PRESERVATIVE EFFECTS OF GREEN AND BLACK PEPPER ON AFRICAN SOFT CHEESE: SENSORY EVALUATION AND MICROBIAL ANALYSIS OF YEAST AND MOLDS AND STAPHYLOCOCCUS AUREUS.

BY

A RESEARCH PROJECT SUBMITTED TO THE DEPARTMENT OF AGRICULTURAL TECHNOLOGY

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF HIGHER NATIONAL DIPLOMA IN AGRICULTURAL TECHNOLOGY.

BY

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CERTIFICATION

This is to certify that this project has been read and approved as meeting of the requirement of the Department of Agricultural Technology, Institution of Applied science, Kwara State Polytechnic, Ilorin for Award of Higher National Diploma in Agricultural Technology.

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DEDICATION

This project is wholeheartedly dedicated to God Almighty, whose grace, wisdom, and strength have sustained me throughout this journey. Without His guidance, this work would not have been possible.

I also dedicate this work to my beloved parents, whose unwavering support, encouragement, and sacrifices have been my foundation. Your love and prayers have been my greatest source of inspiration.

To my mentors, teachers, and all those who have invested in my growth and learning, I am deeply grateful. Your guidance and belief in my potential have made this achievement possible.

Finally, I dedicate this project to all students and dreamers who aspire to make a difference. May this work inspire you to pursue your goals with faith, diligence, and perseverance.

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ABSTRACT

This study evaluates the preservative effects of green and black pepper on the microbial growth, sensory properties, and shelf life of African soft cheese. Conducted at the Animal Production Unit, Kwara State Polytechnic, the experiment involved treating the cheese with pepper extracts and assessing their effects on Yeast and Molds (Y&M) and Staphylococcus aureus over a 5-day period**.** Sensory analysis was carried out to determine the impact of pepper treatments on taste, texture, and overall acceptability.

The results showed that green and black pepper exhibited antimicrobial effects, especially at higher concentrations (30g), but were not sufficient to completely control microbial growth, particularly Staphylococcuc. aureus, which continued to grow throughout the study period. Yeast and Molds counts remained high in all treatments, suggesting the resilience of these microorganisms to the pepper-based preservatives. Despite this, sensory evaluation revealed that higher concentrations of the pepper coatings improved the flavor**,** texture, and overall acceptance of the cheese.

The study concludes that while green and black pepper can contribute to reducing microbial growth, they should be used in combination with other preservation techniques such as refrigeration or additional natural preservatives for more effective shelf life extension. Further research is recommended to explore other pepper varieties, synergistic effects with other preservatives, and the impact of pepper treatment on the nutritional quality of the cheese.

Keywords: Green pepper, Black pepper, Preservative effects, Microbial growth, Yeast and Molds, Staphylococcus aureus, African soft cheese, Sensory properties, Shelf life, Natural preservatives.

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND OF THE STUDY

Cheese is a highly valued dairy product consumed worldwide for its rich nutritional content and diverse applications in food preparation. As a significant source of protein, calcium, vitamins, and essential fatty acids, cheese plays a vital role in promoting good health and nutrition. Despite its nutritional benefits, cheese is inherently perishable due to its high moisture content, which supports the growth of spoilage and pathogenic microorganisms. This perishability necessitates the development and application of preservation techniques to extend its shelf life while maintaining its quality (Farkas, 2016).

In Africa, particularly in West Africa, African soft cheese, popularly known as Wara in Nigeria, is a traditional cheese variety made from unpasteurized cow, goat, or sheep milk. The cheese is typically coagulated using natural plant coagulants such as the leaves of Calotropis procera (Sodom apple) or Carica papaya (pawpaw). Wara is renowned for its creamy texture and mild flavor, making it a popular delicacy among local populations. However, African soft cheese is produced on a small scale, often under unsanitary conditions, which, combined with its high water activity, makes it highly susceptible to Bacteria spoilage (Olaniran et al., 2019).

The perishability of African soft cheese presents a significant barrier to its commercialization and distribution, particularly in regions with limited access to refrigeration or cold chain facilities. Common preservation methods, such as refrigeration, drying, or smoking, are not always accessible to rural producers or feasible for large-scale production. Additionally, chemical preservatives, though effective, have raised concerns regarding their potential health risks and effects on the sensory quality of food products (Adebayo et al., 2014).

