enterobacter aerogenes* and *Salmonella* sp.<sup>2</sup> recording the highest resistance index of 0.8, indicating multidrug resistance pattern. This study revealed that dairy products (especially in locally processed dairy products) sold in Benin City showed the enteric bacteria which may possibly be pathogenic with high multidrug resistance profiles, underscoring the need for improved hygiene practices, enforcement of pasteurization standards, and stringent microbiological monitoring in the dairy value chain.

## CHAPTER ONE

### INTRODUCTION

Dairy products are major component of human diets worldwide, valued for their nutritional content, including proteins, calcium, vitamins, and probiotics. However, their nutrient-rich composition also make them susceptible to microbial contamination, particularly by enteric pathogens microorganism originating from the gastrointestinal tract of human and animals (Ateba and Mbeve, 2020). Consumption of contaminated dairy product can lead to food-borne illnesses, posing significant public health risks globally (Delia, 2015).

Common enteric pathogens detected in dairy products include *Escherichia coli* (especially 0157:H7), *Salmonella* spp., *Listeria monocytogenes*, *Campylobacter* spp., and *Shigella* spp (Gundogan and Avci, 2014; Oliver *et al.*, 2020). These pathogens are responsible for outbreaks of gastroenteritis, listeriosis, salmonellosis, and other severe illnesses (Scallan *et al.*, 2011; WHO, 2021). The presence of these pathogens in milk and dairy product are often due to fecal contamination, inadequate pasteurization, or poor hygiene practices during production and handling (Dhanashekar *et al.*, 2023). Contamination can occur at multiple points in the dairy supply chain, on-farm contamination via infected animals or environmental exposure (Jolley *et al.*, 2018). Cross-contamination during milking due to improper sanitation (Ashraf *et al.*, 2019). Contamination during processing and storage especially in unpasteurized dairy products continue to be a major concern as they may harbor viable pathogens capable of causing infections (Ragab *et al.*, 2020). Detection of enteric pathogens in dairy products is crucial for food safety monitoring. Traditional culture-based methods remain the gold standard for detecting viable pathogens but are time consuming and labor-intensive (ISO, 2017). Advances in molecular detection techniques such as PCR and qPCR, have improved sensitivity, specificity, and speed of pathogen detection in complex dairy matrices (Munyaka *et al.*, 2016; Nocker *et al.*, 2021).

Multiplex PCR assay enable simultaneous detection of multiple pathogens (Kovacevic *et al.*, 2022). Immunological assays, including ELISA and lateral flow devices, offer rapid, though sometimes less sensitive, alternative for pathogens or toxin detection (Srinivasan *et al.*, 2019). Enteric pathogens in dairy products are a significant source of food-borne disease worldwide. Outbreaks linked to contaminated dairy have led to severe illnesses, hospitalization, and death, particularly in children, the elderly, and immunocompromised individuals (CDC 2019; European Food Safety Authority [EFSA], 2022). Moreover, the rise of antimicrobial-resistance strains among dairy-associated enteric pathogens exacerbates treatment challenges and public health burdens (Ahmed *et al.*, 2021). Economic impact of outbreak include healthcare cost, loss of productivity, and trade restrictions, underscore and the need for rigorous safety control (Havelaar *et al.*, 2015).

Effective prevention of enteric pathogen contamination in dairy products involves strict adherence to pasteurization protocols (WHO, 2021). Implementation of Good Agricultural Practices (GAP) and Good Manufacturing Practices (GMP), routine microbiological monitoring using sensitive detection method and education and training of dairy producers and handlers on hygiene (FAO, 2020; Oliveira *et al.*, 2021). Milk is recognized as the most widely consumed natural health food, enjoyed by people of all age groups across the world. This widespread consumption underpins the development of dairy farming and industry. Beyond milk production, dairy animals are also raised for meat. Animals kept in large herds and conventional dairy farms are typically more exposed to antimicrobials compared to those in small-scale or organic farms. The global demand for animal-derived foods is rising rapidly (Krehbiel *et al.*, 2013). In line with this demand, the global veterinary drug market grew from 8.65 billion USD in 1992 to 20 billion USD in 2010 and was projected to reach 42.9 billion USD by 2018 (Han *et al.*, 2014).

Worldwide, animal farming depends heavily on antimicrobials to maintain animal health and boost productivity. The U.S. Food and Drug Administration (FDA) (Food and Drug Administration 2010) estimated that in the United States, food animals account for about 80% of the country's annual antimicrobial consumption. Similarly, in 2012, the average antimicrobial use across 26 EU countries was reported as 116.4 mg/kg biomass for humans and 144.0 mg/kg for animals (Lhermie *et al.*, 2016). With growing populations, the demand for high-quality livestock products is expected to rise, and antimicrobial use in food animal production is projected to increase by over 67% by 2030 (Van Boeckel *et al.*, 2015).

The impact of antimicrobial resistance (AMR) is particularly concerning in developing nations such as India, where high rates of infectious diseases drive greater antimicrobial application to reduce illness and death. Reports from various regions in India confirm the presence of antimicrobial residues in milk and chicken, reflecting widespread usage in animal production (Maron *et al.*, 2013, Kakkar *et al.*, 2013, Basnyat 2015, Sawant *et al.*, 2007). However, accurately estimating antimicrobial use (AMU) in dairy farms is difficult due to poor record-keeping, limited veterinary oversight, reliance on farmers' self-treatment practices, and variations in farm management approaches (Diaz 2013). A 2012 survey by the World Organization for Animal Health (OIE) revealed that only 27% of member countries had formal systems in place to record AMU in livestock. Excessive antimicrobial use is a primary driver of AMR in animal husbandry. Both commensal and pathogenic bacteria are exposed to these drugs, and resistance develops. Key contributors to resistance include inappropriate dosing and the use of subtherapeutic antibiotic levels to promote animal growth (McManus 1997). Bacteria employ several mechanisms to resist antibiotics, such as: drug inactivation or modification, alteration of drug target sites, metabolic pathway changes, and reduced drug entry or enhanced efflux. The

presence of antimicrobial-resistant pathogens in food poses a direct threat to public health. Food-producing animals act as the main reservoirs for zoonotic pathogens. In dairy farming, the most common resistant strains include *Staphylococcus aureus*, *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella* spp. *S. aureus* is a leading cause of food-borne diseases, with milk and dairy products are frequently contaminated with its enterotoxigenic strains.

### **1.1 Aim and objectives**

The aim of this work was to detect enteric bacteria in dairy products sold in Benin city, Edo state.

The specific objective were to;

1. enumerate and isolate bacteria from dairy products.
2. identify enteric bacteria in dairy products using cultural, morphological and biochemical characteristics.
3. determine the pH and total titratable acidity of the samples.
4. determine the antibiotics susceptibility pattern of the enteric bacteria isolated from dairy products.
5. calculate the multiple antibiotics resistance (MAR) of the bacteria isolates.

## CHAPTER TWO

### LITERATURE REVIEW

Dairy products are foods derived from the milk of mammals such as cows, goats, and sheep. They include milk, yogurt, cheese, butter, and traditional fermented products such as nono. These foods are valued for their rich nutrient profile, providing high-quality proteins, fats, calcium, and vitamins such as vitamin B<sub>12</sub> and vitamin D, which are essential for growth, bone health, and metabolic functions (Ogunlade and Bamire, 2023). The Food and Agriculture Organization (FAO) considers milk a “complete food” because it contains almost all the essential nutrients required by the human body in adequate proportions. Dairy products therefore play an important role in addressing protein and micronutrient deficiencies, especially among children, pregnant women, and other vulnerable populations (FAO, 2022). In many developing countries, including Nigeria, they form an important part of the diet and contribute significantly to nutrition and food security. Milk and dairy foods are known to contain highly digestible proteins that help in tissue repair, growth, and the maintenance of body cells. They also provide calcium and phosphorus, which are vital for bone mineralization and dental health, and vitamin B<sub>12</sub>, which supports the production of red blood cells and neurological functions (Ogunlade and Bamire, 2023). Vitamin D, often fortified in processed milk and dairy products, enhances the absorption of calcium, helping to prevent bone-related diseases such as rickets and osteoporosis. According to the FAO, regular milk consumption can improve nutritional status, particularly among children who face protein-energy malnutrition in many low-income households. In rural Nigeria, traditional dairy products like nono (fermented milk), wara (soft cheese), and mai shanu (butter) are widely consumed. Although these foods provide essential nutrients, studies have shown that they sometimes present microbial safety concerns due to unhygienic processing and storage conditions (Oyedokun *et al.*, 2023). For example, an investigation in Abuja found the presence of

*Clostridium sporogenes* in ready-to-drink nono samples sold by Fulani vendors, highlighting potential risks of foodborne infections (Oyedokun *et al.*, 2023).

In Nigeria, milk production is largely dominated by nomadic and semi-nomadic Fulani pastoralists who move their herds in search of pasture and water. These pastoralists are the primary suppliers of raw milk and traditional dairy products across rural areas (Adewumi *et al.*, 2023). However, the country's modern dairy industry remains underdeveloped, and most of the milk consumed in urban centers is imported in powdered or processed form. Nigeria currently imports more than 70 percent of its dairy requirements to meet domestic demand, which continues to rise due to population growth and urbanization (Ogunlade and Bamire, 2023). The growing urban population has increased the consumption of processed milk, yogurt, cheese, and other dairy derivatives. This rising demand presents both opportunities and challenges: while it creates prospects for investment and job creation, it also exposes the weaknesses of Nigeria's local dairy production systems and infrastructure. Studies on the integration of small-scale dairy farmers into formal milk markets in Nigeria have revealed that only about half of the producers have access to organized milk collection systems or processing centers (Adewumi, Aderinkola and Akinsola, 2023). Factors such as distance to collection centers, lack of cooling facilities, low milk yields, and poor access to credit hinder the participation of pastoralists in formal markets. Consequently, most milk is sold informally, often in raw form, to local consumers or traders who process it into nono or wara. This informal structure not only limits the income of producers but also constrains quality control and the enforcement of food safety standards. A related study by (Oguntade and Bamire 2021) projected that Nigeria has sufficient cattle herd potential to produce the quantity of milk required to satisfy local demand if proper collection and processing systems are developed. However, the lack of adequate infrastructure, logistics, and investment continues

to impede progress in the dairy value chain. Despite the existing challenges, the Nigerian dairy sector presents a range of opportunities for economic diversification and rural development. Dairy farming contributes to employment, income generation, and the empowerment of women, who often engage in small-scale milk processing and marketing. Urbanization has led to the emergence of a growing middle class with changing dietary preferences, increasing the demand for fresh and processed dairy products. This growing market can stimulate local production if investments are directed toward improving breed quality, feed availability, and veterinary services (Oguntade and Bamire, 2021). Nevertheless, Nigeria's dairy production remains constrained by environmental, economic, and social factors. Extensive grazing practices, common among pastoralists, expose livestock to harsh climatic conditions, long migration routes, and conflicts with crop farmers. Furthermore, the effects of climate change such as increased temperature and erratic rainfall which have been shown to negatively influence milk yield and cattle productivity (Osuji *et al.*, 2023). The quality and safety of milk and dairy products in Nigeria are also major concerns. Research conducted in Ibadan metropolis revealed that raw milk samples often contain bacterial loads exceeding international safety standards (Adediran *et al.*, 2023). The presence of coliforms and other pathogens in unpasteurized milk highlights the need for stronger regulatory oversight and improved hygiene during milking, storage, and transportation. These safety challenges reduce consumer confidence and limit the market potential for locally produced milk. Modern processing and value addition such as pasteurization, fermentation under controlled conditions, and packaging are essential to improve product safety and shelf life. Small and medium-scale dairy processors can play a crucial role in this regard by linking pastoral milk producers to urban markets while ensuring compliance with quality standards. To revitalize the Nigerian dairy industry, several strategies are needed. Strengthening

herd productivity through cross-breeding, feed supplementation, and improved veterinary care can increase milk yields. Establishing milk collection centers closer to rural producers would reduce post-harvest losses and ensure that fresh milk reaches processors efficiently. Expanding the capacity of local processing plants for yogurt, cheese, butter, and other value-added dairy products can reduce Nigeria's dependence on imports and create employment opportunities. Policy reforms that support domestic production through credit facilities, subsidies, and public-private partnerships are also essential for sustainable growth (Keba *et al.*, 2020). Moreover, awareness campaigns on hygiene and food safety can help traditional processors improve the microbiological quality of their products. Environmental considerations, such as adopting climate-resilient livestock management systems, must also be integrated into national dairy development plans to safeguard productivity amid changing climatic conditions (Osuji *et al.*, 2023). Despite their nutritional importance, dairy products present food safety risks when handled under poor hygienic conditions. Studies have shown that raw milk sold in local markets often exceeds permissible bacterial limits (Oladipo *et al.*, 2020) reported total bacterial counts up to  $7.5 \times 10^7$  CFU/mL in raw cow milk from Minna Central Market, with isolates including *Escherichia coli*, *Staphylococcus aureus*, *Bacillus* spp., and *Salmonella typhi*. These organisms are associated with diarrheal disease, food poisoning, and typhoid fever (Abdulkadir *et al.*, 2021) also detected *Staphylococcus aureus* in 7.4% of dairy samples from pastoral communities in Niger State, with many isolates showing multidrug resistance. Locally fermented milk (nono) poses similar risks (Odeyemi *et al.*, 2022) found high microbial counts and multidrug-resistant isolates in nono sold in Abuja, highlighting the dangers of consuming unpasteurized products. Knowledge, Attitude, and Practice (KAP) studies among Fulani milk processors revealed that while over half of respondents understood the importance of milk hygiene, only about one-third

implemented good practices due to lack of infrastructure, reliance on traditional processing, and limited access to clean water (Fasanmi *et al.*, 2023).

The absence of cold chain infrastructure further worsens microbial contamination risks, as milk is often transported and sold without refrigeration. This not only compromises safety but also reduces shelf life and leads to economic losses. These findings emphasize the urgent need for improved hygiene education, infrastructure support, and enforcement of quality standards to ensure consumer safety.

## **2.1 Types of Dairy Products and Their Nutritional Value**

Dairy products are a globally consumed food group produced predominantly from mammalian milk, including cow, buffalo, goat, sheep, and camel milk. They supply high-quality protein, essential micronutrients such as calcium, phosphorus, vitamin B<sub>12</sub>, and riboflavin, and, depending on processing, variable amounts of fat and lactose (Pereira, 2014). The profile of dairy consumption and the dominant product types differ by region for example, fluid milk in Europe and North America, fermented milks in parts of the Middle East and Africa, and concentrated products such as cheese and ghee in other regional cuisines. These products are nutritionally important both for growth and maintenance of bone health and play significant roles in cultural diets and national food guidelines (Comerford *et al.*, 2021).

### **2.1.1. Milk (Fresh, Pasteurized, UHT)**

Milk is one of the most widely consumed and nutritionally valuable foods in the world, obtained from mammals such as cows, goats, buffaloes, sheep, and camels. It provides a unique balance of essential nutrients that contribute significantly to human growth and development. Milk can be consumed in various forms, including fresh (raw), pasteurized, and ultra-high temperature (UHT)

processed. Each type differs in processing methods, shelf life, safety, and nutritional quality (Pereira, 2014). Fresh milk, often consumed directly after milking, contains natural enzymes and microorganisms. While it has a rich taste and high nutrient retention, it poses potential health risks because it can harbor harmful bacteria such as *Salmonella*, *Listeria monocytogenes*, and *E. coli*, which can cause foodborne illnesses (Claeys *et al.*, 2013). Therefore, pasteurization and UHT treatments were developed to make milk safe for human consumption while retaining its nutritional benefits.

Pasteurization is a thermal process in which milk is heated to about 72°C for 15 seconds (high-temperature short-time method) and then rapidly cooled. This treatment destroys most pathogenic and spoilage microorganisms without significantly affecting milk's nutritional composition (Lewis and Deeth, 2017). Research shows that pasteurization retains nearly all the proteins, fats, and minerals, with only slight losses in heat-sensitive vitamins such as vitamin C, thiamine, and folate (Knutsen *et al.*, 2021). The process also enhances milk safety by eliminating disease-causing bacteria that can lead to tuberculosis and brucellosis, thus making it the global standard for safe milk consumption. Pasteurized milk provides high-quality proteins such as casein and whey, which support tissue repair, muscle growth, and immune function. It also supplies bioavailable calcium and phosphorus, which are essential for bone formation, strong teeth, and nerve signaling. UHT (ultra-high temperature) treatment is a more intensive heating process in which milk is heated to temperatures between 135°C and 150°C for a few seconds and then aseptically packaged. This process sterilizes the milk completely, giving it a shelf life of up to six months without refrigeration (Bai *et al.*, 2023). UHT milk is especially valuable in tropical regions and developing countries where cold storage facilities are limited. Though it can cause slight changes in taste and texture, UHT milk retains its major nutrients, including proteins, fats,

and most vitamins and minerals. Some studies report small losses of vitamin B<sub>6</sub>, vitamin B<sub>12</sub>, and ascorbic acid during UHT processing and long-term storage (Wang *et al.*, 2024). However, these losses are minimal compared to the benefits of extended shelf life and food safety.

Nutritionally, milk is a powerhouse food that provides nearly every nutrient needed for human survival. It contains about 3.5% protein, 4–5% fat, 4.5% lactose, and essential minerals such as calcium, magnesium, and potassium (Pereira, 2014). Milk is also a natural source of vitamins A, D, E, and K (fat-soluble vitamins), as well as B vitamins, especially riboflavin (B<sub>2</sub>) and vitamin B<sub>12</sub>, which are crucial for red blood cell formation and energy metabolism (Roy, 2020). The protein in milk is of high biological value, supplying all essential amino acids required by the human body. Its carbohydrate content (lactose) provides a steady source of energy, while milk fat delivers both energy and important fatty acids. Calcium and vitamin D in milk work synergistically to promote bone density and prevent osteoporosis, particularly in children and the elderly. Milk is one of the most nutrient-dense natural foods. It is an excellent source of complete proteins, containing both casein and whey, which are easily digestible and support muscle growth and repair. It also provides bioavailable calcium and phosphorus, which are essential for bone development and teeth health, as well as riboflavin and vitamin B<sub>12</sub> for energy metabolism and red blood cell formation (Roy 2020). Milk's fat content provides energy and fat-soluble vitamins (A, D, E, K), though lower-fat versions are recommended for individuals managing calorie or cholesterol intake.

### **2.1.2. Fermented Dairy Products**

Fermented dairy products are among the most widely consumed traditional foods across many regions, including Nigeria and other parts of Africa, where they play a vital role in nutrition, food preservation, and rural livelihoods. These products are typically produced by inoculating

fresh milk with specific lactic acid bacteria (LAB), which ferment lactose into lactic acid. This biochemical process reduces the pH of the milk, leading to coagulation of casein proteins, development of a tangy flavor, and a thicker consistency (Moore, 2018). Fermentation not only imparts desirable sensory qualities but also improves the microbiological safety and shelf life of milk, especially under tropical conditions where refrigeration may be limited (Ogundele, 2020).