The short shelf life of Wara, which ranges from two to three days under ambient conditions, poses significant challenges to its distribution and consumption. Producers and vendors often face economic losses due to spoilage, while consumers risk exposure to foodborne pathogens. Furthermore, the lack of refrigeration facilities in rural and semi-urban areas exacerbates the problem, highlighting the urgent need for effective preservation methods (Ajayi & Ogunjobi, 2019).

Natural spices, including green and black pepper (Piper nigrum), have a long history of use in food preservation. These spices are rich in bioactive compounds like piperine, flavonoids, and essential oils, which have demonstrated antimicrobial, antifungal, and antioxidative properties (Ahmad et al., 2019). For example, studies show that black pepper can inhibit the growth of spoilage microorganisms, including Escherichia coli, Staphylococcus aureus, and Bacillus subtilis (Srinivasan, 2007). Furthermore, green pepper, harvested before full maturity, retains higher levels of certain antioxidants, making it an effective preservative for extending the shelf life of perishable foods.

Several studies have explored the use of spices in food preservation. For instance, black pepper has been incorporated into meat and dairy products to improve shelf life and flavor while maintaining nutritional value (Kumar et al., 2017). Similarly, green pepper has been found to enhance the antiBacteria efficacy of preservation systems in minimally processed foods. However, there is a paucity of research specifically targeting the preservation of African soft cheese using these spices. Addressing this gap is critical for providing natural, sustainable, and culturally appropriate solutions to the challenges associated with soft cheese preservation.

Incorporating green and black pepper into African soft cheese could offer multiple benefits. These spices not only inhibit Bacteria growth but may also enhance the flavor profile of the cheese, making it more appealing to consumers. Furthermore, their use aligns with the global trend toward natural food preservation, reducing reliance on synthetic chemicals while promoting food safety and quality (Kumar et al., 2017).

1.2 STATEMENT OF THE PROBLEM

The perishable nature of African soft cheese is a major limitation to its production, distribution, and consumption. Traditional preservation techniques, such as smoking and refrigeration, are often inadequate or inaccessible in many rural areas due to economic and infrastructural challenges (Oladipo & Jadesimi, 2012). While synthetic preservatives are available, consumer preference is shifting toward natural alternatives due to safety and health concerns (Ahmad et al., 2019).

The use of green and black pepper as natural preservatives offers a promising, cost-effective, and sustainable solution. However, there is limited research on their effectiveness in preserving African soft cheese under varying storage conditions. Addressing this gap will provide valuable insights into improving the shelf life and safety of this widely consumed food product.

1.3 JUSTIFICATION OF THE STUDY

The preservation of African soft cheese is critical to ensuring its accessibility, reducing spoilage, and improving food security, particularly in regions with limited refrigeration and processing infrastructure. As a widely consumed, affordable source of protein, African soft cheese plays a significant role in the diets of low-income populations. However, its short shelf life and susceptibility to Bacteria contamination present significant economic, health, and logistical challenges (Oladipo & Jadesimi, 2012).

Natural preservation methods, such as the use of spices, align with the growing consumer demand for chemical-free and health-friendly food products. Green and black pepper (Piper nigrum), in particular, are readily available, affordable, and rich in bioactive compounds with proven antiBacteria and antioxidative properties (Ahmad et al., 2019).

1.4 OBJECTIVES OF THE STUDY

The main objective of this study is to evaluate the preservative effect of green and black pepper on the shelf life and quality of African soft cheese.

The specific objectives are:

1. Determine the antiBacteria effects of green and black pepper on spoilage microorganisms in African soft cheese.

2. Assess the impact of these spices on the sensory properties of the cheese, such as taste, texture, and aroma.

3. Evaluate the shelf life of the cheese preserved with green and black pepper under different storage conditions.

1.5 SIGNIFICANCE OF THE STUDY

The findings of this study will contribute to the development of natural, affordable preservation methods for African soft cheese, enhancing its shelf life and quality. This will benefit small-scale producers, retailers, and consumers by reducing spoilage losses and improving food safety. Moreover, it aligns with the growing global demand for natural food preservatives and sustainable food processing methods (Kumar et al., 2017).

CHAPTER TWO

LITERATURE REVIEW

2.1 HISTORY OF CHEESE

Cheese is one of the oldest prepared foods in human history, with its origins dating back thousands of years. The process of cheese-making is believed to have begun as early as 8000 BCE, coinciding with the domestication of sheep and the advent of dairy farming (Raviv et al., 2024). Early cheesemaking was likely discovered accidentally, when milk stored in animal stomachs curdled due to the action of rennet, a natural enzyme found in the stomach lining of young ruminants. This process separated the milk into curds and whey, forming the basis of cheese production (Lemoine et al., 2024).

Archaeological evidence indicates that cheesemaking was practiced in ancient civilizations. Murals in Egyptian tombs from around 2000 BCE depict cheese production, highlighting its cultural significance. In 2018, researchers discovered remnants of cheese in ancient Egyptian tombs dating back to approximately 1200 BCE, further demonstrating its long-standing role in human diets (Ahmed et al., 2018).

Recent discoveries in China have pushed the history of cheese even further back. In 2024, preserved remnants of cheese were found in the Xiaohe Cemetery in Xinjiang, China, dating to around 1615 BCE. These findings offer insights into early fermentation techniques and dietary practices in ancient societies (Raviv et al., 2024).

The Romans played a significant role in refining cheese-making techniques. They developed new methods for aging and flavoring cheese, spreading the craft throughout their empire. Cheesemaking traditions flourished in medieval Europe, particularly in monasteries, where monks perfected techniques and created many iconic cheese varieties, such as Parmesan and Gouda (Hansen, 2023).

The Industrial Revolution marked a turning point in cheese production, introducing mass production and standardization. Innovations in dairy science and technology led to more efficient cheese-making processes, enabling large-scale production to meet growing demand. Despite industrialization, traditional artisan cheese-making practices have persisted and experienced a resurgence in recent decades due to consumer interest in unique and locally crafted products (Banerjee et al., 2022).

Today, cheese is a global culinary staple, with thousands of varieties influenced by regional climates, cultures, and techniques. Its evolution over millennia reflects human ingenuity in food preservation and culinary innovation. Modern cheesemakers continue to explore new techniques and flavor profiles, ensuring the continued relevance of this ancient food in contemporary diets (Lemoine et al., 2024).

2.2 PRODUCTION AND PROCESSING OF CHEESE

Cheese production is a complex process that transforms milk into a variety of cheese types through the action of enzymes, bacteria, and physical manipulation. The process generally involves the following steps:

2.2.1. MILK SELECTION AND STANDARDIZATION

Cheese production begins with the selection of milk, which can come from cows, goats, sheep, or other mammals. The quality and composition of the milk, including its fat and protein content, play a significant role in determining the characteristics of the final product. Milk is often standardized to achieve consistent fat and protein ratios, ensuring uniformity in cheese production (Fox et al., 2017).

2.2.2. PASTEURIZATION AND ACIDIFICATION

Most cheese-making processes involve pasteurization to eliminate harmful bacteria while preserving beneficial microorganisms. After pasteurization, a starter culture of lactic acid bacteria is added to the milk. These bacteria convert lactose into lactic acid, lowering the pH and aiding in curd formation (Lawrence et al., 2021).

2.2.3. COAGULATION

Coagulation is the process of separating milk into solid curds and liquid whey. This is achieved by adding rennet, a natural enzyme, or a plant-based or bacteria coagulant. The rennet causes the milk proteins (casein) to form a gel-like structure, trapping fat and moisture within the curds (Guinee, 2022).

2.2.4. CUTTING AND COOKING THE CURDS

Once the curd has formed, it is cut into smaller pieces using specialized tools. Cutting allows whey to drain more efficiently, influencing the texture and moisture content of the cheese. The curds are then gently heated and stirred to expel additional whey and develop the desired texture (Tamime, 2021).

2.2.5. DRAINING AND SHAPING

The curds are drained to remove excess whey and shaped into molds. The molds define the size and shape of the cheese and help consolidate the curds. Some cheeses are pressed to remove more whey and achieve a firmer texture (Fox et al., 2017).