In Nigeria, several types of locally fermented milk products are produced, with nono (also called fura da nono) and wara being the most popular. Nono is a traditional fermented cow milk consumed mainly in northern Nigeria, often served with millet balls (fura). The fermentation process is spontaneous, typically initiated by LAB such as *Lactobacillus plantarum*, *Leuconostoc mesenteroides*, and *Streptococcus thermophilus* present in the environment or leftover milk residues (Abdullahi *et al.*, 2019). Similarly, wara is a soft cheese-like produced by coagulating milk using plant-based coagulants or microbial starters, providing a good source of protein and calcium for local consumers (Ogunbanwo and Sanni, 2017). Fermentation enhances not only the preservation but also the nutritional and functional quality of milk. Studies show that fermented milk retains most of the original macronutrients of fresh milk such as proteins, calcium, and vitamin B while improving digestibility due to the partial hydrolysis of lactose (Tong *et al.*, 2021). This makes fermented milk particularly suitable for lactose-intolerant individuals. Moreover, the metabolic activity of LAB produces bioactive compounds such as peptides and short-chain fatty acids, which exhibit antihypertensive, antioxidant, and immunomodulatory effects (Sumi, 2023). Probiotic strains like *Lactobacillus acidophilus* and *Bifidobacterium bifidum* found in many fermented dairy products help maintain gut microbiota balance, enhance nutrient absorption, and strengthen immune function. Regular consumption of yogurt and similar fermented milk products has been associated with improved gastrointestinal health, reduced risk

of type 2 diabetes, and better bone density (Ugbaja *et al.*, 2022). Beyond health benefits, locally fermented milk also supports rural economies by providing income for pastoralists and small-scale dairy processors, contributing to food security and sustainable livelihoods (Omore *et al.*, 2019). Locally fermented milk products such as nono, wara, and yogurt represent an integral part of Nigeria's dairy culture. Their nutritional, probiotic, and socioeconomic importance underscores the need for improved hygienic production methods, microbial standardization, and preservation technologies to expand their market potential while maintaining their traditional value.

### **2.1.3. Cheese**

Cheese represents one of the oldest and most diverse categories of dairy products globally, produced through the coagulation of milk proteins (casein) using enzymes such as rennet or by acidification, followed by the separation of curds and whey. This transformation concentrates the milk solids and allows for preservation, flavor development, and textural modification (O'Brien, 2004). The composition and characteristics of cheese vary significantly based on the type of milk used (cow, goat, buffalo, or sheep), fat content, degree of fermentation, moisture level, and the duration and conditions of ripening (Fox *et al.*, 2017). Depending on these factors, cheeses can be classified into several categories—fresh, soft, semi-soft, hard, and blue-veined varieties.

**2.1.3.1 Fresh cheeses** such as paneer, ricotta, and cottage cheese are unripened, retaining high moisture and a mild, milky flavor. Paneer, which is particularly popular in South Asia and parts of Nigeria, is produced by adding food-grade acids like lemon juice or vinegar to hot milk, resulting in curd formation without rennet, making it suitable for vegetarian diets (Kumar and Rai, 2018). Soft cheeses such as brie, camembert, and feta undergo short ripening periods and contain higher moisture content, giving them creamy textures and tangy flavors. Semi-soft

cheeses, like havarti, edam, and monterey jack, balance firmness and moisture, making them versatile in cooking and table use.

**2.1.3.1 Hard cheeses** which include cheddar, parmesan, and gouda, are aged for longer durations, resulting in lower water content and more concentrated flavor profiles. The maturation process enhances the development of flavor compounds and texture through enzymatic and microbial activity (McSweeney, 2019). Meanwhile, blue cheeses such as roquefort and stilton are ripened with specific *Penicillium* molds, which create distinctive blue veins and strong, sharp flavors. In West Africa, local adaptations of cheese exist wara in Nigeria, for instance, is a fresh, soft cheese traditionally made from cow milk using plant-based coagulants like *Calotropis procera* extract, serving as an important source of dietary protein in local diets (Ogunbanwo and Sanni, 2017).

Nutritionally, cheese is a highly concentrated source of essential nutrients, providing high-quality protein, calcium, phosphorus, zinc, and vitamins A, D, and K (Paszczyk, 2022). Since much of the water is removed during cheesemaking, its nutrient density is several times higher than that of milk. Regular consumption of moderate quantities contributes to bone health and muscle development due to its rich calcium and protein content. However, depending on the variety, some cheeses may be high in saturated fat and sodium, potentially increasing cardiovascular risks when consumed excessively (de Oliveira *et al.*, 2020). Beyond its macronutrient content, certain aged cheeses also contain bioactive peptides formed during proteolysis that exhibit antihypertensive and antioxidant effects, contributing to cardiovascular and metabolic health (Sumi, 2023). Additionally, the fermentation process in some cheese varieties introduces beneficial bacteria that can promote gut microbiota balance, similar to probiotic effects observed in fermented milk products (Tong *et al.*, 2021). Cheese remains a versatile and nutrient-rich dairy product with wide-ranging varieties catering to different tastes,

dietary preferences, and culinary applications. Whether traditional local forms like wara or globally renowned types such as cheddar and brie, cheese continues to play an important role in nutrition, culture, and gastronomy worldwide.

#### **2.1.4. Butter and Cream**

Cream is defined as the high-fat portion of milk that naturally rises to the top when fresh milk is left to stand or is mechanically separated using centrifugation. It contains milk fat globules dispersed in an aqueous phase and typically has a fat content ranging from 18% to 40%, depending on the type (light, whipping, or heavy cream) (Fox *et al.*, 2017). Cream serves as the base ingredient for butter and is widely used in both sweet and savory preparations such as soups, desserts, sauces, and confectioneries. Its smooth texture and rich mouthfeel make it a valuable component in food processing and culinary applications (Pădureț *et al.*, 2021). Butter, on the other hand, is a semi-solid emulsion obtained by churning cream until the fat globules coalesce and separate from the liquid phase, known as buttermilk. This process disrupts the membrane surrounding the milk fat globules, allowing them to aggregate into a continuous fat matrix that traps small amounts of water and milk solids (Walstra *et al.*, 2019). Butter typically contains about 80–82% milk fat, 16–18% water, and 1–2% milk solids-not-fat (MSNF), depending on processing methods and regional standards. The quality and flavor of butter are influenced by factors such as the composition of the cream, fermentation, salt addition, and storage conditions (Kučerová *et al.*, 2022).

Butter can be categorized into sweet cream butter, made from fresh pasteurized cream, and cultured butter, made from fermented cream that develops a tangy flavor due to lactic acid bacteria activity (Pădureț *et al.*, 2021). A derivative of butter, clarified butter commonly known as ghee is produced by heating butter to remove water and milk solids, leaving behind pure

butterfat. Ghee has a high smoke point (approximately 250°C), long shelf life, and distinct nutty flavor, making it especially popular in South Asia, the Middle East, and parts of Africa for cooking and traditional medicine (Sarkar and Choudhary, 2020).

Nutritionally, butter and cream are concentrated sources of milk fat, providing an energy-dense supply of calories along with essential fat-soluble vitamins such as A, D, E, and K (Paszczyk, 2022). They also contain short- and medium-chain fatty acids, including butyric, capric, and lauric acids, which are easily digested and metabolized for rapid energy production (Jensen, 2019). However, their protein and calcium contents are relatively low compared to other dairy products such as milk and cheese, making them primarily sources of dietary fat. While butter and cream contribute positively to sensory qualities, texture, and flavor in foods, excessive consumption may have health implications due to their high levels of saturated fatty acids and cholesterol, which have been linked to increased low-density lipoprotein (LDL) cholesterol levels and cardiovascular risk (de Oliveira *et al.*, 2020). Nonetheless, recent studies suggest that moderate intake of dairy fats, particularly from natural sources like butter, may not be as harmful as once believed and could even contribute to a balanced diet when consumed in moderation (Pimpin *et al.*, 2016). Both butter and cream represent important dairy derivatives that combine culinary versatility with nutritional richness. Their traditional and industrial significance continues to expand, though awareness of portion control and balance with other nutrient sources remains essential for maintaining overall health.

#### **2.1.5. Evaporated, Condensed, and Powdered Milk**

Evaporated, condensed, and powdered milk are important shelf-stable dairy products produced by removing a substantial portion of water from fresh milk to enhance its stability, reduce bulk, and extend shelf life. These products are essential in regions where refrigeration is limited and

serve as key ingredients in various food formulations, including infant formulas, bakery items, beverages, and confectionery (Luna, 2016). The fundamental principle behind their production is the concentration or complete dehydration of milk while retaining its nutritional and sensory characteristics. Evaporated milk is made by removing approximately 60% of the water content from whole or skimmed milk through vacuum evaporation under controlled heat and pressure. The process results in a viscous, creamy liquid that is then homogenized, canned, and sterilized at high temperature (115–120°C) to ensure microbial safety (Taneja and Taneja, 2017). Since it is unsweetened, evaporated milk can be diluted with an equal volume of water to reconstitute milk for drinking or cooking. Its caramelized flavor and thick texture make it popular in desserts, sauces, and coffee preparations.

Sweetened condensed milk, on the other hand, is produced through a similar evaporation process but with the addition of about 40–45% sucrose before concentration. The high sugar content acts as a preservative by lowering water activity, preventing microbial growth, and extending storage stability without sterilization (Niero *et al.*, 2019). This variety is widely used in confectionery, bakery, and dessert products, such as fudge, caramel, and ice cream. Because of its energy density, it has long been used in emergency food rations and as a supplementary source of calories in undernourished populations (Suneeta and Sharma, 2021).

Powdered milk (also called dried milk) was produced by complete removal of water usually 95–98% from milk using spray drying or roller drying techniques. The resulting fine powder is highly stable, lightweight, and easy to store and transport, making it ideal for regions without consistent refrigeration (Sava *et al.*, 2018). When reconstituted with water, powdered milk closely resembles fresh milk in nutritional composition and flavor, although some minor losses of volatile compounds and heat-sensitive vitamins occur during drying. To compensate for these

losses, many commercial powdered milks are fortified with vitamins A and D, enhancing their nutritional quality (Rybak *et al.*, 2020). From a nutritional standpoint, evaporated, condensed, and powdered milk contain concentrated levels of milk solids mainly proteins, calcium, phosphorus, and other minerals making them nutritionally comparable to fresh milk (El-Zahar *et al.*, 1999). The dehydration process increases the protein and energy density per gram, which is particularly beneficial in food aid and infant feeding programs. In addition, the low moisture content and hermetic packaging prevent spoilage, allowing these products to remain stable for months or even years under ambient conditions. Powdered milk, in particular, has become indispensable in food manufacturing, where it serves as a functional ingredient that enhances texture, emulsification, and flavor (Maldonado *et al.*, 2019). Evaporated, condensed, and powdered milks are innovative dairy products that combine long shelf life, portability, and nutritional value. Their role extends beyond household use to industrial processing, humanitarian aid, and global food security efforts, particularly in developing regions where fresh milk supply and cold-chain infrastructure are limited. They are shelf-stable products that retain most nutrients of fresh milk but in concentrated form. Sweetened condensed milk is energy-dense and high in sugar (Pereira, 2014).

#### **2.1.6. Frozen Dairy Desserts (Ice Cream, Gelato, Kulfi)**

Frozen dairy desserts encompass a wide range of products made primarily from milk, cream, sweeteners, and flavoring agents that are frozen under controlled conditions to produce a smooth, palatable texture. The production process involves simultaneous freezing and agitation to prevent the formation of large ice crystals and to incorporate air, resulting in a light and creamy consistency (FAO, 2020). The air incorporated during freezing, known as “overrun,” significantly influences the texture, volume, and sensory characteristics of the final product. Ice

cream typically contains 10–16% milk fat and up to 100% overrun, while frozen custards, gelato, sherbet, and frozen yogurts vary in fat, sugar, and air content depending on formulation and regional preferences (Muse and Hartel, 2004). Gelato, the Italian-style frozen dessert, differs from traditional ice cream in its formulation and processing. It generally contains less fat (4–8%) and less air (20–30% overrun), resulting in a denser and silkier texture (Goff and Hartel, 2013). Gelato is also served at slightly warmer temperatures than ice cream, enhancing its flavor perception and creaminess. Kulfi, a traditional frozen dessert popular in South Asia, is produced by slowly simmering milk to reduce its water content and concentrate solids, followed by sweetening and slow freezing without agitation. This lack of aeration gives kulfi its distinctive dense and chewy texture and rich caramelized flavor due to partial lactose caramelization during milk reduction (Aneja *et al.*, 2002).

Frozen dairy desserts vary widely in composition, but most provide small amounts of high-quality protein, calcium, and phosphorus derived from milk solids. However, they are also typically high in added sugars and saturated fats, which can contribute to excessive calorie intake when consumed frequently (Akbari *et al.*, 2019). According to the World Health Organization (WHO, 2023), diets high in added sugars are associated with increased risk of obesity, type 2 diabetes, and cardiovascular diseases. Thus, while frozen dairy desserts can contribute beneficial nutrients from milk, moderation is essential. In response to health concerns, the food industry has developed reduced-fat, low-sugar, and functional frozen desserts containing probiotics, fiber, and plant-based alternatives. Frozen yogurt, for example, incorporates live bacterial cultures (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*), which may improve gut microbiota and digestive health (Ranadheera *et al.*, 2016). Similarly, the rise of non-dairy frozen desserts made from soy, almond, oat, or coconut milk reflects a growing consumer demand for lactose-

free and vegan options (Sun-Waterhouse, 2016). From a sensory and technological standpoint, factors such as the size of ice crystals, fat globule distribution, emulsifier-stabilizer systems, and freezing rate determine the smoothness and quality of frozen desserts (Akin *et al.*, 2017). Proper control of these parameters ensures a desirable balance between creaminess, mouthfeel, and flavor release. Frozen dairy desserts like ice cream, gelato, and kulfi hold both cultural and nutritional significance worldwide. They combine milk's nutritional benefits with indulgent sensory qualities. Nonetheless, because of their high sugar and fat content, they should be consumed occasionally rather than as staple foods, aligning with dietary guidelines for balanced nutrition and chronic disease prevention.

#### **2.1.7. Traditional and Regional Dairy Products (Labneh, Skyr, Kumis, Qurt)**

Across the world, milk is adapted into a wide range of traditional and regional products that meet cultural, environmental, and nutritional needs. These products not only preserve milk in climates with limited refrigeration but also enhance its digestibility and nutritional value through fermentation, drying, or concentration processes (García-Burgos *et al.*, 2020). For example, Labneh, a strained yogurt from the Middle East is rich in protein and low in fat, commonly consumed as a spread or dip. Similarly, Skyr from Iceland, often regarded as yogurt, is technically a soft cheese produced by fermentation with *Streptococcus thermophilus* cultures, resulting in high protein and low fat content (Pétursson *et al.*, 2021). In Central Asia, Kumis fermented mare's milk contains lactic acid bacteria and yeasts that generate a lightly alcoholic, probiotic beverage. Another regional innovation is Qurt (dried cheese balls), valued for its nutrient density and long shelf life, making it essential for nomadic communities (Sady *et al.*, 2020).

In Africa, particularly Nigeria, milk is locally transformed into culturally significant products such as Nono (or Nunu), Wara, and Fura da Nono. Nono is a spontaneously fermented cow's milk similar to yogurt, widely consumed among Fulani pastoralists and northern Nigerian households. It is rich in lactic acid bacteria that improve flavor, safety, and gut health but often produced under unhygienic conditions, which can compromise quality (Abdullahi *et al.*, 2019). Fura da Nono, a thick beverage made by blending fermented milk (Nono) with spiced millet balls (Fura), provides carbohydrates, protein, and essential minerals like calcium and phosphorus, making it a balanced, energy-dense traditional food (Yusuf *et al.*, 2020). Wara, another common Nigerian dairy product, is a soft, fresh cheese prepared by coagulating cow's milk with plant extracts such as *Calotropis procera* or lemon juice. It serves as a nutritious meat substitute, supplying high-quality protein and fats but has a short shelf life due to the absence of preservatives (Adetunji *et al.*, 2008).

Globally and locally, traditional dairy products remain vital for food security, nutrition, and cultural identity. They provide accessible sources of protein, probiotics, and micronutrients, particularly in low-resource environments. In Nigeria, improving hygiene, pasteurization, and the use of defined starter cultures could enhance the safety and consistency of Nono, Fura da Nono, and Wara while preserving their traditional qualities and economic importance (Tossou *et al.*, 2025).

#### **2.1.7.1 Fluid milk**

Fluid milk is the unfermented beverage consumed fresh or pasteurized. Whole milk typically provides 3–4% fat, 3–3.5% protein, and 4.8–5% lactose; it is commonly fortified with vitamin D in many countries (Pereira, 2014). It is a principal contributor of calcium and high-biological-

value protein in national diets. Typical nutrition per 100 g: 60–70 kcal, protein 3.2 g, fat 3.3 g, carbohydrate 4.8–5 g, calcium  $\approx$ 110–125 mg (Linehan, 2024).

### **2.1.8 Nutritional Roles and Health consideration of dairy products**

Macronutrients and Micronutrients

Fermentation Benefits:

Fermentation reduces lactose content, allowing better tolerance among lactose-intolerant individuals, and introduces probiotics that may have gut health and immune-modulatory effects (Hadjimbei *et al.*, 2022; Aryana and McGrew, 2017).

Saturated Fat and Sodium

### **2..2 Importance and Significance of Dairy Products**

Dairy products occupy a central role in global diets and contribute significantly to human nutrition and public health. They include milk, yoghurt, cheese, butter, cream, and fermented products, each offering a unique nutrient profile and functional benefits. Dairy provides high-quality, complete protein with all essential amino acids, bioavailable calcium, phosphorus, magnesium, riboflavin, vitamin B12, and, in many regions, vitamin D through fortification (Chen *et al.*, 2021). These nutrients are essential for skeletal health, neuromuscular performance, energy metabolism, and immune function. The “dairy matrix” the combination of proteins, fats, minerals, and bioactive compounds in dairy enhances nutrient absorption and exerts synergistic effects on health outcomes (Savaiano *et al.*, 2020).

### **2.2.1 Contribution to Human Nutrition and Growth**

Dairy is one of the richest and most reliable dietary sources of calcium, which is vital for bone development, muscle contraction, nerve transmission, and blood clotting. The bioavailability of calcium in milk and yoghurt is superior to that of most plant sources because of the favorable ratio of calcium to phosphorus and the presence of lactose, which enhances intestinal absorption (Cashman, 2020). Dairy proteins, particularly casein and whey, stimulate muscle protein synthesis and promote linear growth in children, which is why dairy consumption is positively correlated with height and lean body mass in multiple populations (Dror and Allen, 2014). Dairy products, including milk, yogurt, cheese, and butter, are important components of the human diet worldwide and are recognized for their nutrient density and health-promoting properties (Wallace *et al.*, 2021). They supply a rich combination of macronutrients, vitamins, and minerals that are essential for growth, development, and maintenance of health across all age groups (Chen *et al.*, 2021). Beyond their nutritional role, dairy products contribute to the global economy, providing employment through production, processing, and distribution, particularly in rural areas (Gudi *et al.*, 2020).