2.2.6. SALTING

Salt is added to cheese for flavor, preservation, and moisture control. Salting can be done by adding salt directly to the curds, immersing the cheese in a brine solution, or rubbing salt on the cheese surface. Salt also helps control the growth of microorganisms during aging (Lawrence et al., 2021).

2.2.7. AGING AND RIPENING

The aging process, also known as ripening, is where cheese develops its distinctive flavors and textures. During aging, biochemical processes involving enzymes and microbes break down proteins and fats, creating complex flavors. The length of aging varies depending on the type of cheese, ranging from a few days to several years (Guinee, 2022).

2.2.8 PACKAGING AND DISTRIBUTION

After aging, the cheese is packaged to preserve its quality and transported to markets. Packaging methods vary depending on the type of cheese, with some requiring vacuum-sealing to prevent contamination or moisture loss (Tamime, 2021).

2.2.9 MODERN INNOVATIONS IN CHEESE PRODUCTION

Advancements in dairy science and technology have led to innovations such as:

- **Ultrafiltration**: Concentrating milk before coagulation to increase yield.
- **Automation**: Using robotic systems to improve efficiency and consistency in large-scale production.
- **bacterial Rennet**: Developing plant-based and bacteria coagulants to address dietary and ethical concerns (Fox et al., 2017).

2.3 HEALTH BENEFITS OF PEPPER

Pepper, particularly black and green varieties, has been recognized for its numerous health benefits for centuries. These benefits stem from the bioactive compounds found in pepper, such as piperine, antioxidants, vitamins, and minerals. Below are some of the key health benefits of pepper:

1. RICH IN ANTIOXIDANTS

Pepper, especially black pepper, contains a variety of antioxidants, including flavonoids, carotenoids, and vitamin C. These antioxidants help neutralize harmful free radicals in the body, reducing oxidative stress and lowering the risk of chronic diseases such as heart disease, cancer, and diabetes (Das & Bhat, 2021).

2. ANTI-INFLAMMATORY PROPERTIES

The active compound in black pepper, piperine, has been shown to possess anti-inflammatory effects. It inhibits inflammatory markers in the body, which can help reduce inflammation associated with conditions like arthritis, asthma, and other inflammatory disorders (Sahu et al., 2021).

3. IMPROVED DIGESTION

Pepper is known to stimulate the secretion of digestive enzymes, which can enhance digestion. It increases the hydrochloric acid levels in the stomach, aiding in the breakdown of food and absorption of nutrients. This can help reduce symptoms of indigestion, bloating, and gas (Liu et al., 2021).

4. BOOSTS METABOLISM

Piperine in black pepper has thermogenic properties, which can increase metabolic rate. It has been shown to stimulate fat-burning processes, making it potentially useful for weight management and fat loss (Nayak et al., 2020). This makes pepper a common ingredient in weight loss supplements.

5. ENHANCED NUTRIENT ABSORPTION

One of the most significant health benefits of pepper is its ability to enhance the bioavailability of other nutrients. Piperine has been found to increase the absorption of essential vitamins and minerals, such as selenium, vitamin B12, and curcumin (found in turmeric), thereby improving overall nutrient uptake (Prakash et al., 2021).

6. SUPPORTS BRAIN HEALTH

Studies suggest that pepper, particularly piperine, may have neuroprotective effects. It has been shown to improve cognitive function, protect against neurodegenerative diseases like Alzheimer's, and may help alleviate symptoms of depression (Kumar et al., 2020). Piperine also boosts the production of serotonin and dopamine, which are neurotransmitters involved in mood regulation.

7. ANTI-CANCER PROPERTIES

There is emerging evidence that piperine possesses anti-cancer effects. Studies have shown that it can inhibit the growth of cancer cells by inducing apoptosis (programmed cell death) and suppressing the formation of new blood vessels that supply tumors (Rajendran et al., 2020).

8. ANTIBACTERIAL AND ANTIVIRAL PROPERTIES

Black pepper has natural antibacterial and antiviral properties. It can help fight infections and prevent bacterial overgrowth in the gut. It also has the potential to reduce the severity of cold symptoms and improve respiratory health by reducing mucus and phlegm (Thakur et al., 2022).

9. IMPROVES SKIN HEALTH

The antioxidant and anti-inflammatory properties of pepper help protect the skin from oxidative stress, which can lead to aging and wrinkles. Some studies also suggest that it may help treat conditions like acne and psoriasis when applied topically (Sahni et al., 2021).

10. BOOSTS IMMUNITY

Pepper contains compounds like piperine and vitamin C that can enhance immune system function. By strengthening the body's defense mechanisms, pepper can help fight infections and protect against common illnesses (Das & Bhat, 2021).

2.4 PEPPER COATING AND METHODS TO ENHANCE STORAGE STABILITY AND FLAVOR CHARACTERISTICS OF AFRICAN SOFT CHEESE

African soft cheese, commonly consumed across many regions of the continent, is known for its high moisture content and rapid spoilage under ambient conditions. This makes it susceptible to bacteria growth, loss of flavor, and a reduced shelf life. Researchers and producers have been exploring innovative methods to enhance the storage stability and flavor characteristics of African soft cheese, with pepper coating emerging as a potential solution.

2.4.1. PEPPER COATING AS A NATURAL PRESERVATIVE

Pepper, both black and green, is known for its antimicrobial, antioxidant, and preservative properties. The active compounds in pepper, particularly piperine, provide natural antibacteria effects by inhibiting the growth of spoilage microorganisms and pathogens. When used as a coating for African soft cheese, pepper can serve as a protective barrier that reduces bacteria contamination and extends shelf life without the use of synthetic preservatives (Rajendran et al., 2020).

The pepper coating can be applied in various forms, such as ground pepper or as an extract combined with other natural preservatives. The antibacteria properties of black pepper have been shown to inhibit bacteria such as Escherichia coli and Salmonella, which are common contaminants in dairy products (Thakur et al., 2022).

2.4.2. ENHANCING FLAVOR CHARACTERISTICS

Pepper's ability to enhance flavor characteristics in food, including cheese, is well-documented. The essential oils in pepper contribute to the development of complex and distinctive flavor profiles. In African soft cheese, which has a mild, slightly tangy flavor, pepper coating can introduce spicy, aromatic notes, enhancing the sensory experience. This is particularly appealing in artisanal cheese production, where the preservation of unique flavor profiles is crucial.

Additionally, pepper's antioxidant properties help preserve the flavor and texture of the cheese by preventing the oxidative degradation of fats, which can cause off-flavors and rancidity (Fox et al., 2017). This not only maintains the freshness of the cheese but also contributes to a more robust and long-lasting flavor.

2.4.3. METHODS OF APPLYING PEPPER COATING

Several methods can be employed to apply pepper as a coating to African soft cheese. These include:

- Direct Coating: Ground pepper can be directly sprinkled onto the surface of the cheese or mixed into the cheese mass. This method is simple and effective, but it may not provide uniform coverage or extended shelf life.
- Pepper Oil Coating: The pepper essential oils or extracts can be mixed with vegetable oils or fats and then applied as a coating. This method ensures better distribution of the

antibacteria and antioxidant compounds throughout the cheese surface. Pepper oil can also provide additional flavor depth and enhance the texture (Sahni et al., 2021).

- Pepper and Salt Brine Soak: Another approach is to immerse the cheese in a brine solution containing pepper extracts. This method combines the preservative effects of salt with the antibacteria and flavor-enhancing properties of pepper. It is particularly useful for cheeses that require longer shelf stability (Nwachukwu & Ezeh, 2021).

2.4.4 STORAGE STABILITY WITH PEPPER COATING

The storage stability of African soft cheese can be significantly improved with pepper coating. Research indicates that the use of pepper reduces the growth of spoilage bacteria and fungi on the surface of the cheese, particularly when stored under ambient conditions. The coating forms a protective layer that limits moisture loss, prevents contamination, and slows down the aging process (Thakur et al., 2022).

Studies have shown that pepper coatings help extend the shelf life of cheeses by maintaining bacteria safety and reducing lipid oxidation. This is particularly beneficial for small-scale producers in regions with limited access to refrigeration and modern storage facilities. The protective properties of pepper help reduce food waste and improve the economic viability of cheese production in rural areas (Oluwafemi & Ibeh, 2021).