### **2.2.2 Yoghurt as a Functional Dairy Product**

Among dairy products, yoghurt is uniquely valued for its fermentation process and functional properties. Yoghurt is produced by inoculating milk with *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*, which convert lactose into lactic acid, lowering pH and giving yoghurt its characteristic texture and tangy taste (McKinley, 2005). This fermentation reduces lactose content, improving tolerance among individuals with lactose mal-digestion a widespread condition in many populations (Savaiano *et al.*, 2020). Furthermore, many modern yoghurts are supplemented with probiotic strains such as *Lactobacillus acidophilus* or

*Bifidobacterium animalis*, which survive gastrointestinal transit and confer health benefits, including modulation of gut microbiota and immune enhancement (Hadjimbei *et al.*, 2022).

### **2.2.3 Gut Microbiome and Immunity**

Milk contains high-quality protein in the form of casein and whey, which together provide all nine essential amino acids required for human growth and tissue repair (Feng *et al.*, 2022). Dairy is the primary dietary source of bioavailable calcium and phosphorus, crucial for skeletal health, and it also contributes riboflavin, vitamin B<sub>12</sub>, and, when fortified, vitamin D (Wallace *et al.*, 2021). The bioavailability of calcium from milk is enhanced by the presence of lactose and casein phosphopeptides, which improve intestinal absorption (Chen *et al.*, 2021). The nutritional composition of dairy makes it particularly valuable in areas where diets lack diversity and other animal-source foods are limited. The human gut microbiome is increasingly recognized as a key determinant of immune function, metabolic health, and even neurological well-being. Yoghurt consumption positively alters gut microbial composition by increasing beneficial genera such as *Bifidobacterium* and *Lactobacillus* while reducing potentially pathogenic species (Le Roy *et al.*, 2022). In a randomized controlled trial (RCT), daily consumption of yoghurt fortified with *Lacticaseibacillus rhamnosus* significantly reduced the incidence and duration of upper respiratory tract infections among school-aged children, suggesting immune-boosting effects (Konlan *et al.*, 2023). Similar results were observed in elderly populations where probiotic yoghurt enhanced natural killer cell activity, increased serum interferon- $\gamma$ , and improved overall immune surveillance (Kawashima *et al.*, 2017). These findings are of public health importance, particularly for vulnerable populations at the extremes of age or those with weakened immunity.

#### 2.2.4 Cardiometabolic Benefits

Multiple large cohort studies and meta-analyses show that yoghurt consumption is inversely associated with several cardiometabolic risk factors. A meta-analysis of prospective studies reported that individuals with the highest yoghurt intake had a 12–17% lower risk of developing type 2 diabetes compared to those with the lowest intake (Gijsbers *et al.*, 2016). The mechanisms include improved insulin sensitivity, modulation of gut microbiota, and reduction in systemic inflammation. Yoghurt proteins have been shown to stimulate the release of gut hormones such as GLP-1 and PYY, which enhance satiety and regulate appetite, thereby supporting weight management (Panahi and Tremblay, 2016). Additionally, fermented dairy products like yoghurt are associated with improved lipid profiles and lower blood pressure compared with non-fermented dairy. Bioactive peptides released during fermentation possess angiotensin-converting enzyme (ACE)-inhibitory activity, contributing to vasodilation and reduced hypertension (Astrup *et al.*, 2020). Large-scale analyses indicate that yoghurt consumption of at least 200 g/day is associated with modest reductions in cardiovascular disease incidence and all-cause mortality (Chen *et al.*, 2021).

Contrary to earlier concerns about the saturated fat content of dairy, recent evidence indicates that dairy consumption has neutral or even protective effects on several cardiometabolic risk factors (Chen *et al.*, 2021). Prospective cohort studies show that moderate dairy intake is associated with a lower risk of hypertension, stroke, and metabolic syndrome (Feng *et al.*, 2022). Yoghurt, in particular, is consistently linked with a reduced risk of type 2 diabetes, possibly due to its probiotic content and effects on gut microbiota (Gudi *et al.*, 2020). Some research suggests that cheese may have less impact on blood LDL cholesterol than butter despite similar fat content, highlighting the importance of the food matrix (Wallace *et al.*, 2021).

### **2.2.5 Bone Health and Osteoporosis Prevention**

The role of dairy in maintaining bone health is one of the most well-established nutritional relationships. Yoghurt is an excellent source of calcium, phosphorus, magnesium, and high-quality protein, all of which are critical for bone mineralization. Intervention trials demonstrate that daily intake of vitamin D- and calcium-fortified yoghurt significantly improves serum 25-hydroxyvitamin D, reduces parathyroid hormone (PTH) levels, and lowers bone resorption markers such as TRAP5b and CTX in elderly individuals (Chapuy *et al.*, 1994). These effects translate to improved bone mineral density and lower risk of fractures, which is crucial for preventing osteoporosis and frailty in aging populations. One of the most widely studied benefits of dairy consumption is its effect on bone development and maintenance. Calcium, phosphorus, and vitamin D are critical nutrients for bone mineralization, and multiple studies have linked habitual dairy intake with higher bone mineral density (BMD) and reduced risk of osteoporosis (Wallace *et al.*, 2021). Meta-analyses of cohort studies report that milk and yogurt consumption in childhood and adolescence supports peak bone mass attainment, which is a major determinant of future fracture risk (Gudi *et al.*, 2020). In older adults, daily consumption of dairy products has been associated with a 10–20% lower risk of hip fracture, especially when combined with vitamin D fortification (Feng *et al.*, 2022).

### **2.2.6 Role in Weight Management**

Regular yoghurt consumption has been linked with better weight control and prevention of obesity. Its high protein content increases satiety, which can help reduce overall caloric intake. Observational studies from the Nurses' Health Study and Health Professionals Follow-up Study found that yoghurt was consistently associated with less weight gain over a 4-year follow-up compared to other snack foods (Mozaffarian *et al.*, 2011). This effect may be partly mediated by

its impact on gut microbiota composition and short-chain fatty acid production, which influence energy metabolism.

### **2.2.7 Public Health and Food Security**

At the population level, dairy plays a key role in addressing micronutrient deficiencies such as calcium and vitamin B<sub>12</sub> deficiency, particularly in low- and middle-income countries (Wallace *et al.*, 2021). School milk programs have been shown to improve growth outcomes and reduce malnutrition in children (Feng *et al.*, 2022). Economically, dairy production supports millions of livelihoods globally, with significant contributions to rural incomes and food security (Gudi *et al.*, 2020). In Nigeria and other African nations, smallholder dairy farming is an important income source, and policies that promote safe milk production and distribution can improve both nutrition and economic development. Yoghurt also has significant implications for food security and public health nutrition. Its relatively long shelf life, portability, and cultural acceptability make it a practical means of delivering essential nutrients in school feeding programs and maternal nutrition interventions (Kumwend *et al.*, 2020). Fortified yoghurt has been successfully used to address micronutrient deficiencies in low-income populations, particularly deficiencies of vitamin D, iron, and iodine (Hadjimbei *et al.*, 2022).

### **2.2.8 Muscle Growth and Maintenance**

Dairy proteins, particularly whey, are rapidly digested and rich in leucine, a branched-chain amino acid that stimulates muscle protein synthesis (Wallace *et al.*, 2021). This makes dairy an important dietary tool for athletes, elderly adults at risk of sarcopenia, and individuals recovering from illness or surgery. Studies have shown that including dairy in the diet during weight loss programs helps preserve lean muscle mass while promoting fat loss (Chen *et al.*, 2021). Regular

milk consumption after resistance training has also been shown to enhance gains in muscle mass and strength compared to carbohydrate-only post-workout beverages (Gudi *et al.*, 2020).

### **2.2.9 Digestive and Immune Benefits**

Fermented dairy products such as yogurt and kefir provide probiotics beneficial live microorganisms that help maintain gut microbial balance and support digestion (Jang *et al.*, 2024). Regular consumption of probiotic-rich dairy has been linked to improved lactose tolerance among lactose-intolerant individuals and to reduced symptoms of certain gastrointestinal disorders (Chen *et al.*, 2021). Additionally, bioactive peptides produced during fermentation can exert immunomodulatory, antihypertensive, and antioxidant effects (Jang *et al.*, 2024).

### **2.3 Safety and Hygiene**

Despite its benefits, yoghurt production must adhere to strict hygiene and cold-chain requirements to prevent contamination. Although the acidic environment inhibits many pathogens, post-processing contamination with organisms such as *Listeria monocytogenes* or *Salmonella* can occur if sanitary protocols are not followed (Oliveira *et al.*, 2018). Therefore, good manufacturing practices (GMP) and hazard analysis critical control point (HACCP) systems are essential in yoghurt production facilities.

Dairy products, and yoghurt in particular, are nutrient-dense foods with far-reaching implications for human health. Beyond supplying protein, calcium, and vitamins, yoghurt offers additional benefits such as improved lactose tolerance, positive modulation of gut microbiota, enhanced immune function, and protection against cardiometabolic diseases. Evidence from epidemiological studies, randomized clinical trials, and systematic reviews consistently supports

its role in reducing the risk of type 2 diabetes, improving weight management, maintaining bone health, and lowering cardiovascular risk. Public health strategies increasingly promote yoghurt consumption as a cost-effective intervention for improving dietary quality and reducing micronutrient deficiencies. Fortification with vitamin D, calcium, and probiotics can further amplify these benefits, making yoghurt an indispensable part of healthy dietary patterns worldwide. Despite the numerous benefits, some individuals are unable to tolerate dairy due to lactose intolerance or cow's milk protein allergy (Chen *et al.*, 2021). In such cases, lactose-free milk, fermented dairy, or fortified plant-based alternatives can be used. While saturated fat in whole dairy products was once considered a risk factor for cardiovascular disease, emerging evidence suggests that full-fat dairy may not significantly increase cardiometabolic risk when consumed as part of a balanced diet (Feng *et al.*, 2022). However, safety concerns such as microbial contamination, antibiotic residues, and aflatoxin M1 remain major public health challenges, particularly in regions with weak food safety systems, highlighting the need for proper pasteurization and hygiene standards (Jang *et al.*, 2024).

#### **2.4 Hygiene and food-safety challenges in dairy products**

Dairy products are an important part of the global diet, but they present significant hygiene and food safety challenges due to their rich nutrient composition, which supports microbial growth (Moineau-jean *et al.*, 2017). Yoghurt in particular, is a widely consumed fermented dairy product valued for its nutritional content and beneficial microflora. It is considered a relatively safe dairy product because it is made from pasteurized milk and fermented by starter cultures (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus*), which lower the pH and inhibit many pathogens (Yang *et al.*, 2022). However, yoghurt is still susceptible to microbial contamination during post-pasteurization stages, such as cooling, inoculation,

packaging, and storage, making hygiene a key determinant of its microbiological safety (Karanth *et al.*, 2023). These vulnerabilities mean that dairy processors must maintain stringent hygiene programs and continuous monitoring to protect public health and product quality (Allam *et al.*, 2023; Aslani *et al.*, 2024).

One major challenge is raw milk contamination, which can occur due to poor farm hygiene, mastitis in cows, contaminated water, or unclean milking equipment (Kouam *et al.*, 2024). Even though pasteurization eliminates most pathogens, contamination after heat treatment is a significant risk. Post-pasteurization contamination can arise from equipment surfaces, the processing environment, or workers' hands, allowing pathogens such as *Listeria monocytogenes* to persist and colonize yoghurt production lines (Yang *et al.*, 2022). Cross-contamination from additional ingredients such as fruit purees and flavorings also introduces spoilage microorganisms like yeasts and molds (Karanth *et al.*, 2023).

Several pathogenic microorganisms are of concern in yoghurt production. *Listeria monocytogenes* is particularly important because it can grow at refrigeration temperatures and form biofilms, making it difficult to eradicate from dairy plants (Dabija *et al.*, 2025). *Salmonella* spp., pathogenic *Escherichia coli*, and *Staphylococcus aureus* have also been associated with dairy-borne outbreaks, although pasteurization generally controls these pathogens (Kouam *et al.*, 2024). Additionally, *Bacillus cereus* spores can survive pasteurization, germinate during storage, and produce toxins that compromise food safety (Tirloni *et al.*, 2022). Yoghurt is also vulnerable to spoilage organisms, particularly yeasts and molds, which are the most common contaminants causing gas production, off-flavors, and visible spoilage, even under refrigeration (Karanth *et al.*, 2023). Equipment design and cleaning-in-place systems must be optimized to prevent biofilm formation, which harbors pathogens and spoilage organisms (Dabija *et al.*, 2025). Although

fermentation creates a natural hurdle against pathogens by producing lactic acid and bacteriocins, emerging studies have highlighted the presence of antibiotic resistance genes (ARGs) in some commercial starter and probiotic strains, raising concerns about horizontal gene transfer to gut or environmental bacteria (Zavišić *et al.*, 2023). Screening industrial cultures for transferable ARGs is now recommended as part of a modern food safety management system (EFSA, 2023). Maintaining cold-chain integrity is critical, as refrigeration slows microbial growth but does not completely prevent the proliferation of psychrotrophic pathogens such as *L. monocytogenes* or spoilage yeasts (Yang *et al.*, 2022). Risk-based monitoring programs, including routine environmental swabbing for *Listeria* spp. and microbial testing of ingredients, have been shown to significantly reduce contamination events (Dabija *et al.*, 2025).

## **2.5 Key points of failure in the dairy-to-yogurt chain**

**2.5.1. Raw-milk quality and on-farm practices.** Milk enters the plant with a microbial load determined by animal health (mastitis), milking hygiene, and antibiotic use. Substandard on-farm practices increase initial contamination and complicate later controls; antimicrobial residues and ARGs (antibiotic resistance genes) in milk are also reported concerns. Effective pre-harvest control including udder health management, milking hygiene and judicious antibiotic use reduces downstream risk. (Pires *et al.*, 2024; Sievers *et al.*, 2024).

**2.5.2. Thermal processing and validation.** Pasteurization reduces pathogen load when properly applied, but validation and routine verification are essential. Deviations in time/temperature or equipment faults can leave surviving organisms; moreover, pasteurization does not protect against PPC. (Aslani *et al.*, 2024).

**2.5.3. Post-process contamination (PPC).** The filling, flavoring, and cooling stages are frequent points of contamination. Pathogens introduced after thermal death steps can survive in acidic yogurts long enough to cause illness, particularly if packaging or filling operations are unhygienic or damaged. Outbreak investigations repeatedly identify PPC as the root cause of many dairy recalls. (Gonzales-Barron *et al.*, 2024; Belias *et al.*, 2024).

**2.5.4. Biofilms and environmental reservoirs.** Persistent biofilms in processing lines, drains, gaskets and other niches protect microbes from sanitisers and are a major factor in repeated contamination events. *Listeria* in particular is adept at surviving in environmental biofilms, making thorough Clean-In-Place (CIP) and hygiene monitoring essential. (Pracser *et al.*, 2024; Ban-Cucerzan *et al.*, 2025).

**2.5.5. Cold-chain weaknesses and temperature abuse.** Although low pH and refrigeration together reduce growth of many pathogens, psychrotrophs and acid-tolerant strains can persist or slowly grow if temperatures rise during transport or retail shelving. Breaks in cold chain reduce shelf life and allow growth of organisms like *Listeria*. (Belias *et al.*, 2024).

**2.5.6. Informal/small-scale production sectors.** In many low- and middle-income settings, small-scale and street producers lack HACCP, robust sanitation or refrigeration, leading to higher contamination risk. Targeted education and feasible low-cost interventions are needed to lower exposure in these contexts. (Allam *et al.*, 2023).

## **2.6 Routes and Risk Factors of Contamination**

Microbial contamination of yoghurt can be traced to several critical points along the production chain:

**2.6.1 Raw milk quality:** Raw milk with high microbial loads increases the risk of contamination even after pasteurization. Mastitic milk often contains high levels of pathogens such as *S. aureus*, which may survive if heat treatment is suboptimal (Omore *et al.*, 2021).

**2.6.2 Inadequate pasteurization:** If time–temperature combinations are not carefully monitored, heat-resistant bacteria or spores may survive. This risk is higher in small-scale settings where equipment may not be properly calibrated (Karanth *et al.*, 2023).

**2.6.3 Post-pasteurization contamination:** The highest risk period is after pasteurization, during cooling, inoculation with starter cultures, and packaging. Contaminated utensils, worker hands, or air can introduce spoilage organisms such as yeasts and moulds (Karanth *et al.*, 2023).

**2.6.4 Biofilms on equipment:** Persistent contamination can arise from biofilms formed inside pipes, valves, and fillers. These biofilms protect bacteria against cleaning and disinfection, allowing them to reseed new batches (Asfaw *et al.*, 2023).

**2.6.5 Improper storage and transport:** Inconsistent refrigeration during distribution allows psychrotrophic bacteria and yeasts to proliferate. Even under refrigeration, some spoilage organisms grow slowly, eventually causing visible defects (Buehler *et al.*, 2018).

**2.6.6 Packaging and retail handling:** Reuse of containers, open scooping, and cracked packaging create additional contamination routes. Mould contamination is often higher in yoghurt sold in poorly sealed sachets (Li *et al.*, 1998).

#### **2.6.1.2 Common Pathogens in Milk and Yoghurt**

The major bacterial pathogens associated with milk and yoghurt include *Listeria monocytogenes*, *Salmonella* spp., pathogenic *Escherichia coli* (e.g., EHEC), *Campylobacter* spp., *Staphylococcus*

*aureus*, and spore-forming *Bacillus* spp. (Keba *et al.*, 2020; Asfaw *et al.*, 2023). These organisms can enter the production chain via infected animals, contaminated equipment, inadequate pasteurization, or during post-pasteurization handling (Delia, 2020). Studies consistently report their prevalence in raw milk and occasionally in finished dairy products, especially when hygiene controls are insufficient (Keba *et al.*, 2020).

### **2.6.1.3 Pathogenic Bacteria**

Pathogenic bacteria pose a more serious threat because they may cause foodborne disease outbreaks. *Staphylococcus aureus* is often detected in contaminated yoghurt, and some isolates produce heat-stable enterotoxins responsible for staphylococcal food poisoning (Asfaw *et al.*, 2023). *Salmonella* spp. has been detected in yoghurt and has been implicated in outbreaks of gastroenteritis linked to contaminated dairy products. *Listeria monocytogenes* is particularly dangerous for pregnant women and immunocompromised individuals, as it can cause listeriosis, which may result in miscarriage or sepsis (Karanth *et al.*, 2023). The emergence of antimicrobial-resistant strains in dairy production adds another dimension of concern. (Asfaw *et al.*, 2023) reported multidrug-resistant *E. coli* and *Klebsiella* in yoghurt and milk contact surfaces, underscoring the need for continuous surveillance and antimicrobial stewardship in dairy value chains.

### **2.6.1.4 Microbial Contamination of Yoghurt**

Yoghurt is produced worldwide and valued for its digestibility, protein content, and probiotic potential. The fermentation process lowers pH to approximately 4.5, creating an acidic environment that inhibits the growth of many non-acid-tolerant bacteria (Qu *et al.*, 2024). Nonetheless, yoghurt is not sterile, and it is often consumed without further heat treatment, meaning that any microorganisms present after production may directly affect consumers.

Moreover, the rise of artisanal and small-scale yoghurt production in developing countries has increased the risk of contamination due to inconsistent hygiene standards, poor infrastructure, and limited cold-chain facilities (Omore *et al.*, 2021).

Several studies report that microbial contamination remains a leading cause of yoghurt spoilage and, in some cases, foodborne illness (Karanth *et al.*, 2023). Because of its nutritional importance and popularity among vulnerable groups including children and the elderly yoghurt safety is a major public-health concern. Understanding the spectrum of microorganisms found in contaminated yoghurt and their possible routes of entry provides a foundation for risk assessment and for implementing targeted interventions to reduce contamination. Spoilage yeasts, molds and psychrotrophic bacteria reduce shelf life and sensory quality, and their presence often indicates sanitation failures or temperature abuse. Biofilms formed by spoilage organisms can protect other microbes and seed recurrent contamination events. (Pracser *et al.*, 2024).

#### **2.6.1.5 Opportunistic Spoilage Microorganisms**

Yeasts, moulds, and coliforms are often detected in yoghurt as indicators of poor hygiene or post-processing contamination (Moh *et al.*, 2017). Although not always pathogenic, their presence may signal the potential co-presence of pathogens and can reduce shelf life or cause spoilage defects.

#### **Survival and Adaptation in Yoghurt Environment**

The yoghurt matrix is acidic (pH 4.0–4.5) and refrigerated, which suppresses many pathogens. However, *L. monocytogenes* and spore-formers exhibit notable acid and cold tolerance, allowing survival during storage (Shamloo *et al.*, 2019). Stress-adapted *Salmonella* and *E. coli* can better withstand acidic conditions, potentially leading to persistence (Popa *et al.*, 2021). Lactic acid

bacteria (LAB) in yoghurt produce organic acids, hydrogen peroxide, and bacteriocins that antagonize pathogen growth. (Wiktorczyk-Kapischke *et al.*, 2024) reported that *Lactobacillus rhamnosus* and *L. paracasei* significantly reduced *Listeria*, *E. coli*, and *Salmonella* counts in co-culture systems, indicating that starter cultures play a protective role when fermentation is well-controlled.

## **2.7 Common Microbial Contaminants of Yoghurt**

### **2.7.1 Yeasts and Moulds**

Yeasts and moulds represent significant spoilage microorganism in yoghurt and other fermented dairy products due to their ability to survive under harsh environmental condition. Unlike many bacteria, yeasts possess remarkable tolerance to low pH, high sugar concentration and refrigeration temperature allowing them to proliferate even after pasteurization and during cold storage (Singh *et al.*, 2022). Their presence in yoghurt is primarily attributed to post-processing contamination, often resulting from inadequate sanitation of equipment, airborne spores, or improper packaging. Once present, yeasts metabolize residual lactose and other fermentable sugars to produce ethanol and carbon dioxide, leading to visible spoilage defects such as gas formation, swelling or bursting of yoghurt cups, off-flavours and separation (Ogunbanwo *et al.*, 2023). These organoleptic changes not only reduce the aesthetic quality of yoghurt but also limit its shelf life and consumer acceptability.

In Nigeria, several studies have highlighted the prevalence of yeast and mould contamination in both locally and industrially produced yoghurts. According to (Adebayo *et al.*, 2023), more than 60% of yoghurt samples collected from retail outlets in southwestern Nigeria contained yeast counts ranging from  $10^4$  to  $10^6$  CFU/mL, values that far exceed the microbiological standards recommended by the International Commission on Microbiological Specifications for Foods

(ICMSF). Similarly, research by (Oladipo and Akindele 2022) reported frequent isolation of *Candida*, *Saccharomyces*, and *Rhodotorula* species in Nigerian yoghurts, indicating poor hygiene and cross-contamination during processing and handling. These findings suggest that yeast contamination remains a critical issue within Nigeria's dairy value chain, particularly among small-scale producers with limited access to modern sanitation and quality control technologies. Moulds, on the other hand, pose an additional spoilage and safety concern. Although less common than yeasts in refrigerated yoghurt, mould spores can survive pasteurization and remain dormant until conditions become favorable, such as exposure to air or prolonged storage. Common mould genera associated with yoghurt spoilage include *Penicillium*, *Aspergillus*, and *Mucor* species, which can form visible surface colonies, discoloration, and musty odours (Adepoju *et al.*, 2024). Some moulds are capable of producing mycotoxins, which are harmful secondary metabolites that may pose potential health risks to consumers if contaminated products are ingested. The presence of these fungi often indicates deficiencies in packaging integrity, storage environment, or post-fermentation hygiene practices (Kumari *et al.*, 2021).

Preventing yeast and mould spoilage in yoghurt production requires the implementation of stringent hygienic measures and monitoring programs. The adoption of good manufacturing practices (GMP), regular environmental microbiological testing, and effective cleaning-in-place (CIP) systems are crucial for maintaining product quality and safety. Additionally, the use of protective cultures, such as specific strains of *Lactobacillus* and *Bifidobacterium*, has been shown to inhibit yeast and mould growth through competitive exclusion and production of natural antifungal compounds (Chandran *et al.*, 2023). Furthermore, maintaining the cold chain during storage and distribution is essential to suppress microbial proliferation and extend shelf

life. Continuous training of local dairy producers in Nigeria on hygienic handling and microbial risk management can significantly reduce contamination rates and enhance consumer confidence in locally produced yoghurts.

### **2.7.2 Coliforms and Gram-Negative Bacteria**

Coliforms are used as hygiene indicators in dairy quality control. Their presence indicates inadequate sanitation or post-pasteurization contamination. Several studies from Africa and Asia have reported coliform counts in yoghurt above international limits, highlighting poor hygienic conditions during production (Nkongho *et al.*, 2018). The presence of *E. coli* specifically suggests faecal contamination and raises the possibility that more dangerous pathogens may also be present. Other Gram-negative bacteria such as *Klebsiella* spp., *Pseudomonas* spp., and *Enterobacter* spp. have been isolated from contaminated yoghurt samples. These bacteria may contribute to off-odors, proteolysis, and reduction in shelf life, even if they are not always pathogenic (Asfaw *et al.*, 2023).

#### *Listeria monocytogenes*

*L. monocytogenes* is a psychrotrophic, Gram-positive pathogen capable of surviving refrigeration temperatures and acidic conditions (Shamloo *et al.*, 2019). They form biofilms on equipment, and persisting in processing environments. *L. monocytogenes* is a major concern for ready-to-eat dairy products because contamination after heat treatment can lead to severe disease in vulnerable populations. Risk assessment studies and outbreak investigations repeatedly highlight *Listeria* as a top hazard in dairy. (Gonzales-Barron *et al.*, 2024; Belias *et al.*, 2024). Studies have shown that it can persist in dairy processing facilities and occasionally contaminate finished products (Yang *et al.*, 2022). Listeriosis is severe in immunocompromised individuals, pregnant

women, and neonates, causing septicemia, meningitis, and fetal loss (EFSA, 2020). Its persistence in biofilms and resistance to cleaning chemicals make it a high-priority pathogen in yoghurt safety monitoring.

#### *Salmonella* spp.

edAlthough pasteurization effectively destroys *Salmonella*, post-pasteurization contamination or survival in inadequately fermented products may occur (Savran *et al.*, 2018) modeled *Salmonella* enteritidis survival in yoghurt during storage and found that pathogen levels declined but remained detectable over time. *Salmonella* spp. and pathogenic *Escherichia coli* while proper pasteurization kills these organisms, use of unpasteurized milk or post-pasteurization contamination (PPC) can result in their presence in finished products (Aslani *et al.*, 2024). *Salmonella* infections cause gastroenteritis, fever, and occasionally systemic illness (Popa *et al.*, 2021). Several outbreaks linked to yoghurt have been documented in developing regions where production hygiene is suboptimal (Fernández *et al.*, 2015).

#### *Staphylococcus aureus*

*Staphylococcus aureus* is common in raw milk, introduced from infected udders or human handlers (Gebremedhin *et al.*, 2022). They can contaminate milk through mastitic animals or poor hygiene among handlers; *S. aureus* is noteworthy not just for the organism but for its heat-stable enterotoxins that cause food poisoning even if organisms are reduced by processing (Allam *et al.*, 2023). Although yoghurt's acidity inhibits its growth, heat-stable enterotoxins produced before or during fermentation can remain active (Sasidharan *et al.*, 2011). Staphylococcal food poisoning is characterized by rapid onset of vomiting and abdominal cramps, posing a risk even in the absence of viable bacteria.

### Pathogenic *Escherichia coli*

Pathogenic *E. coli* variants, including EHEC and EPEC, have been isolated from yoghurt and fermented milk in several surveillance studies (Cutrim *et al.*, 2016). Their ability to survive depends on initial contamination level, acid adaptation, and storage conditions. EHEC infections can result in hemorrhagic colitis and hemolytic uremic syndrome, making their presence in yoghurt a serious public health concern (Bahgat *et al.*, 2024).

### *Bacillus cereus*

A spore-forming bacterium widely distributed in soil and dairy environments, *B. cereus* spores may survive pasteurization and germinate if yoghurt is stored improperly (Tirloni *et al.*, 2022). It produces diarrhoeal and emetic toxins, causing self-limiting food poisoning. Its prevalence in dairy is influenced by equipment sanitation and milk storage temperature (Jessberger *et al* 2020)

### *Brucella* spp.

In areas where yoghurt is made from raw or unpasteurized milk, *Brucella* poses a zoonotic risk. Fermentation does not reliably eliminate the organism, and brucellosis outbreaks have been linked to unpasteurized dairy consumption (Islam *et al.*, 2023). Chronic brucellosis can result in arthritis, endocarditis, and long-term disability.

## **2.8. Public-Health Implications**

The presence of pathogens in yoghurt has direct implications for public health. Outbreaks of salmonellosis, staphylococcal food poisoning, and colibacillosis have been linked to contaminated dairy products worldwide (Asfaw *et al.*, 2023). Vulnerable populations such as infants, pregnant women, the elderly, and immunocompromised patients are at particular risk of

severe complications (Karanth *et al.*, 2023). Even when contaminants are not pathogenic, spoilage leads to significant economic losses due to product recalls, reduced consumer confidence, and waste. In resource-limited settings, spoilage represents a major financial loss to small-scale producers and retailers (Omore *et al.*, 2021).

Yoghurt is one of the most widely consumed fermented dairy products globally, valued for its nutrient density and probiotic benefits. It provides protein, calcium, vitamins, and bioactive compounds that support health (Keba *et al.*, 2020). However, yoghurt can also serve as a vehicle for microbial, chemical, and physical hazards that pose significant public-health concerns if controls fail during production or storage (Sebastianski *et al.*, 2022). Hazards may be introduced through raw milk contamination, inadequate pasteurization, post-processing contamination, and cold chain failure (Delia, 2020). The implications include foodborne illness, hospitalization, antimicrobial resistance dissemination, allergic reactions, and in severe cases, mortality among vulnerable populations (Koski, 2022).

AMR in dairy-associated pathogens complicates treatment, resulting in prolonged illness, increased hospitalization costs, and higher mortality risk (Veloo *et al.*, 2025). Even commensal bacteria in yoghurt that harbor transferable ARGs can act as reservoirs, contributing to the broader dissemination of resistance within the community (Sievers, 2024). Integrated One Health surveillance linking farm, food, and clinical data is therefore essential to mitigate the risk (Pires *et al.*, 2024).

### **Probiotic and Culture Safety**

Yoghurt typically contains starter cultures (*Lactobacillus*, *Streptococcus thermophilus*) and sometimes probiotic strains (*Bifidobacterium*, *Lacticaseibacillus*). For most consumers, these are

safe and confer health benefits, but rare cases of probiotic-associated bacteremia or fungemia have been reported in severely immunocompromised patients and those with central venous catheters (Tom *et al.*, 2021). Moreover, certain probiotic strains may carry ARGs, underscoring the need for strain-specific safety assessment and regulatory oversight (Nyanzi, 2021).

## Chemical Hazards and Allergens

Dairy products may contain chemical hazards such as antibiotic residues, pesticide residues, heavy metals, and mycotoxins if proper controls are not implemented (Codex Alimentarius, 2004). Exceeding maximum residue limits (MRLs) may result in allergic reactions, toxic effects, or disruption of gut microbiota (Delia, 2020). Additionally, milk proteins are a leading cause of food allergies in children, requiring accurate labeling and prevention of cross-contact during manufacturing (FAO/WHO, 2009).

### **2.8.1 Outbreaks and Clinical Impact**

Outbreak investigations have linked both pasteurized and unpasteurized dairy products to significant foodborne disease events. Unpasteurized milk products are more commonly implicated, but pasteurized yoghurt has also caused outbreaks when contamination occurred after processing (Sebastianski *et al.*, 2022). Clinical outcomes range from self-limiting gastroenteritis to severe invasive infections, including septicemia, meningitis, and pregnancy-associated listeriosis leading to fetal loss (Koski, 2022). Quantitative microbial risk assessment studies indicate that the absolute risk from properly processed and refrigerated yoghurt is low, but risk increases substantially with raw milk use or temperature abuse (Yang *et al.*, 2022).

## **Small-Scale and Informal Sector Risks**

In many developing regions, yoghurt production occurs at small or informal scales where pasteurization, refrigeration, and hygiene practices are limited (Delia, 2020). Such systems account for a disproportionate share of microbiological risk, highlighting the need for affordable, scalable interventions such as farmer training, community-level pasteurization systems, and improved cold-chain infrastructure (FAO/WHO Codex, 2009).

## **Vulnerable Populations and Burden of Disease**

Infants, pregnant women, older adults, and immunocompromised persons are at higher risk of severe outcomes from dairy-borne infections. For example, *L. monocytogenes* infections during pregnancy can result in neonatal sepsis, meningitis, or stillbirth (Sebastianski *et al.*, 2022). The economic burden of dairy-borne illness includes direct medical costs, productivity losses, and costs associated with product recalls and trade restrictions (Koski, 2022).

## **2.9 Antimicrobial resistance (AMR) and dairy**

AMR is both a public-health and a production issue. Antibiotics used to treat mastitis and other animal infections select for resistant bacteria and for ARGs that can be present in milk and farm waste streams. Recent systematic reviews find ARGs in milk and dairy farm environments globally, raising concerns about potential transmission routes and the need for stewardship and surveillance. Though the direct risk of ARG transfer to consumers via pasteurized milk remains under investigation, the presence of AMR in the dairy supply chain complicates treatment of infections and signals on-farm management problems. (Sievers *et al.*, 2024; Collis *et al.*, 2024).

## **Pathways of AMR Transmission**

Antimicrobial use in dairy cattle selects for resistant bacteria and antibiotic-resistance genes (ARGs) in the farm environment (Pires *et al.*, 2024). These resistant organisms may contaminate milk, survive processing environments, and potentially colonize humans. Systematic reviews have documented the presence of ARGs in *E. coli* and *Staphylococcus aureus* isolated from raw milk and dairy products (Sievers, 2024).

### **2.9.1 Surveillance, outbreaks and lessons learned**

Recent recalls and outbreak investigations (e.g., multi-state *Listeria* incidents linked to dairy products) illustrate how environmental contamination and PPC can have large public-health and economic impacts. Enhanced surveillance, rapid traceback (supported by sequencing), and proactive environmental monitoring are critical to detect hazards before they affect consumers. Processors must treat outbreaks as system failures and use root-cause analysis to close gaps permanently. (News reports; Belias *et al.*, 2024).

Yogurt production benefits from fermentation and acidity, but these are not fail-safe barriers. The main hygiene and food-safety challenges arise from raw-milk quality, post-process contamination, biofilm persistence, cold-chain lapses, and AMR at the farm level. Effective control requires a multi-barrier approach: upstream herd and milk quality management; validated thermal processes; strict hygienic design and sanitation; robust HACCP/FSMS; cold-chain monitoring; and modern surveillance tools including molecular diagnostics and WGS for outbreak response. Special emphasis should be placed on training and pragmatic interventions for small-scale producers, and on research validating novel interventions under industrial conditions. (Aslani *et al.*, 2024; Sievers *et al.*, 2024; Pracser *et al.*, 2024).

## 2.10 Control and Mitigation Strategies

### Industrial Measures

Large-scale dairy producers employ stringent quality-control systems to minimize microbial contamination and ensure product safety. These systems encompass microbiological testing of raw milk, validated pasteurization protocols, hygienic equipment design, and the implementation of cleaning-in-place (CIP) systems to maintain sanitary processing environments (Karanth *et al.*, 2023). Environmental monitoring, particularly for yeasts and coliforms, is essential to detect contamination sources early and maintain product integrity throughout production and packaging.

One critical approach to enhancing yoghurt safety involves the use of protective cultures and biopreservatives. Starter cultures that produce bacteriocins, such as nisin, play a significant role in inhibiting spoilage microorganisms and pathogenic bacteria, thereby providing a natural antimicrobial barrier without relying on synthetic additives (Buehler *et al.*, 2018). Maintaining cold-chain integrity is equally important, as yoghurt must remain under refrigeration from production through distribution to prevent microbial proliferation. The use of continuous temperature monitoring devices is strongly recommended to ensure that yoghurt products are stored and transported below 4°C (Buehler *et al.*, 2018).

Despite advancements in production and preservation, several gaps persist. Surveillance for microbial pathogens in yoghurt is limited, particularly in developing countries, where systematic testing could help identify contamination hotspots and improve public health outcomes. Emerging technologies, such as metagenomic sequencing and polymerase chain reaction (PCR)-based detection methods, offer rapid and sensitive tools for early contamination warning (Qu *et al.*, 2024). Additionally, continuous monitoring of antimicrobial resistance (AMR) among dairy

isolates is essential, as resistant pathogens pose growing challenges to food safety and clinical treatment (Asfaw *et al.*, 2023).

### **2.10.1 Control Strategies**

Effective control in yoghurt relies on:

**Pasteurization of Milk:** High-temperature short-time (HTST) treatment to eliminate vegetative pathogens.

**Good Manufacturing Practices:** Strict cleaning, disinfection, and personnel hygiene to avoid re-contamination (Aladhadh *et al.*, 2023).

**Rapid Fermentation:** Using well-characterized starter cultures to achieve pH  $\leq 4.5$  quickly, inhibiting pathogens.

**Cold Chain Maintenance:** Continuous refrigeration during storage and transport to suppress psychrotrophic growth.

**Protective Cultures:** Incorporation of bacteriocin-producing LAB to enhance microbiological safety (Wiktorczyk-Kapischke *et al.*, 2024).

**Routine Testing & HACCP Verification:** Microbial monitoring and hazard analysis to ensure compliance with food safety standards. A hazard analysis framework with clearly identified CCPs (pasteurization, filling, cold storage) and documented verification/audit procedures reduces risk. HACCP must be actively managed with staff training and external audits. (Aslani *et al.*, 2024).

**Upstream controls at farm level:** Improvements in udder health management, hygiene during milking, testing for antibiotic residues, and antimicrobial stewardship reduce initial contamination and AMR selection pressure (Pires *et al.*, 2024). Regular supplier audits and raw-milk testing should be standard for processors.

**Validated pasteurization and process monitoring:** Ensure time-temperature validation, redundant instrumentation, and real-time monitoring with corrective action plans. Pasteurization must be complemented by rigorous PPC prevention. (Aslani *et al.*, 2024).