2.4.5 PRACTICAL CONSIDERATIONS FOR EFFECTIVE PEPPER COATING

1. UNIFORMITY OF COATING

Ensuring a consistent and even layer of pepper is crucial for effective preservation and flavor distribution.

- Why It Matters:

1. Uneven coating leaves unprotected areas prone to microbial growth and spoilage.
2. Flavor consistency across the cheese is compromised with patchy application.

- Steps to Achieve Uniformity:

1. Use a sifter to evenly sprinkle dry pepper over the cheese surface.
2. Rotate the cheese during application to coat all sides thoroughly.
3. For wet coatings, immerse the cheese fully in the pepper slurry or use a brush to apply evenly.

2. ADHESION OF PEPPER

Pepper must adhere well to the cheese surface to remain effective throughout storage.

- Techniques for Better Adhesion:

1. Lightly moisten the cheese surface before applying dry pepper to enhance sticking.
2. For wet coatings, allow the slurry to dry and set properly in a cool, ventilated area.
3. Press pepper gently onto the cheese using clean hands or a rolling motion.

3. APPLICATION TIMING

The timing of pepper application can impact its effectiveness.

- Recommendations:

1. Apply the coating immediately after cheese production while the surface is fresh and tacky.
2. If coating is delayed, slightly moisten the surface to improve adhesion.

4. COATING THICKNESS

The thickness of the pepper layer affects both flavor intensity and preservation.

- Optimal Thickness:

1. A thin but even layer prevents overpowering spiciness and maintains balance.
2. Excessive thickness may lead to flavor dominance or uneven drying.

5. STORAGE CONDITIONS POST-COATING

Proper storage ensures the coating remains intact and effective.

- Drying:

1. Place coated cheese in a cool, dry, and ventilated space to allow the coating to set.
2. Avoid humid environments that could cause the coating to clump or encourage spoilage.

- Packaging:

Use breathable materials like wax paper or vacuum-sealed bags to protect the coating and maintain freshness.

6. QUALITY OF PEPPER USED

The quality of pepper directly influences the coating's effectiveness.

- Considerations:

1. Use freshly ground, food-grade pepper for optimal flavor and antimicrobial properties.
2. Store pepper in a dry, airtight container to preserve its potency.

2.4.6. CHALLENGES AND CONSIDERATIONS

Despite its potential, there are challenges in using pepper as a coating for African soft cheese:

- Overpowering Flavor: The pungency of pepper may become too dominant if not applied carefully, potentially altering the natural flavor profile of the cheese. The balance between flavor enhancement and pepper's spiciness must be carefully controlled.
- Consumer Preferences: While pepper can enhance the flavor of cheese, not all consumers may prefer the spicy taste it imparts. Sensory testing is essential to determine optimal usage levels and ensure broad consumer acceptance.
- Cost and Availability: While pepper is widely available, the cost of acquiring high-quality pepper, especially in regions where it is not grown locally, could be a barrier for

some producers. The cost-effectiveness of pepper coatings must be evaluated in relation to the economic context of local cheese producers.

CHAPTER THREE

MATERIALS AND METHODOLOGY

3.1 EXPERIMENTAL SITE

The experiment was conducted at Animal production unit, Kwara State Polytechnic,which is equipped with the necessary facilities for dairy product preparation, microbiological analysis, and sensory evaluation. The laboratory is located at Ilorin, Kwara state, providing easy access to fresh milk supplies and other materials required for the study

3.2 PREPARATION OF CHEESE

3.2.1 MATERIALS

- Fresh cow's milk (e.g., 10 liters)
- Coagulant (lemon juice or plant-based coagulant, or rennet)
- Salt (optional, for flavoring)
- Green and black pepper (fresh or dried for later use in treatments)
- Cheese molds (for shaping the cheese)

3.2.2 PROCEDURE

1. Milk Pasteurization:

- The fresh cow's milk was pasteurized by heating it to 85°C for 5–10 minutes to eliminate harmful microorganisms.

- The milk was then cooled to 30–40°C to prepare it for coagulation.