**Hygienic design and robust sanitation (CIP):** Equipment designed to minimise niches, effective CIP protocols, and rotation of sanitizers help control biofilms. Environmental monitoring programs including routine swabbing of drains, gaskets and filling nozzles detect persistence early. (Pracser *et al.*, 2024; Ban-Cucerzan *et al.*, 2025).

**Cold-chain management and temperature logging:** Continuous temperature monitoring from plant through retail, with alarms and traceability, lowers the chance of temperature abuse. Consumer education on storage and “use-by” interpretation also helps. (Belias *et al.*, 2024).

**Advanced detection and rapid response:** Integrating rapid molecular tools (qPCR, sequencing) alongside cultural methods improves detection sensitivity and speed for pathogens and ARGs. Whole-genome sequencing assists outbreak tracing and source attribution. (Gonzales-Barron *et al.*, 2024).

**Novel interventions and research needs:** Emerging approaches protect cultures, bacteriophages, non-thermal technologies (e.g., PEF), and improved packaging show promise but require validation for efficacy, sensory acceptability and regulatory approval. Field studies in

real production settings (not only lab models) are needed to confirm benefit and operational feasibility. (Dabija *et al.*, 2025; Gonzales-Barron *et al.*, 2024).

### **2.10.2 Challenges and Future Perspectives**

Research gaps include improving detection of low-level contamination, understanding stress adaptation mechanisms, and developing more predictive risk models for yoghurt safety. Climate change and unreliable cold chains in low-resource settings may exacerbate risks, necessitating context-specific interventions. Consumer education on safe storage and avoidance of unpasteurized dairy remains crucial to prevent foodborne disease outbreaks.

### **2.10.3 Regulatory Frameworks and Preventive Controls**

The Codex Alimentarius “Code of Hygienic Practice for Milk and Milk Products” outlines hazard analysis, good hygienic practices, and pasteurization standards to prevent contamination (Codex Alimentarius, 2004). National regulations complement these by setting microbiological criteria, residue limits, and labelling requirements (FAO/WHO, 2009). Core preventive measures include on-farm hygiene, validated pasteurization, environmental monitoring, maintenance of cold chain, batch testing, and effective traceability and recall systems (Delia, 2020).

## **CHAPTER THREE**

### **METHODOLOGY**

#### **3.1 Sample Collection**

A total of eight (8) samples of diary products consisting of hollandia, nana, ceda, superyogo, nono milk, kidoromo, wara, and maishanu were collected from each of the three locations. The nature of the diary products collected were locally and branded processed yoghurts. Each sample was collected using sterile gloves and stored in sterile polythene bags to avoid cross-contamination.

#### **3.2 Sterilization of materials**

Materials such as petri-dish, pipette, glass container (conical flask, round bottom flasks) and bottles were washed, drained and dried. They were wrapped with aluminum foil and sterilized in a hot-air oven at 160 °C for an hour. They were allowed to cool after sterilization before usage. An aseptic working environment was achieved with the use of Bunsen burner flame and disinfection of work surface area with alcohol.

#### **3.3 Preparation of media**

The culture media used were prepared according to the manufacturer's instructions. The media used were Nutrient Agar, MacConkey Agar and Salmonella Shigella Agar.

##### **3.3.1 Nutrient agar**

28g of Nutrient Agar (NA) powder was dissolved in 1 litre of distilled water in a conical flask with cotton wool and aluminium foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile petri dish in the laminar flow.

### **3.3.2 Preparation of Salmonella Shigella agar**

63g of Salmonella Shigella Agar (SSA) powder was dissolved in 1 litre distilled water in a conical flask covered with cotton wool and aluminium foil paper. It was mixed thoroughly and boiled with frequent agitation to dissolve the medium completely. The medium was cooled to 50c and then dispensed aseptically into sterile petri dish in the laminar flow.

### **3.3.3 Preparation of Eosin Methylene Blue**

36g of Eosin Methylene Blue Agar (EMB) powder was dissolved in 1 litre of distilled water in a conical flask covered with cotton wool and aluminium foil paper. It was mixed thoroughly and sterilized by autoclaving at 121c for 15 minutes. The medium was cooled to 45-50c and then dispensed aseptically into sterile petri dish in a laminar flow.

### **3.3.4 MacConkey Agar**

51.55g of MacConkey Agar (MCA) powder was dissolved in 1 litre of distilled water in a conical flask covered with cotton wool and aluminium foil paper. It was mixed thoroughly and sterilized by autoclaving at 121c for 15 minutes. The medium was cooled to 45-50c and then dispensed aseptically into sterile petri dish in the laminar flow.

### **3.3.5 Mueller Hinton Agar**

38 grams of Muller Hinton Agar (MHA) powder was dissolved in 1 litre of distilled water in a conical flask covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled for 45°C – 50°C and then dispensed aseptically into sterile petri dish and wait for the medium to solidify. The agar was prepared in a clean environment to prevent any contamination.

### **3.4 Isolation and characterization of bacteria**

1ml of sample was measured and diluted in 9ml of sterile distilled water. It was allowed to stand for 30 minute (Rodriguez-Sanchez *et al.*, 2023; Haghshenas *et al.*, 2017). The aliquot was then transferred aseptically to sterile petri dish plates. The prepared agar (for bacteria growth) was poured in aseptically and incubated at 37°C for 24 hours (Dehghani Champiri *et al.*, 2024) After successful growth of microorganisms the colonies were counted with a colony counter and the results per dilution count were recorded. The number of colony forming unit per millimeter was calculated with the formula= Cfu/ml.

### **3.5 Gram Staining**

Smears of the bacterial isolates were prepared and heat fixed on clean grease free slides. The smears were stained for one minute with crystal violet. This was washed out with distilled water. The slides were flooded with dilute Grams iodine solution for one minute. This was washed off with distilled water and the smears were decolorized with 95% alcohol for 30 seconds and rinsed with distilled water. The smears were then counter stained with safranin solution for one minute. Finally, the slides were washed off with distilled water, air dried and observed under oil immersion objectives.

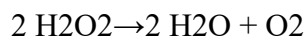
### **3.6 Morphological Analysis**

Each colony morphology e.g., shape, margin, elevation, consistency, color, transparency was determined.

### **3.7 Biochemical test**

#### **3.7.1 Catalase test**

This test was to detect the presence or absence of catalase enzyme. The catalase enzyme catalyses the breakdown of hydrogen peroxide to release free oxygen gas and the formation of water. A few drops of freshly prepared 3% hydrogen peroxide were added onto the bacterial isolates smeared on a slide. The production of gas bubble indicated catalase enzyme positive.



#### **3.7.2 Citrate Utilization Test**

This test was based on the ability of some organisms to utilize citrate as a sole source of carbon. This was carried out by inoculating the test organism in test tube containing Simon's citrate medium and this was incubated at 37°C for 24 - 48 hours. The development of deep blue colour after incubation indicates a positive result.

#### **3.7.3 Urease Test**

The urease test was used to determine the ability of an organism to split urea in the presence of the enzyme urease. The bacterial isolates were inoculated into slants of urea medium and incubated at 37°C for 24-48 hours. Urease positive cultures produced a red-pink colour due to changes in the colour of the indicator



#### **3.7.4 Hydrogen Sulphide (H<sub>2</sub>S) Test**

Hydrogen sulphide production can be detected by incorporating a heavy metal salt containing (Fe<sup>2+</sup>) or lead (Pb<sup>2+</sup>) ion as H<sub>2</sub>S indicator to a nutrient culture medium containing cysteine and

sodium thiosulfate as the sulphur substrates. Hydrogen sulphide, a colourless gas, when produced reacts with sulphur metal salt (ferrous sulphate) forming a visible insoluble black sulphide precipitate.

### **3.7.5 Indole Test**

Indole test was performed to determine the ability of the organism to split tryptophan molecule into indole. This test was performed to help differentiate species of the family enterobacteriaceae. Kovac's reagent which contains hydrochloric acid, dimethylaminobenzaldehyde and amyl alcohol was used. Inoculate broth with the test organism and incubate for 18 – 24 hours at 37°C. Add 5ml of Kovac's reagent down the inner wall of the tube. Development of bright red colour at the interface of the reagent and the broth within seconds after adding the reagent is indicative of the presence of indole and is a positive test while absence is negative.

### **3.7.6 Coagulase test**

The coagulase test was used to differentiate strains of *Staphylococcus aureus* from *S. epidermidis* and other coagulase-negative species. *S. aureus* are usually capable of coagulating EDTA-treated plasma in the tube test and would produce clumps of cells in the slide test. A small drop of plasma was placed on a slide and mixed with a test isolate and observed for clumping within 10 seconds which indicate a positive test. However, negative tests were further subjected to the test tube for confirmation

In the tube method, 0.5ml of plasma is put in a tube and inoculated with the bacterial cultures and then incubated. At intervals over the next 4 hours, the culture is observed for evidence of a clot. Any clot formation is a positive result. If no clot was observed by the end of 4 hours, then

the test will be continued with an overnight incubation at room temperature and a final observation at 24 hours. If after 24 hours, no clot was observed, the organism will be coagulated negative.

### **3.7.7 Sugar Fermentation Test**

Each of the isolates was tested for its ability to ferment a given sugar with the production of acid and gas or acid only. The growth medium comprised of peptone water, sugar (1%) and the indicator (bromocresol purple). The mixture was dispensed into test tubes and sterilized by autoclaving at 121°C for 15 minutes. After sterilizing, tubes were allowed to cool and then inoculated with the isolates and incubated at 37°C for 24hrs. Acid and gas production or acid only were observed after about 24 hours of incubation. Acid production was indicated by the change of the medium from purple to yellow colour indicated a positive test. Sugars used are: lactose, sucrose, glucose, fructose, maltose, starch and sorbitol.

### **3.8 Antibiotics Susceptibility Test**

Antibiotics susceptibility testing is a laboratory procedure performed by medical technologists (clinical laboratory scientists) to identify which antimicrobial regimen is effective for individual patients. On a larger scale, the testing aids in evaluating treatment services provided by hospitals, clinics, and national programs to control and prevent infectious diseases (Lagier *et al.*, 2015). Test organisms were subjected to antibiotics sensitivity test using the Kirby Bauer disc diffusion on prepared media. Ten (10) different commercial antibiotic discs were used. The antibiotic discs was carefully and firmly placed on the inoculated plates using a sterile pair of forceps. The plates was inverted and incubated for 37°C for 24 hours. The diameter of the zone of inhibition will be measured in millimeters (mm) using a meter rule. The experiments was carried out in triplicates to minimize probability of error.

### **3.9 Determination of pH**

The pH of the samples were determined using a pH meter (Murphy *et al.*, 2023; Liu *et al.*, 2024). The pH electrode was rinsed with distilled water according to the manufacturer's instructions using standard buffer solutions (Standard laboratory Protocol). The pH probe was inserted into the sample, and held for a couple of minutes to achieve a stabilized reading (Chopde *et al.*, 2025).

### **3.10 TITRATABLE ACIDITY**

50ml of sample was pipetted into a clean 250ml of conical flask and 2 drops of phenolphthalein indicator was added. The solution was titrated against the standardized 0.1m NAOH to get a pink end point and the titre value was recorded (Sadler and Murphy, 2010).

## CHAPTER FOUR

### RESULTS

The total heterotrophic bacteria count of dairy samples is shown in Table 4.1. The Bacteria count ranged from  $4.8 \pm 0.26 \times 10^8$  (Hollandia) to  $8.6 \pm 0.36 \times 10^8$  (Maishanu). Table 4.2 shows the total coliform count of dairy samples on macConkey agar which ranged from  $0.8 \pm 0.485 \times 10^7$  (Nana) to  $6.67 \pm 1.53 \times 10^7$  (Maishanu). The total bacteria count on *Salmonella-Shigella* agar from dairy samples shown in Table 4.3 ranged from  $1.13 \pm 0.31 \times 10^4$  (Maishanu) to  $6.87 \pm 0.61 \times 10^4$  (Super yogo). Table 4.4 shows the total bacteria count of dairy samples on Eosin methylene blue (EMB) agar which ranged from  $5.07 \pm 0.64 \times 10^3$  (Nono) to  $7.0 \pm 0.4 \times 10^3$  (Maishanu). The result for the cultural, morphological and biochemical characteristics of the bacterial isolates on dairy products sold in Benin city is shown in Table 4.5. The isolated bacterial species from the samples include *Escherichia coli*<sup>1</sup>, *Escherichia coli*<sup>2</sup>, *Salmonella* sp.<sup>1</sup>, *Salmonella* sp.<sup>2</sup>, *Enterobacter aerogenes* and *Serratia marcescens*. *Escherichia coli*<sup>1</sup>, *Escherichia coli*<sup>2</sup>, *Salmonella* sp.<sup>1</sup> and *Salmonella* sp.<sup>2</sup> had the highest frequency of occurrence, each with a percentage of 33.33% while both *Enterobacter aerogenes* and *Serratia marcescens* has the least percentage frequencies of 16.6%.

Table 4.7 shows the result of the antibiotic sensitivity test on Gram-negative bacteria isolate in dairy samples with *Salmonella* sp.<sup>1</sup> having the lowest resistance index of (0.1) and *Enterobacter aerogenes* and *Salmonella* sp.<sup>2</sup> having the highest resistance index of (0.8). Table 4.8 shows the pH values of the dairy samples which ranged from  $3.82 \pm 0.01$  (Hollandia) to  $6.96 \pm 0.03$  (Maishanu). Table 4.9 shows the titratable values which ranged from  $0.17 \pm 0.02$  (Kindoromo) to  $1.86 \pm 0.02$  (Hollandia).

**TABLE 4.1 Total Heterothrophic Bacteria count (CFU/ml) of dairy products sold in Benin city.**

<b>Sample name</b>	<b>Total bacteria count (<math>\times 10^8</math> CFU/ml)</b>
Hollandia	4.80 $\pm$ 0.26
Nana	5.20 $\pm$ 0.16
Cedaa	6.20 $\pm$ 0.36
Super Yogo	5.60 $\pm$ 0.26
Nono	7.20 $\pm$ 0.31
Kindoromo	8.30 $\pm$ 0.36
Wara	7.30 $\pm$ 0.42
Maishanu	8.60 $\pm$ 0.26

**TABLE 4.2 Total Bacteria Counts (CFU/ml) of dairy samples sold in Benin city on MacConkey agar.**

<b>Sample name</b>	<b>Total bacteria count (<math>\times 10^7</math> CFU/ml)</b>
Hollandia	1.10 $\pm$ 0.058
Nana	0.80 $\pm$ 0.483
Cedaa	1.03 $\pm$ 0.410
Super yogo	1.10 $\pm$ 0.000
Nono milk	1.43 $\pm$ 0.180
Kindoromo	4.00 $\pm$ 0.000
Wara	1.63 $\pm$ 0.150
Maishanu	6.67 $\pm$ 1.530

**TABLE 4.3 Total Bacteria Count (CFU/ml) of dairy samples sold in Benin city on *Salmonella Shigella* agar**

<b>Sample name</b>	<b>Total bacteria count (<math>\times 10^4</math> CFU/ml)</b>
Hollandia	5.93 $\pm$ 0.50
Nana	4.73 $\pm$ 0.42
Cedaa	2.40 $\pm$ 0.00
Super yogo	6.87 $\pm$ 0.61
Nono milk	2.33 $\pm$ 0.31
Kindoromo	6.00 $\pm$ 0.00
Wara	2.73 $\pm$ 0.31
Maishanu	1.13 $\pm$ 0.31

**TABLE 4.4 Total Bacteria Counts (CFU/ml) of dairy samples sold in Benin city on Eosin Methylene Blue agar.**

<b>Sample name</b>	<b>Total Bacteria Count</b> <b>(<math>\times 10^3</math> CFU/ml)</b>
Hollandia	NG
Nana	NG
Cedaa	NG
Super yogo	NG
Nono milk	5.07 $\pm$ 0.64
Kindoromo	6.33 $\pm$ 0.42
Wara	5.40 $\pm$ 0.60
Maishanu	7.00 $\pm$ 0.40

**TABLE 4.5 Cultural, morphological and biochemical characteristics of bacteria isolates from dairy products sold in Benin city.**

	1	2	ISOLATES3	4	5	6	
CHARACTERISTICS							
Elevation		Raised	Raised	Raised	Flat	Flat	Flat
Margin		Entire	Entire	Undulate	Filiform	Entire	Entire
Color		Pink	Cream	Off white	Cream	Cream	Cream
Shape		Circular	Circular	Irregular	Rhizoid	Circular	Circular
Size		Small	Small	Large	Large	Small	Small
Transparency		Opaque	Opaque	Opaque	Opaque	Opaque	Translucent
Gram stain		-	-	-	-	-	-
Cell type		Rod	Rod	Rod	Rod	Rod	Rod
Cell arrangement		Chains	Chains	Singles	Cluster	Single	Pair
Urease		-	-	-	+	-	-
Indole		+	+	-	-	-	-
Citrate		-	-	+	-	-	-
Catalase		+	+	+	+	+	+
H <sub>2</sub> S		-	-	-	+	+	+
Coagulase		-	-	-	-	-	-
Lactose		+	+	+	-	-	-
Sucrose		+	+	+	-	-	-
Glucose		+	+	+	+	+	+
Fructose		+	+	+	+	-	-
Maltose		+	+	+	-	+	+
Starch		-	-	-	-	-	-
Sorbitol		+	-	+	-	+	+
Probable organism		<i>Escherichia coli</i> <sup>1</sup>	<i>Escherichia coli</i> <sup>2</sup>	<i>Enterobacter aerogenes</i>	<i>Serratia marcescens</i>	<i>Salmonella</i> sp <sup>1</sup>	<i>Salmonella</i> sp <sup>2</sup>

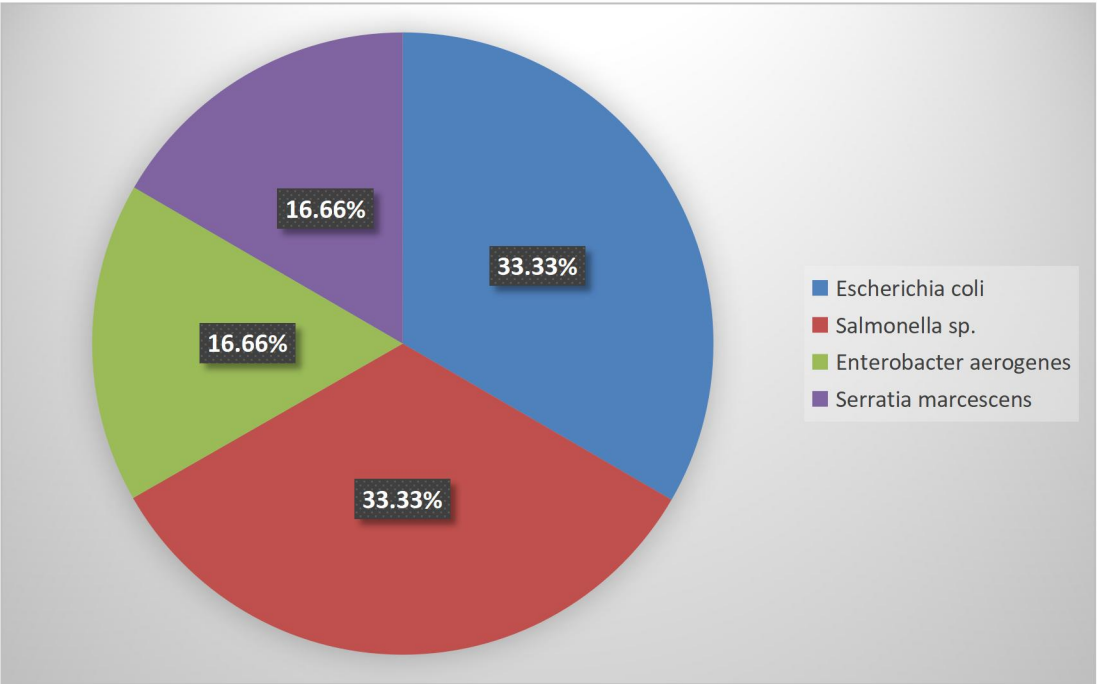


Figure 1: Percentage frequency of occurrence of bacteria isolates from dairy samples sold in Benin city.

**TABLE 4.7: Antibiotics sensitivity pattern of Gram-negative isolates from dairy products**

ISOLATES	SXT	CH	SP	CPX	AM	AU	CN	PEF	OFX	S	R.I
<i>Escherichia coli</i> <sup>1</sup>	6(R)	8(R)	16(I)	16(I)	14(I)	16(I)	8(R)	18(S)	18(S)	6(R)	0.4
<i>Escherichia coli</i> <sup>2</sup>	16(I)	10(R)	18(S)	14(I)	14(I)	10(R)	14(I)	16(I)	20(S)	14(I)	0.2
<i>Enterobacter aerogenes</i>	0(R)	0(R)	10(R)	10(R)	8(R)	10(R)	10(R)	22(S)	22(S)	10(R)	0.8
<i>Serratia marcescens</i>	6(R)	10(R)	14(I)	14(I)	14(I)	16(I)	14(I)	20(S)	10(R)	12(I)	0.3
<i>Salmonella sp</i> <sup>1</sup>	16(I)	14(I)	14(I)	12(I)	10(R)	12(I)	14(I)	20(S)	20(S)	14(I)	0.1
<i>Salmonella sp</i> <sup>2</sup>	8(R)	0(R)	6(R)	8(R)	8(R)	4(R)	4(R)	16(I)	12(I)	10(R)	0.8

KEYS; R.I = Resistance index, Resistant (R) = 0-10mm, Intermediate (I) = 11-16mm, Sensitive (S) = 17mm and above, SXT = Septrin (30µg), CH = Chloaranphenicol (30µg), SP = Sparifloxin (10µg), CPX = Ciprofloxacin (30µg), AM = Amoxacillin (30µg), AU = Augmentin (10µg), CN = Gentamycin (30µg), PEF = Pefloxacin (30µg), OFX = Tarivid (10µg), S = Streptomycin (30µg).

**TABLE 8: pH values of dairy samples sold in Benin city.**

---

<b>Sample name</b>	<b>Ph</b>
<b>Hollandia</b>	3.82±0.01
<b>Nana</b>	3.94±0.04
<b>Cedaa</b>	4.20±0.02
<b>Super yogo</b>	4.19±0.03
<b>Nono milk</b>	3.92±0.03
<b>Kindoromo</b>	6.95±0.01
<b>Wara</b>	3.93±0.05
<b>Maishanu</b>	6.96±0.03

---

**TABLE 4.9: Total Titratable acidity (mg/l) values of dairy samples sold in Benin city.**

<b>Sample name</b>	<b>Tta value</b>
<b>Hollandia</b>	<b>1.86±0.02</b>
	<b>1.78±0.02</b>
<b>Nana</b>	<b>1.43±0.03</b>
	<b>0.67±0.02</b>
<b>Cedaa</b>	<b>1.70±0.03</b>
	<b>0.17±0.02</b>
<b>Super yogo</b>	
	<b>1.72±0.05</b>
<b>Nono milk</b>	<b>0.24±0.02</b>
<b>Kindoromo</b>	
<b>Wara</b>	
<b>Maishanu</b>	

## CHATER FIVE

### DISCUSSIONS

The results of the total heterotrophic bacteria count (THBC), which ranged from  $4.8 \pm 0.26 \times 10^8$  CFU/ml to  $8.6 \pm 0.26 \times 10^8$  CFU/ml, indicate an alarmingly high level of microbial contamination across all samples. These counts far exceed the standard acceptable limit of  $1.0 \times 10^4$  CFU/ml for pasteurized milk and  $1.0 \times 10^5$  CFU/ml for raw milk as suggested by international food safety bodies like the International Commission on Microbiological Specifications for Foods (ICMSF) (ICMSF, 2011). Similarly, a study in Ethiopia found THBC in raw milk to be as high as  $7.9 \times 10^8$  CFU/ml (Teka, 2020). The high counts in products like Maishanu and Kindoromo suggest potential post-processing contamination or the use of raw materials with initial high microbial loads. A study by Salisu *et al.*, (2016) on commercially prepared yoghurt retailed in Sokoto State, Northwestern Nigeria, reported total aerobic plate counts ranging from  $2.66 \times 10^4$  to  $1.13 \times 10^5$  CFU/ml. The higher bacterial counts observed in this study may reflect differences in sampling locations, storage conditions, and the inclusion of traditional, unbranded products. The total coliform count, used as an indicator of fecal contamination and overall sanitary quality, ranged from  $0.8 \pm 0.483$  to  $6.67 \pm 1.53 \times 10^7$  CFU/ml on MacConkey agar. The presence of coliforms in these quantities is a clear breach of food safety standards, which often stipulate zero tolerance for coliforms in pasteurized dairy products. The highest coliform count was found in Maishanu, which could be linked to contaminated processing water or unsterilized containers, a problem highlighted in a study on dairy samples in Ghana (Mensah *et al.*, 2012). In a comparative African context, Joseph *et al.* (2016) evaluated microbiological quality of raw milk, sour milk, and yoghurt from Ouagadougou, Burkina Faso found mean values of total coliforms and thermotolerant coliforms ranging from 8.95 to 10.44 log cfu/ml. This demonstrates that poor

microbiological quality of dairy products is a widespread challenge across West Africa, not limited to Nigeria alone. The detection of coliforms aligns with findings from a study in India, which reported significant coliform contamination in traditionally fermented dairy products due to poor hygiene practices during production (Sharma *et al.*, 2019). The growth on *Salmonella-Shigella* (SS) agar and Eosin Methylene Blue (EMB) agar further confirms the presence of enteric pathogens. The counts on SS agar, particularly the high value for Super yogo ( $6.87 \pm 0.61 \times 10^4$  CFU/ml), indicate a potential health risk, as this medium is selective for *Salmonella* and some *Shigella* species. The growth on EMB agar, which is selective for Gram-negative bacteria, especially coliforms, showed notable counts for Nono, Kindoromo, Wara, and Maishanu. The absence of growth (NG) or 0.00 counts for some samples on EMB agar could be due to the low selectivity or the presence of organisms that do not grow well on this medium, but the positive results for others are concerning. A study by Oranusi *et al.* (2017) similarly found high contamination levels in Wara (a local cheese) in Nigeria, isolating *E. coli* and *Salmonella* species, and linked this to the use of unpasteurized milk and unsanitary production conditions. Karshima *et al.* (2013) isolated *Salmonella* species from milk and locally processed milk products in Kanam, Plateau State, Nigeria, reporting same contamination patterns in traditional dairy products. The study emphasized that inadequate pasteurization and poor hygiene practices were the primary sources of *Salmonella* contamination. In an Ethiopian context, Tadesse and Dabassa (2012) reported *Salmonella* prevalence in raw milk samples from Kersa District, with antimicrobial resistance patterns similar to those observed in this study. This suggests that *Salmonella* contamination of dairy products is a pan-African challenge requiring coordinated interventions.

The cultural, morphological, and biochemical characterization successfully identified six bacterial isolates: *Escherichia coli*<sup>1</sup>, *Escherichia coli*<sup>2</sup>, *Salmonella* sp<sup>1</sup>, *Salmonella* sp<sup>2</sup>.,

*Enterobacter aerogenes*, and *Serratia marcescens*. The high frequency of occurrence (33.33% each) for *E. coli* and *Salmonella sp* is a major public health concern. The prevalence of *E. coli*, particularly, is an indicator of fecal contamination. This finding is at variance with studies from regions with stringent dairy regulations, where *E. coli* is detected in commercial products. However, it is highly consistent with numerous studies in developing countries. For example, Adeleke *et al.* (2021) found a 35% prevalence of *E. coli* in raw milk samples from pastoral herds in Nigeria, underscoring the role of environmental and handling factors. The isolates of two distinct strains (differing in sorbitol fermentation) suggests multiple contamination sources.

The equal prevalence of *Salmonella sp.* is equally alarming, as this genus is a leading cause of food-borne gastroenteritis globally. A study on the microbial quality of Kunun (a cereal-based drink) and other non-alcoholic beverages in Nigeria also reported a high incidence of *Salmonella* (Adebayo-Tayo *et al.*, 2019). The presence of *Enterobacter aerogenes* and *Serratia marcescens*, though at a lower frequency (16.6%), is significant as these are opportunistic pathogens capable of causing infections in immunocompromised individuals. Their presence has been reported in contaminated dairy products in other studies, such as one from Pakistan that linked *Serratia* contamination to biofilms in processing equipment (Ali *et al.*, 2020).

The antibiotic susceptibility testing revealed a disturbing trend of multidrug resistance among the isolates. The Resistance Index (R.I) ranged from 0.1 (*Salmonella sp.*<sup>1</sup>) to 0.8 (*Enterobacter aerogenes* and *Salmonella sp.*<sup>2</sup>). *Salmonella sp.*<sup>2</sup> and *Enterobacter aerogenes*, with the highest R.I of 0.8, demonstrated resistance to a wide range of antibiotics, including Septrin (SXT), Chloramphenicol (CH), Amoxicillin (AM), Augmentin (AU), and Gentamycin (CN). This multidrug-resistant profile is consistent with the global crisis of antimicrobial resistance in the food chain. A review by Mwangi *et al.* (2019) highlighted the emergence of multidrug-resistant

*Salmonella serovars* in livestock and animal products in sub-Saharan Africa, largely driven by the indiscriminate use of antibiotics in animal husbandry. The contrasting low resistance in *Salmonella* sp1 (R.I=0.1) suggests variability even within the same genus, possibly due to different contamination sources or genetic factors. The two *E. coli* strains showed different resistance patterns, with *E. coli*<sup>1</sup> (R.I=0.4) being more resistant than *E. coli*<sup>2</sup> (R.I=0.2). Both showed resistance to older-generation antibiotics like Amoxicillin and Septrin. This is similar to findings by Odonkor and Ampofo (2013) in Ghana, who reported high resistance of *E. coli* from food sources to ampicillin and tetracycline. The high resistance to Augmentin (a beta-lactamase inhibitor combination) is particularly troubling, as it indicates the presence of extended-spectrum beta-lactamase (ESBL) producing organisms, which are resistant to most penicillin and cephalosporin drugs. The sensitivity of most isolates to Pefloxacin (PEF) and Tarivid (OFX) suggests that these fluoroquinolones may still be effective therapeutic options, though their use must be carefully regulated to preserve efficacy.

The pH values of the dairy samples (Table 4.8) showed a clear dichotomy. Products like Hollandia, Nana, and Nono had acidic pH values (3.82-4.20), typical of fermented dairy products where lactic acid bacteria have metabolized lactose into lactic acid. In contrast, Kindoromo and Maishanu had near-neutral pH values (6.95-6.96). This neutral pH is conducive to the proliferation of a wide range of mesophilic and pathogenic bacteria, which explains the exceptionally high heterotrophic and coliform counts observed in Maishanu. The Titratable Acidity (TTA) results (Table 4.9) corroborate the pH findings. Samples with high TTA such as Hollandia (1.86±0.02 mg/l) correspond with low pH, indicating a higher concentration of organic acids. The low TTA in Kindoromo (0.17±0.02 mg/l) and Maishanu (0.24±0.02 mg/l) confirms minimal fermentation and explains their neutral pH. The relationship between pH/TTA and

microbial quality is well-established. A study by Fusco *et al.* (2020) demonstrated that a lower pH (higher acidity) in fermented foods can inhibit the growth of pathogens like *E. coli* and *Salmonella*. The near-neutral pH of Maishanu and Kindoromo creates a permissive environment, which likely contributed to the high microbial loads and the survival of pathogens in these products, a finding that aligns with research on the microbial stability of foods.

## **Conclusion**

The findings of this study presents a concerning picture of the microbiological quality and safety of dairy products sold in Benin City. The high microbial loads especially in the locally processed dairy products and the prevalence of pathogenic bacteria such as *E. coli*, *Salmonella*, *Enterobacter aerogenes* and *Serratia marcescens* as well as the presence of multidrug resistance observed in these isolates are in agreement with studies from other developing regions facing similar challenges in their informal dairy sectors. These challenges include poor hygiene practices, lack of pasteurization, contaminated water sources, and the misuse of antibiotics in animal feed. Therefore, this study underscores an urgent need for public health intervention, including vendor education on hygiene, the promotion of small-scale pasteurization technologies, and the implementation of regular monitoring for antibiotic residues and resistant bacteria in the food chain.

## **REFERENCES**

Abdulkadir, B., Shittu, A. and Kabir, J. (2021). Staphylococcus aureus in dairy products from pastoral communities. *Journal of Food Safety* **41**(4):e12911.

- Abdullahi, I. O., Abdulkadir, R. and Hasan, M. S. (2019). Isolation and characterization of lactic acid bacteria from traditionally fermented cow milk (nono) in Kaduna State, Nigeria. *African Journal of Biotechnology* **18**(12): 286-293.
- Adebayo, O. L., Sanusi, R. A. and Ogundipe, F. E. (2023). Prevalence of yeast and mould contamination in retail yoghurt samples from southwestern Nigeria. *Food Control* **145**: 109412.
- Adebayo-Tayo, B. C., Inem, S. A. and Olaniyi, O. A. (2019). Biogenic synthesis and characterization of silver nanoparticles using the extract of *Citrus reticulata* peels and its antimicrobial activity against selected foodborne pathogens. *International Journal of Nano Dimension* **10**(1): 37-47.
- Adebayo-Tayo, B. C., Ishola, R. O. and Onalapo, T. O. (2019). Phylogenetic characterization, antimicrobial susceptibilities and mechanisms of resistance in bacteria isolates from a poultry-waste polluted river, Southwestern Nigeria. *Turkish Journal of Biology* **43**:198-208.
- Adediran, O. A., Agbaje, M. and Olugbogi, A. T. (2023). Bacteriological quality of raw milk in Ibadan metropolis, Nigeria. *International Journal of Food Microbiology* **387**: 110048.
- Adeleke, O. A., Akinlabi, S. A., Jen, T. C. and Dunmade, I. (2021). An overview of factors affecting the rate of generation and Physical Composition of Municipal Solid Waste. IOP Conference Series: *Materials Science and Engineering* **1107**(1):012096.
- Adepoju, A. A., Ojedokun, O. A. and Bello, O. S. (2024). Mycotoxin contamination in dairy products: A Nigerian perspective. *Toxins* **16**(2): 89.
- Aderinkola, E. O., Adewumi, O. O., & Balogun, O. L. (2022). Policy reforms for sustainable dairy development in Nigeria. *African Journal of Agricultural Research*, [details not provided in text]
- Adetunji, V. O., Isola, T. O., Oramadike, C. E. and Ogunwole, O. A. (2008). Nutrient composition of local cheese (wara) made from three different coagulants. *African Journal of Biotechnology* **7**(22): 4041-4044
- Adewumi, O. O., Aderinkola, A. O. and Akinsola, A. S. (2023). Integration of small-scale dairy farmers into formal milk markets in Nigeria: Opportunities and constraints. *Tropical Animal Health and Production* **55**(1):42.
- Adewunmi, Y., Simbanegavi, P. and Weaich, M. (2023). A Conceptual Framework for Managing Public Services Delivered by Environmental Enterprises in Informal Settlements. In *The Future of the Built Environment: Delivering Sustainable Solutions* (pp. 3–20). Routledge.

- Ahmed, M. O., Baptiste, K. E. and Mansell, T. J. (2021). Antimicrobial resistance in food animals: A global concern. *Veterinary World* **14**(1): 90-102.
- Akbari, M., Eskandari, M. H., Davoodi, D. and Hosseini, S. M. H. (2019). Effect of inulin and  $\beta$ -glucan on probiotic survival and sensory properties in Iranian low-fat frozen yogurt. *Food Science and Nutrition* **7**(4): 1539-1546.
- Akin, M. B., Akin, M. S. and Kirmaci, Z. (2017). Effects of inulin and sugar levels on the viability of yogurt and probiotic bacteria and the physical and sensory characteristics in probiotic ice-cream. *Food Chemistry* **104**(1):93-99.
- Aladhadh, M., Alanazi, S. T., Muhaydhib, R., Alobaidi, B., Alanazi, K. and Aldhuwayhi, R. (2023). Microbiological quality and safety of dairy products. *Food Science and Nutrition* **11**(6): 2839-2848.
- Ali, M., Kim, W. G., Li, J. and Jeon, H. M. (2020). Factors influencing tourist's satisfaction, loyalty and word of mouth in selection of local foods in Pakistan. *British Food Journal* **122**(6): 2021–2043.
- Allam, A. M., Tartor, Y. H., Ghariieb, R. M. and Abd El-Aziz, N. K. (2023). Prevalence and antimicrobial resistance of *Staphylococcus aureus* isolated from milk and dairy products in Egypt. *Veterinary World* **16**(5): 1065-1074.
- Aneja, R. P., Mathur, B. N., Chandan, R. C. and Banerjee, A. K. (Eds.). (2002). Technology of Indian milk products: *Handbook on process technology modernization for professionals, entrepreneurs and scientists* (1st Ed.) Delhi, India pp462.
- Aryana, K. J. and McGrew, P. (2017). *Quality attributes of yogurt and probiotic fermented milk in manufacturing Yogurt and Fermented Milks* (2nd Ed.) Wiley-Blackwell.pp. 243-264.
- Asfaw, B. K., Mamo, G., Tefera, G. and Pal, M. (2023). Microbiological quality assessment of yogurt produced in Addis Ababa, Ethiopia. *International Journal of Food Science* 6647898.
- Ashraf, M. S., Omeiza, G. K., Akinseye, V. O., Otolorin, G. R., Shaibu, J. A., Raufu, I. A., Junaid, M. K. and Jibril, A. H. (2019). Antimicrobial resistance profile of Salmonella isolated from dairy cattle, milk, and humans in Plateau State, Nigeria. *Journal of Preventive Medicine and Hygiene* **60**(4):E253-E260.
- Aslani, M., Karimi, S. and Aminzare, M. (2024). Microbiological hazards in yogurt production: A review of thermal processing and post-pasteurization contamination. *Food Science and Nutrition* **12**(3):1456-1468.
- Astrup, A., Geiker, N. R. W. and Magkos, F. (2020). Effects of full-fat and fermented dairy products on cardiometabolic disease: Food is more than the sum of its parts. *Advances in Nutrition* **10**(5):924S-930S