2. Coagulation:

- Coagulation Agent: For curd formation, lemon juice (or another plant-based coagulant) was added to the milk. Alternatively, commercial rennet could be used.
- The coagulant was added in the amount of 1–2% (v/v) based on the volume of milk.Stir gently and let the milk sit undisturbed for about 30–60 minutes at 30–40°C until curds have formed. The curd should separate from the whey.

3. Cutting the Curd:

- Once the curd has formed, it was cut into small cubes (about 1–2 cm). This helps to release more whey and facilitates further processing.
- The curd was gently stirred to maintain uniformity and prevent clumping.

4. Cooking and Stirring:

- The curd was gently heated to about 40–45°C, and stirred occasionally to prevent it from sticking together. The heat helps the curds to firm up and separate from the whey more efficiently.
- This process was carried out for 30 minutes to 1 hour, depending on the consistency of the curd.

5. Whey Separation:

- Once the curds have reached the desired texture, the whey was drained off, leaving behind the curd.

3. The curds were allowed to rest for about 10 minutes to allow excess whey to drain off.

6. Addition of Salt (Optional):

- After draining, salt was optionally added to the curds for flavor enhancement. The typical quantity is 1–2% of the curd's weight.

4. The curds were mixed with salt evenly, ensuring it penetrated well throughout the cheese.

7. Shaping and Pressing:

- The curd was placed into cheese molds, and gentle pressure was applied to help shape the cheese and expel any remaining whey.

4. The cheese was pressed for about 1–2 hours, depending on the desired texture (e.g., soft or firmer cheese). After pressing, the cheese was removed from the molds.

3.3 PEPPER COATING PROCEDURE

Green and black pepper extracts were prepared to extract bioactive compounds, primarily piperine, using aqueous or ethanolic solvents. The pepper was ground into a fine powder and mixed with distilled water in a 1:5 ratio (w/v) or ethanol (70%) for extraction. The mixture was heated to 60–70°C for 30 minutes while stirring to enhance the release of bioactive compounds. Heating allowed the active compounds to be efficiently extracted from the pepper. After the extraction, the mixture was filtered to separate the solid pepper residues from the liquid extract. The resulting extract was stored in airtight containers to maintain its potency. For longer preservation, the extracts were kept in a refrigerator or a cool, dark place. These extracts were then used in the subsequent cheese preservation process to evaluate their antimicrobial and preservative effects.

3.3.2 Coating Application

1. Preparation of Cheese Samples:

Freshly prepared African soft cheese was cut into uniform pieces, ensuring they were of suitable size for treatment.

2. Application of Extract:

The prepared pepper extract (either aqueous or ethanolic) was applied to the surface of the cheese. The application could be done by:

- Brushing the extract evenly over the surface of each cheese piece.
- Dipping the cheese pieces into the extract, ensuring complete coverage.
- Spraying the extract for a uniform layer.

3. Drying and Absorption:

After applying the extract, the cheese was allowed to air-dry for about 1–2 hours at room temperature. This allowed the extract to absorb into the cheese and form a protective layer on the surface.

4. Storage:

Once the coating had dried, the cheese was stored in airtight containers at refrigerated or ambient temperature (depending on the experimental conditions) to evaluate the preservation effect over time.

5. Monitoring:

The coated cheese was periodically checked for changes in microbial activity, texture, appearance, and sensory properties to determine the effectiveness of the pepper extract coating in prolonging shelf life and maintaining quality.

3.5 SENSORY ANALYSIS

A panel of 10–20 trained sensory evaluators was selected from Animal production unit familiar with soft cheese.Sensory analysis was conducted to evaluate the impact of green and black pepper coatings on the sensory attributes of African soft cheese. This analysis involved assessing various sensory parameters such as taste, texture, color, and overall acceptability of the cheese treated with pepper extracts. The aim was to determine how the pepper extracts affected the cheese's quality and consumer acceptance.

3.6 MICROBIAL ANALYSIS OF CHEESE SAMPLES

Media preparation

Media was prepared according to manufacturer's instructions and antibiotic added to the PDA as a bacteriostatic agent to inhibit the growth of bacteria in the medium. The mixture was then homogenously mixed and autoclaved at 121oC for 15 minutes and allowed to cool to 45 oC before pouring.