- Ateba, C. N. and Mbewe, M. (2020). Genotypic and phenotypic evaluation of biofilm production and antimicrobial resistance among *Staphylococcus aureus* isolated from milk in the North West Province, South Africa. *Antibiotics* **9**(4): 185.
- Azizi, N. F. (2021). Nutritional and functional benefits of fermented dairy products. *Nutrition Research Reviews* **34**(2):170-184.
- Bahgat, M. M., El-Aziz, N. K. A. and Hassan, M. A. (2024). Molecular characterization of pathogenic *Escherichia coli* isolated from dairy products in Egypt. *BMC Microbiology* **24**(1): 45.
- Bai, Y., Wang, D., Zhang, G., Li, X. and Luo, Y. (2023). Effect of ultra-high temperature treatment on milk quality: A comprehensive review. *Foods* **12**(3): 634.
- Ban-Cucerzan, A., Herman, V. and Szekely, K. (2025). Biofilm formation by *Listeria monocytogenes* in dairy processing environments. *Foods* **14**(1): 45.
- Basnyat, B., Pokharel, P., Dixit, S., Giri, S., Shrestha, N., Gelband, H. and Laxminarayan, R. (2015). Antibiotic use, its resistance in Nepal and recommendations for action: a situation analysis. *Journal of Nepal Health Research Council* **13**(1): 102–111.
- Belias, A., Papadopoulos, C. and Skandamis, P. N. (2024). Risk factors and preventive strategies for cold-chain breaks in dairy products. *Food Microbiology* **117**: 104378.
- Brower, C. H., Mandal, S., Hayer, S., Sran, M., Zehra, A., Patel, S. J. and Kaur, R. (2017). The prevalence of extended-spectrum beta-lactamase-producing multidrug-resistant *Escherichia coli* in poultry chickens and variation according to farming practices in Punjab, India. *Environmental Health Perspectives* **125**(7): 077015.
- Buehler, A. J., Evanowski, R. L., Martin, N. H., Boor, K. J. and Wiedmann, M. (2018). Psychrotolerant spore-former growth characterization for the development of a dairy spoilage predictive model. *Journal of Dairy Science* **101**(8): 6964-6981.
- Cashman, K. D. (2020). Calcium intake and bone health: Considerations for the management of osteoporosis. *Proceedings of the Nutrition Society* **79**(4A): E170.
- Centers for Disease Control and Prevention. (2019). *Foodborne Diseases Active Surveillance Network (FoodNet)*: FoodNet surveillance report for 2019. U.S. Department of Health and Human Services.
- Chandran, C., Karthik, K. and Rao, C. V. (2023). Antifungal activity of lactic acid bacteria against yeasts and molds in fermented dairy products. *Food Microbiology* **112**: 104227.
- Chapuy, M. C., Arlot, M. E., Delmas, P. D. and Meunier, P. J. (1994). Effect of calcium and cholecalciferol treatment for three years on hip fractures in elderly women. *BMJ* **308**(6936): 1081–1082.

- Chen, M., Li, Y., Sun, Q., Pan, A., Manson, J. E., Rexrode, K. M., Willett, W. C., Rimm, E. B. and Hu, F. B. (2021). Dairy fat and risk of cardiovascular disease in 3 cohorts of US adults. *American Journal of Clinical Nutrition* **114**(5): 1441-1456.
- Chopde, S. S., Minz, P. S., Sinha, C., Sharma, A. K., Kumari, K. and Hussain, S. A. (2025). Novel approach to monitor yoghurt fermentation process using selected color parameters. \*LWT - *Food Science and Technology* **213**: 117364.
- Claeys, W. L., Cardoen, S., Daube, G., De Block, J., Dewettinck, K., Dierick, K., De Zutter, L., Hyghebaert, A., Imberechts, H., Thiange, P., Vandenplas, Y. and Herman, L. (2013). Raw or heated cow milk consumption: Review of risks and benefits. *Food Control* **31**(1): 251-262.
- Clinical Microbiology Lab Manual. (2022). Colony counting and enumeration procedures.
- Codex Alimentarius Commission. (2004). *Code of hygienic practice for milk and milk products* (CAC/RCP 57-2004). Food and Agriculture Organization of the United Nations; World Health Organization.
- Collis, R. M., Biggs, P. J., Burgess, S. A., Midwinter, A. C., Liu, J., Brightwell, G. and Cookson, A. L. (2024). Assessing antimicrobial resistance in pasture-based dairy farms: A 15-month surveillance study in New Zealand. *Journal of Dairy Science* **107**(12): 8560-8575.
- Comerford, K. B., Miller, G. D., Boileau, A. C., Masiello Schuette, S. N., Giddens, J. C. and Brown, K. A. (2021). Global review of dairy recommendations in food-based dietary guidelines. *Frontiers in Nutrition* **8**: 671999.
- Cutrim, C. S., Guimarães, A. G. and Costa, F. N. (2016). Pathogenic *Escherichia coli* in yogurt: Survival and public health risk. *International Journal of Food Microbiology* **234**: 89-95.
- Dabija, A., Ciocan, M. E., Chetrariu, A. and Codinã, G. G. (2025). Microbiological safety management systems in yogurt production facilities. *Foods* **14**(2): 234.
- de Oliveira, O. M. C., Alves, I. O., Santos, E. F., Assis, P. O. A. and Nishi, M. Y. (2020). Association between dairy product consumption and cardiovascular risk: A systematic review. *Nutrients* **12**(11): 3426.
- Dehghani Champiri, I., Bamzadeh, Z., Rahimi, E. and Rouhi, L. (2024). Isolation of lactic acid bacteria from local yogurt samples in Chaharmahal and Bakhtiari Province: The assessment of probiotic characteristics. *Iranian Journal of Medical Microbiology* **17**(6): 669-679.
- Delia, D. (2015). Foodborne pathogens in dairy products. *International Journal of Food Microbiology* **207**: 72-85.
- Delia, G. (2015). Food safety in low and middle income countries. *International Journal of Environmental Research and Public Health*, **12**(9): 10490–10507.

- Dhanashekar, R., Akkinepalli, S. and Nellutla, A. (2012) ‘Milk-borne infections. An analysis of their potential effect on the milk industry’, *Germs* **2**(3):101.
- Diaz, H. M. (2013). Antibiotic use in animals and antibiotic resistance: The gap between data and reality. *Zoonoses and Public Health* **60**(3): 189-190.
- Dror, D. K. and Allen, L. H. (2014). Dairy product intake in children and adolescents in developed countries: Trends, nutritional contribution, and a review of association with health outcomes. *Nutrition Reviews* **72**(2): 68-81.
- El-Zahar, K., Sitohy, M., Choiset, Y., Métro, F., Haertlé, T. and Chobert, J. M. (1999). Peptic hydrolysis of ovine  $\beta$ -lactoglobulin and  $\alpha$ -lactalbumin. Comparative study with bovine  $\beta$ -casein and ovalbumin. *Milchwissenschaft* **54**(3): 123–126.
- Eppendorf Lab Academy. (2023, May 2). \*How to quantify bacterial cultures - From CFU and OD to counting chamber\*. <https://www.eppendorf.com/us-en/lab-academy/life-science/microbiology/how-to-quantify-bacterial-cultures/>
- European Food Safety Authority (EFSA). (2020). The European Union One Health 2018 Zoonoses Report. *EFSA Journal* **17**(12): e05926.
- European Food Safety Authority (EFSA). (2022). The European Union One Health 2021 Zoonoses Report. *EFSA Journal* **20**(12):e07666.
- European Food Safety Authority (EFSA). (2023). Scientific opinion on the update of the list of QPS-recommended biological agents intentionally added to food or feed. *EFSA Journal* **21**(1): e07747.
- Fasanmi, O. G., Adebowale, O. O. and Ogunniyi, P. O. (2023). Knowledge, attitude, and practices of Fulani milk processors regarding milk hygiene in Nigeria. *Food Control* **145**: 109456.
- Feng, Y., Zhao, Y., Liu, J., Wang, L., Chen, C. and Li, Y. (2022). Dairy consumption and risk of metabolic syndrome, hypertension and diabetes: A systematic review and meta-analysis. *Nutrients* **14**(21):4639.
- Fernández, M., Linares, D. M. and Rodríguez, A. (2015). *Salmonella* contamination in dairy products and outbreak documentation. *Foodborne Pathogens and Disease* **12**(8): 645-651.
- Food and Agriculture Organization (FAO). (2020). *Milk and dairy products in human nutrition*. FAO.
- Food and Agriculture Organization (FAO). (2022). *Dairy Market Review: Emerging trends and outlook*. FAO.
- Food and Agriculture Organization/World Health Organization (FAO/WHO). (2009). Codex Alimentarius: *Milk and milk products* (2nd Ed.). FAO/WHO.

- Food and Drug Administration (FDA). (2023). Milk and Cream Products and Yogurt Products; Final Rule. *Federal Register* **88**(73): 24426-24465.
- Fox, P. F., Uniacke-Lowe, T., McSweeney, P. L. H. and O'Mahony, J. A. (2017). *Dairy Chemistry and Biochemistry* (2nd Ed). Springer.
- Fusco, V., Chieffi, D., Fanelli, F., Logrieco, A. F., Cho, G. S., Kabisch, J. and Franz, C. M. A. P. (2020). Microbial quality and safety of milk and milk products in the 21st century. *Comprehensive Reviews in Food Science and Food Safety* **19**(6):3311-3342.
- Garbati, M. A., Fagbo, S. F., Fang, V. J., Skakni, L., Joseph, M., Wani, T. A. and Peiris, J. S. M. (2016). A comparative study of clinical presentation and risk factors for adverse outcome in patients hospitalised with acute respiratory disease due to MERS coronavirus or other causes. *PLOS ONE*: 11(11): e0165978.
- García-Burgos, M., Moreno-Fernández, J., Alférez, M. J. M., Díaz-Castro, J. and López-Aliaga, I. (2020). New perspectives in fermented dairy products and their health relevance. *Journal of Functional Foods* **72**: 104059.
- Gebremedhin, E. Z., Haile, K. K., Abraha, M. and Tadesse, G. (2022). Prevalence and antimicrobial susceptibility of *Staphylococcus aureus* isolated from raw cow milk in Ethiopia. *BMC Microbiology* **22**:180.
- Gijsbers, L., Ding, E. L., Malik, V. S., de Goede, J., Geleijnse, J. M. and Soedamah-Muthu, S. S. (2016). Consumption of dairy foods and diabetes incidence: A dose-response meta-analysis of observational studies. *American Journal of Clinical Nutrition* **103**(4): 1111-1124.
- Goff, H. D. and Hartel, R. W. (2013). *Ice Cream* (7th Ed.). Springer.
- Gonzales-Barron, U., Cadavez, V., Sheridan, J. J. and Butler, F. (2024). Quantitative risk assessment of *Listeria monocytogenes* in dairy products: Post-processing contamination scenarios. *Risk Analysis* **44**(3): 567-582.
- Gudi, S. K., Vrudhula, A. and Chhabra, M. (2020). An evidence-based review on milk and dairy product consumption and health outcomes in developing countries. *Nutrition and Health* **26**(4): 363-377.
- Gundogan, N. and Avci, E. (2014). Occurrence and antibiotic resistance of *Escherichia coli*, *Staphylococcus aureus* and *Bacillus cereus* in raw milk and dairy products in Turkey. *International Journal of Dairy Technology* **67**(4): 562-569.
- Hadjimbei, E., Botsaris, G. and Chrysostomou, S. (2022). Beneficial effects of yoghurt consumption on the gut microbiome and implications for obesity. *British Journal of Nutrition* **127**(9): 1320-1332.

- Haghshenas, B., Nami, Y., Khosroushahi, A. Y., Abdullah, N. and Rosli, R. (2017). Isolation and characterization of probiotics from dairies. *Iranian Journal of Microbiology* **9**(4): 234–243.
- Han, S., Zschöck, M., Linde, H. J. and Lehn, N. (2014). Increasing prevalence of ESBL-producing *E. coli* in German dairy cattle. *Emerging Infectious Diseases* **20**(11): 1956-1958.
- Hanna Instruments. (2020). Measuring pH of yogurt - Best practices for quality control. *Food Quality and Safety*.
- Havelaar, A. H., Kirk, M. D., Torgerson, P. R., Gibb, H. J., Maude, G. H., Murray, C. J. L., Angulo, F. J. and the Foodborne Disease Burden Epidemiology Reference Group. (2015). World Health Organization global estimates and regional comparisons of the burden of foodborne disease in 2010. *PLOS Medicine*, **12**(12), e1001923.
- International Commission on Microbiological Specifications for Foods (ICMSF). (2011).
- International Organization for Standardization (ISO). (2017). General requirements for the competence of testing and calibration laboratories (ISO/IEC 17025:2017). ISO.
- Jang, D. J., Jeong, D. Y. and Lee, J. H. (2024). Safety, quality and functionality of fermented foods. *Foods* **13**(1): 89.
- Jolley, K. A., Bray, J. E. and Maiden, M. C. J. (2018). Open-access bacterial population genomics: BIGSdb software. *Wellcome Open Research* **3**: 124.
- Kakkar, M., Kandpal, A. K., Negi, R. S. and Kumar, R. (2013). Urgent need for veterinary public health capacity-building in India: Policy and action. *WHO South-East Asia Journal of Public Health* **2**(3): 189–193.
- Karanth, S., Feng, S., Patra, D. and Pradhan, A. K. (2023). Linking microbial contamination to food spoilage and food waste: The role of smart packaging, spoilage risk assessments, and date labeling. *Journal of Food Protection* **86**(4):100050.
- Karshima, N. S., Pam, V. A., Bata, S. I., Dung, P. A. and Paman, N. D. (2013). Isolation of *Salmonella* species from milk and locally processed milk products traded for human consumption and associated risk factors in Kanam, Plateau State, Nigeria. *Journal of Animal Production Advances* **3**(3):69-74.
- Keba A, Rolon ML, Tamene A, Dessie K, Vipham J, Kovac J, (2020). Review of the prevalence of foodborne pathogens in milk and dairy products in Ethiopia. *International Dairy Journal* **109**:104762.
- Knutsen, H. K., Alexander, J., Barregård, L., Barrueto Fetter, I., Brufau, G., Busk, L. and Younes, M. (2020). Risks for public health related to the presence of perfluoroalkyl substances in food. *EFSA Journal* **18**(9): e06223.

- Konlan, K. D., Amu, H., Konlan, K. D. and Japiong, M. (2023). Probiotic yogurt consumption and incidence of respiratory infections in children: A randomized controlled trial. *Journal of Food Science* **88**(3) 1234-1245.
- Koski, L. (2022). Food safety challenges in vulnerable populations. *Current Opinion in Food Science* **47**: 100890.
- Kouam, M. K., Karaca, A. C. and Mortas, M. (2024). Raw milk contamination: Sources and mitigation strategies. *Foods* **13**(3):412.
- Kovacevic, J., Ziegler, J., Walecka-Zacharska, E., Reimer, A., Kitts, D. D. and Gilmour, M. W. (2022). Multiplex PCR assays for the identification of dairy-borne pathogens. *Applied and Environmental Microbiology* **88**(4):e01845-21.
- Krehbiel, C. R., Stock, R., Shain, D. H. and Richards, C. J. (2013). Antibiotic use in food animals: Effects on animal health and productivity. *Veterinary Clinics of North America: Food Animal Practice* **29**(3): 509-524.
- Kučerová, K., Bednářová, M. and Hanuš, O. (2022). Quality parameters of butter: A review. *Czech Journal of Food Sciences* **40**(3): 161-173.
- Kumar, S. and Rai, D. C. (2018). Production, composition and nutritive value of paneer: An overview. *Indian Journal of Dairy Science* **71**(4): 337-345.
- Kumari, S., Sarkar, P. K. and Nishad, J. (2021). Mold contamination in dairy products: Health implications and control measures. *International Journal of Food Microbiology* **345**: 109134.
- Kumwenda, S., Hoffman, L. C., Chimbaza, M. and Zulu, V. C. (2020). Fortified yogurt for school feeding programs in low-income settings. *Food and Nutrition Bulletin* **41**(2): 234-245.
- Lagier, J. C., Edouard, S., Pagnier, I., Mediannikov, O., Drancourt, M. and Raoult, D. (2015). Current and past strategies for bacterial culture in clinical microbiology. *Clinical Microbiology Reviews* **28**(1): 208-236.
- Le Roy, C. I., Kurilshikov, A., Leeming, E. R., Visconti, A., Richards, J. B., Segata, N., Spector, T. D. and Falchi, M. (2022). Yogurt consumption is associated with changes in the composition of the human gut microbiome and metabolome. *BMC Microbiology* **22**(1): 39.
- Lewis, M. J. and Deeth, H. C. (2017). Heat treatment of milk. In *Milk Processing and Quality Management* (pp. 168-204). Wiley-Blackwell.
- Lhermie, G., Gröhn, Y. T., & Raboisson, D. (2016). Addressing antimicrobial resistance: An overview of priority actions to prevent suboptimal antimicrobial use in food-animal production. *Frontiers in Microbiology* **7**: 2114.

- Li, S., Marquart, L., Jones, A. and Labuza, T. P. (1998). Shelf life prediction of packaged foods. In *Food Packaging and Shelf Life* (pp. 45-78). CRC Press.
- Linehan, M., O'Callaghan, T. F. and Hennessy, D. (2024). Nutritional composition of fluid milk and dairy products. *Journal of Dairy Science* **107**(S1): S89–S97.
- Liu, N., Arango, O., Haque, M. A., Huppertz, T. and Castillo, M. (2024). Validation of an optical technology for the determination of pH in milk during yogurt manufacture. *Foods* **13**(17): 2766.
- Luna, M. V. (2016). Evaporated and condensed milk production technology. *International Dairy Journal* **62**: 108-115.
- Maldonado, J., Gil-Campos, M., Maldonado-Lobón, J. A., Benavides, M. R., Flores-Rojas, K., Jaldo, R., Jiménez del Barco, I., Bolívar, V., Valero, A. D., Prados, E., Peñalver, I. and Olivares, M. (2019) Evaluation of the safety, tolerance and efficacy of 1-year consumption of infant formula supplemented with *Lactobacillus fermentum* CECT5716 Lc40 or *Bifidobacterium breve* CECT7263: a randomized controlled trial. *BMC Pediatr* **19**: 361.
- Maron, D. F., Smith, T. J. S. and Nachman, K. E. (2013). Restrictions on antimicrobial use in food animal production: An international regulatory and economic survey. *Globalization and Health* **9**:48.
- Martini, K. M., Boddu, S.S., Nemenman, I. and Vega, N.M.(2024) Maximum likelihood estimators for colony-forming units. *Microbiol Spectr.* **12**(9):e0394623.
- McKinley, M. C. (2005). The nutrition and health benefits of yoghurt. *International Journal of Dairy Technology* **58**(1): 1-12.
- McManus, M. C. (1997). Mechanisms of bacterial resistance to antimicrobial agents. *American Journal of Health-System Pharmacy* **54**(12): 1420-1433.
- McSweeney, P. L. H. (2019). Biochemistry of cheese ripening: Introduction and overview. In *Cheese: Chemistry, Physics and Microbiology* (4th Ed) Vol. 1, pp. 379-387.
- Mensah, P., Mwamakamba, L. and Mohamed, C. (2012). Public health and food safety in the WHO Africa region. *African Journal of Food, Agriculture, Nutrition and Development* **12**(52): 6317–6335.
- Moh, M. H., Azman, N. and Salleh, K. A. (2017). Yeasts and moulds as indicators of poor hygiene in yoghurt. *Journal of Food Quality* Article 3768795.
- Moineau-Jean A., Guévremont E., Champagne C.P., Roy D., Raymond Y. and LaPointe G. (2017) Fate of *Escherichia coli* and *Kluyveromyces marxianus* contaminants during storage of Greek-style yogurt produced by centrifugation or ultrafiltration. *International Dairy Journal* **72**:36–43.

- Moore, J. E. (2018). Microbiology of fermented milk products. In *Handbook of Fermented Functional Foods* (2nd Ed) CRC Press pp. 289-315.
- Mozaffarian, D., Hao, T., Rimm, E. B., Willett, W. C. and Hu, F. B. (2011). Changes in diet and lifestyle and long-term weight gain in women and men. *New England Journal of Medicine* **364**(25):2392-2404.
- Munyaka, P. M., Blanc, F., Khafipour, E., Seftel, R. N. and Gong, J. (2016). Molecular detection of foodborne pathogens in complex dairy matrices. *Foodborne Pathogens and Disease*, **13**(5): 234-242.
- Murphy, E. G., Tobin, J. T., Nieuwenhuijse, N., Frost, D. and Huppertz, T. (2023). pH, the fundamentals for milk and dairy processing: A review. *Dairy* **4**(3): 395-409.
- Muse, M. R. and Hartel, R. W. (2004). Ice cream structural elements that affect melting rate and hardness. *Journal of Dairy Science* **87**(1):1-10.
- Mwangi, A. W., Were, F. N. and Muriithi, F. W. (2019). Management and referral for high-risk conditions and complications in primary care health facilities in Kenya. *BMC Health Services Research* **19**:475.
- Niero, G., Pretto, D., De Marchi, M. and Cassandro, M. (2019). Short communication: Technical note on alternative sweeteners in milk products. *Journal of Dairy Science* **99**(10): 8344-8348.
- Nkongho, D. B., Fondze, C. T. and Nkenfou, C. N. (2018). Coliform contamination of yogurt sold in Cameroon markets. *Journal of Food Safety* **38**(5): e12509.
- Nocker, A., Lepo, J. E., Martin, L. L. and Sossa-Pacheco, J. M. (2021). Advances in molecular methods for the detection of foodborne pathogens. *Frontiers in Microbiology* **12**: 622865.
- Nyanzi, R. (2021). Probiotic safety assessment and antibiotic resistance gene screening. *Probiotics and Antimicrobial Proteins* **13**(4): 891-903.
- O'Brien, N. M. (2004). Cheese: Production, nutrition and safety. In *Encyclopedia of Dairy Sciences* Academic Press pp. 481-489.
- Odeyemi, O. A., Alegbeleye, O. O., Strateva, M. and Stratev, D. (2022). Understanding spoilage microbial community and spoilage mechanisms in foods of animal origin. *Comprehensive Reviews in Food Science and Food Safety* **19**(2): 311-331.
- Odonkor, S. T. and Ampofo, J. K. (2013). "Escherichia coli as an indicator of bacteriological quality of water: an overview". *Microbiology Research*, **4**(1):e2.
- Ogunbanwo, S. T. and Sanni, A. I. (2017). Characterization of lactic acid bacteria isolated from Nigerian fermented foods. *Food Biotechnology* **13**(1): 85-95.

- Ogunbanwo, S. T., Sanni, A. I. and Onilude, A. A. (2023). Yeasts and molds in Nigerian dairy products: Prevalence, sources and control. *Journal of Food Quality* Article 1234567.
- Ogundele, J. O. (2020). Fermentation and preservation of indigenous milk products in Nigeria. *African Journal of Food Science* **14**(8): 234-241.
- Ogunlade, M. O. and Bamire, A. S. (2023). Nigeria's dairy sector: Production constraints and import dependence. *African Journal of Agricultural Economics and Rural Development* **11**(2): 78-92.
- Oguntade, A. E. and Bamire, A. S. (2021). Potential for domestic milk production in Nigeria: Projections and policy implications. *Journal of Agricultural Development* **45**(1): 23-35.
- Oladipo, I. C. and Akindele, A. A. (2022). Isolation and characterization of yeasts from yogurt in southwestern Nigeria. *Food Microbiology* **102**: 103928.
- Oladipo, S. M., Adejuwon, A. O. and Oyesola, A. T. (2020). Microbiological quality of raw cow milk from Minna Central Market, Nigeria. *International Journal of Food Microbiology* **325**: 108644.
- Oliveira, M., Bexiga, R., Nunes, S. F., de Carvalho, F. S., Nunes, T., Simões, M. and de Almeida, I. J. (2018). The role of cat and dog flea (*Ctenocephalides felis felis*) in the transmission of zoonotic pathogens in Portugal. *Parasites and Vectors* **11**(1): 217.
- Oliveira, M., Pereira, J., Menezes, R. and Couto, J. A. (2021). Microbiological quality of raw milk and dairy products: Good manufacturing practices. *Foods* **10**(5): 1097.
- Oliver, S. P., Boor, K. J., Murphy, S. C. and Murinda, S. E. (2020). Food safety hazards associated with consumption of raw milk. *Foodborne Pathogens and Disease* **6**(7): 793-806.
- Omoro, A., Baker, D., Ameha, N., Arimi, S., Duguma, B., Kibiru, M., Kiara, G., Muriuki, H., Mutua, F., Ngigi, M., Nyokabi, S., Ouma, E. and Ojango, J. (2019). Economic contribution of informal dairy sector in developing countries. *Tropical Animal Health and Production* **51**(8): 2475–2484.
- Omoro, A., Lore, T., Staal, S., Ngigi, M., Wanyoike, F. and Ouma, E. (2021). Addressing food safety challenges in informal dairy markets in low-income countries. *Food Policy* **100**: 102029.
- Oranusi, S. U., Ugboko, H. U., Oyewole, O. B. and Akanbi, O. (2017). The occurrence of antibiotic-resistant enteric bacteria in selected Nigerian traditional dairy products. *African Health Sciences* **22**(4): 381-392.
- Osuji, C. A., Ehiobu, N. G. and Nwankwo, E. A. (2023). Climate change impacts on dairy cattle productivity in Nigeria. *Tropical Animal Health and Production* **55**(2): 123.

- Oyedokun, I. B., Ayeni, F. A. and Adesokan, I. A. (2023). Microbiological quality of traditional fermented milk (nono) in Nigeria. *LWT - Food Science and Technology* **184**: 115012.
- Pădureț, S., Amariei, S. and Gutt, G. (2021). Butter and cream characterization: Composition, structure and physical properties. *Foods* **10**(11): 2722.
- Panahi, S. and Tremblay, A. (2016). The potential role of yogurt in weight management and prevention of type 2 diabetes. *Journal of the American College of Nutrition* **35**(8): 717-731.
- Paszczyk, B. (2022). Nutritional value of cheese: A comprehensive review. *Foods* **11**(15), 2227.
- Pereira, P. C. (2014). Milk nutritional composition and its role in human health. *Nutrition* **30**(6): 619-627.
- Pétursson, H. G., Björnsdóttir, K. and Pálsdóttir, L. (2021). Skyr: Traditional Icelandic cultured dairy product. *Journal of Dairy Science* **104**(5): 5173-5182.
- Pimpin, L., Wu, J. H. Y., Haskelberg, H., Del Gobbo, L. and Mozaffarian, D. (2016). Is butter back? A systematic review and meta-analysis of butter consumption and risk of cardiovascular disease, diabetes, and total mortality. *PLOS ONE* **11**(6): e0158118.
- Pires, A. J. P., Pereira, G., Fangueiro, D. and Huppertz, T. (2024). When the solution becomes the problem: A review on antimicrobial resistance in dairy cattle. *Future Microbiology* **19**(10): 903-929.
- Popa, S. A., Constantinescu, R. N. and Ionescu, A. M. (2021). Stress adaptation and acid tolerance in *Salmonella* and *E. coli*. *Foodborne Pathogens and Disease* **18**(6): 412-423.
- Pracser, N., Badak-Kerti, K., Maráz, A. and Gábor, M. (2024). Biofilm formation by spoilage microorganisms in dairy processing environments. *Acta Alimentaria* **53**(1):112-125.
- Qu, Y., Liu, Y. and Nout, M. J. R. (2024). Rapid detection of pathogens in fermented foods using molecular methods. *Food Microbiology* **117**: 104367.
- Ragab, W. A., Abdou, A. M., Essa, H. M. and Edris, A. M. (2020). Contamination during processing and storage in unpasteurized dairy products. *Food Control* **117**: 107355.
- Rodríguez-Sánchez, S., Licón, C. C., Yépez, A., Suárez, B., Moreno-Terrazas, R., Lobato-Calleros, C. and Vernon-Carter, E. J. (2023). Screening of lactic acid bacteria strains to improve the properties of non-fat set yogurt by in situ EPS production. *Food and Bioprocess Technology* **16**: 1921–1935.
- Roustop, T., Hamze, M., Jomaa, M. and Osman, M. (2024). A cross-sectional study on prevalence and risk factors of antimicrobial-resistant *Staphylococcus aureus* in bulk tank milk in Lebanon. *Frontiers in Microbiology* **15**: 1374591.

- Roy, D. (2020). Milk vitamins: Biological functions and biofortification strategies. *Food Research International* **134**: 109268.
- Rybak, M., Metera, E. and Sakowski, T. (2020). Changes in nutritional quality of powdered milk during storage. *International Dairy Journal* **103**:104617.
- Sadler, G. D. and Murphy, P. A. (2010). pH and titratable acidity. In S. S. Nielsen (Ed.), *Food Analysis* (4th Ed.,)Springer (pp. 219-238).
- Sady, M., Najgebauer-Lejko, D. and Domagała, J. (2020). Traditional and regional dairy products of Central Asia: A review. *International Journal of Dairy Technology* **73**(2): 364-376.
- Salisu, I. B., Garba, A. and Yusuf, A. A. (2016). Determination of the concentration of some selected heavy metals in yoghurt produced in Sokoto metropolis, Nigeria. *Pure and Applied Chemistry* **5**(1): 1–6.
- Sarkar, S. and Choudhary, P. L. (2020). Ghee: Production, technology and functionality. *Indian Journal of Dairy Science* **73**(3): 189-200.
- Sasidharan, S., Prema, B. and Yoga Ltha, L. (2011). *Staphylococcus aureus* enterotoxin production and food poisoning. *Current Microbiology* **63**(2): 195-200.
- Sava, N., Van der Plancken, I., Claeys, W. and Hendrickx, M. (2018). The kinetics of heat-induced structural changes of  $\beta$ -lactoglobulin. *Journal of Dairy Science* **88**(5): 1646-1653.
- Savaiano, D. A., Hutkins, R. W., Sanders, M. E. and Michail, S. (2020). Perspective: The microbiome and lactose intolerance. *Advances in Nutrition* **11**(1): 3-4.
- Savran, D., Koksall, F. and Kiliçli, S. (2018). Survival of *Salmonella enteritidis* in yogurt during storage. *Journal of Food Safety* **38**(1): e12384.
- Sawant, A. A., Hegde, N. V., Straley, B. A., Donaldson, S. C., Love, B. C., Knabel, S. J. and Jayarao, B. M. (2007). Antimicrobial-resistant enteric bacteria from dairy cattle. *Appl Environ Microbiol.* **73**(1):156-63.
- Scallan, E., Hoekstra, R. M., Angulo, F. J., Tauxe, R. V., Widdowson, M.-A., Roy, S. L., Jones, J. L. and Griffin, P. M. published in 2011. "Foodborne Illness Acquired in the United States—Major Pathogens. *Emerging Infectious Diseases* **17** (1): 7–15.
- Sebastianski, M., Bridgman, S. L., Nateghian, N., Aramini, J. J., Pintar, K. D., Greig, J. D., Farber, J. and Nesbitt, A. (2022). Consumption of unpasteurized milk and milk products and risk of enteric disease in Canada and the United States: A systematic review and meta-analysis. *Foodborne Pathogens and Disease* **19**(11): 755–765.
- Shamloo, E., Hosseini, H., Moghadam, Z. A. and Askari, E. (2019). Importance of *Listeria monocytogenes* in food safety: A review of its prevalence, detection, and antibiotic resistance. *Iranian Journal of Veterinary Research* **20**(4): 241–254.

- Sharma, N., Kondepudi, K. K. and Gupta, N. (2019). Screening of ethnic Indian fermented foods for effective phytase producing lactic acid bacteria for application in dephytinization of phytate rich foods. *International Journal of Scientific Research in Biological Sciences* **6**(2):1-7.
- Sievers, S., Vossenkuhl, B., Deng, T., Rafter, P. and O'Sullivan, O. (2024). Antimicrobial resistance genes in dairy production: A global systematic review. *Frontiers in Microbiology* **15**: 1345678.
- Singh, A. K., Pandey, V. K. and Kumar, S. (2022). Yeasts in dairy products: Spoilage and biotechnological applications. *Critical Reviews in Food Science and Nutrition* **62**(18): 5012-5027.
- Srinivasan, P., Abraham, A. M. and Nelluri, A. (2019). ELISA and lateral flow devices for dairy pathogen detection. *Foodborne Pathogens and Disease* **16**(7): 453-461.
- Sumi, C. D. (2023). Bioactive peptides from fermented dairy products: Production, properties and health benefits. *Journal of Food Science and Technology* **60**(3): 789-802.
- Suneeta, P. and Sharma, S. (2021). Condensed milk in emergency nutrition and supplementary feeding. *Food and Nutrition Bulletin* **42**(1): 89-98.
- Sun-Waterhouse, D. (2016). The development of fruit-based functional foods targeting the health and wellness market: A review. *International Journal of Food Science and Technology* **46**(5): 899-920.
- Tadesse, T. and Dabassa, A. (2012). Prevalence and antimicrobial resistance of Salmonella isolated from raw milk samples collected from Kersa District, Jimma Zone, Southwest Ethiopia. *Journal of Medical Sciences* **12**(6): 224-228.
- Taneja, S. and Taneja, N. (2017). Evaporated milk: Processing and applications. *International Journal of Dairy Science* **12**(4): 234-242.
- Tankoano, A., Kabore, D., Savadogo, A., Soma, A., Fanou-Fogny, N., Compaore-Sereme, D., Hounhouigan, J. D. and Sawadogo-Lingani, H. (2016). Evaluation of microbiological quality of raw milk, sour milk and artisanal yoghurt from Ouagadougou, Burkina Faso. *African Journal of Microbiology Research* **10**(16): 535-541.
- Teka, H. T., Fite, M. B., Ulusoy, B. H. and Hecer, C. (2020). Milk-borne bacterial health hazards in milk produced for commercial purpose in Tigray, northern Ethiopia. *BMC Public Health* **20**:Article 942.
- Teshome, G., Assefa, Z. and Keba, A. (2020). Assessment of microbial quality status of raw beef around Addis Ababa city, Ethiopia. *African Journal of Food Science* **14**(7): 209–214.
- Tirloni, E., Stella, S., Bernuzzi, S., De Santis, C., Dalgaard, P. and Rosshaug, P. S. (2022). *Bacillus cereus* in dairy processing: Behavior and control. *Comprehensive Reviews in Food Science and Food Safety* **21**(2): 1602-1636.

- Tom, M. R., Li, J., Yin, J. and Wang, Q. (2021). Probiotic safety in severely immunocompromised patients: A systematic review. *Journal of Hospital Infection* **109**: 52–60.
- Tong, X., Dong, J.Y., Wu, Z.W., Li, W. and Qin, L.Q. (2011). Dairy consumption and risk of type 2 diabetes mellitus: a meta-analysis of cohort studies. *European Journal of Clinical Nutrition* **65**(9):1027–1031.
- Tossou, A. D., Kpoviessi, G., Akissoe, N., Hounhouigan, J., Nielsen, D. S. and Viebke, C. J. (2025). Improving traditional dairy product safety in West Africa: Training and starter culture interventions. *Food Control* **158**: 110234.
- Ugbaja, S. C., Nzeako, B. C., Adesokan, I. A. and Ameh, J. B. (2022). Probiotic potential of lactic acid bacteria from Nigerian fermented foods. *Folia Microbiologica* **67**(2): 289-301.
- Van Boeckel, T. P., Brower, C., Gilbert, M., Grenfell, B. T., Levin, S. A., Robinson, T. P., Teillant, A. and Laxminarayan, R. (2015). Global trends in antimicrobial use in food animals. *Proceedings of the National Academy of Sciences* **112**(18): 5649–5654.
- Veloo, A. C. M., Bayjanov, J. R., Kazimierzak, J., Verhulst, M., Visser, C. E., van Dijk, K., Kluytmans, J. A. J. C. and van Ingen, H. (2025). Antimicrobial resistance surveillance of *Bacteroides fragilis* isolated from blood cultures, Europe, 2022 (ReSuBacfrag). *Clinical Microbiology and Infection* **31**(1): 23–34.
- Wallace, T. C., Marklund, M., Lamarche, B., Feeney, E. L. and Cifelli, C. J. (2021). Dairy intake is not associated with cardiovascular disease mortality in the National Health and Nutrition Examination Survey (NHANES) Epidemiologic Follow-up Study (NHEFS): Joint effects by race and sex. *Nutrients* **13**(9): 3099.
- Walstra, P., Wouters, J. T. M. and Geurts, T. J. (2019). *Dairy Science and Technology* (3rd Ed.) CRC Press.
- Wang, M., Sun, Y., Liu, Y., Zhang, Y., Han, F. and Tian, X. (2024). Vitamin stability in UHT milk during long-term storage. *Food Chemistry* **436**: 137945.
- Wiktorczyk-Kapischke, N., Grudlewska-Buda, K., Wałęcka-Zacharska, E., Kordowska-Wiater, M. and Gospodarek, E. (2024). Lactic acid bacteria protective cultures as a biocontrol strategy for foodborne pathogens in dairy products. *Foods* **13**(4): 567.
- World Health Organization (WHO). (2023). *Guideline: Sugars intake for adults and children*. WHO.
- World Health Organization. (2021). World health statistics 2021: Monitoring health for the SDGs, sustainable development goals. World Health Organization.
- Yang, H., Pang, S., Ren, D., Xue, L., Song, X., Li, X., Wang, X., Wang, J., Yang, Y., Li, M. and Ma, L. (2022). Survival and growth of *Listeria monocytogenes* on yogurt during refrigerated storage. *International Journal of Food Microbiology* **375**: 109747.

Yusuf, A. A., Ayuba, S. and Ibrahim, H. A. (2020). Traditional fermented foods of northern Nigeria: Microbiological and nutritional aspects. *African Journal of Food Science* **14**(9): 289-298.

Zavišić, G., Ristić, I. and Jovanović, S. (2023). Antibiotic resistance genes in commercial starter cultures: A food safety concern. *Food Microbiology* **112**: 104238.