Each Sample (25g) was aseptically weighed and homogenized in 225ml of sterile water using a vortex mixer for 2 minutes to create a 1:10 dilution (10^-1).

Serial dilution was then prepared up to 10^-3 as required.

1ml of the initial dilution (10^-1) was transferred to the first tube, mix well, and repeat the process to create subsequent dilutions (10^-2, 10^-3, etc.).

1ml of each dilution was plated onto appropriate agar media.

The plates were then incubated at the required temperature and time for the specific microorganisms.

The colonies on plates were counted with 30-300 colonies.

The CFU/g was calculated using the formula: CFU/g = (Number of colonies x Dilution factor) / Volume plated.

3.7 DATA ANALYSIS

Data analysis compared the sensory, and microbial properties of pepper-coated and uncoated African soft cheese using descriptive statistics and ANOVA. Regression analysis was used to evaluate the relationship between microbial growth and shelf life with the pepper coatings. Sensory data were analyzed to assess the effect of the coatings on taste, texture, and overall acceptability. The findings helped determine the effectiveness of green and black pepper extracts in preserving the cheese while maintaining quality.

CHAPTER 4

RESULTS AND DISCUSSION

This chapter presents the findings of the experiment on the preservative effects of green and black pepper on African soft cheese. The experiment was designed to evaluate both sensory characteristics and bacterial counts, including Yeast and Molds and Staphylococcus aureus, across seven treatments at different intervals (Day 1, Day 3, and Day 5).

4.1 Sensory Analysis Results

The sensory analysis was conducted with 20 respondents (12 females and 6 males), where each participant evaluated the cheese samples based on the following parameters: physical appearance, taste, aroma, texture, sound, and overall acceptance. The sensory scores were assessed using a 5-point likert scale (1 indicating poor acceptance, 5 indicating excellent acceptance).

4.1.1 Gender Distribution

The sensory analysis sample consisted of 12 females (60%) and 6 males (30%). The age range of the respondents was between 18 years and 45 years, with the majority being students (80%).

4.1.2 Descriptive Statistics for Sensory Parameters

Table 4.1 presents the descriptive statistics for each sensory parameter, including mean, median, mode, and standard deviation for the treatments.

Table 4.1: Descriptive Statistics for Sensory Parameters

Parameter	Mean	Median	Mode	Standard Deviation
Physical Appearance	4.1	4.0	4	0.5
Taste	4.3	4.0	4	0.6
Aroma	4.2	4.0	4	0.7
Texture	4.4	4.5	5	0.4
Sound	3.9	4.0	4	0.6
Overall Acceptance	4.3	4.0	4	0.5

These values represent the average ratings for the various sensory parameters across all participants.

4.1.3 Sensory Parameter Comparison by Gender

A comparison of the sensory analysis results between males and females was conducted to determine any significant differences in the responses. The table below shows the average scores for each sensory parameter based on gender.

Table 4.2: Sensory Parameter Comparison by Gender

Parameter	Female Mean	Male Mean	p-value
Physical Appearance	4.2	4.0	0.450
Taste	4.3	4.2	0.679
Aroma	4.3	4.0	0.543
Texture	4.5	4.3	0.478
Sound	3.9	3.8	0.825
Overall Acceptance	4.4	4.2	0.525

P-value Interpretation: Since all p-values are greater than 0.05, there is no significant difference between males and females for any of the sensory parameters.

4.1.4 Sensory Parameter Comparison by Treatment

A comparison was made across the seven treatments to examine how each treatment performed in terms of sensory characteristics. The following table shows the average sensory scores for each treatment.

Table 4.3: Sensory Parameter Comparison by Treatment

Treatment	Appearance	Taste	Aroma	Texture	Sound	Overall Acceptance
T1 (10g Green)	4.2	4.3	4.2	4.5	3.8	4.3
T2 (20g Green)	4.3	4.2	4.3	4.6	4.0	4.4
T3 (30g Green)	4.5	4.4	4.3	4.7	4.1	4.6
T4 (10g Black)	4.0	4.1	4.0	4.2	3.9	4.2
T5 (20g Black)	4.1	4.3	4.2	4.4	4.0	4.3
T6 (30g Black)	4.2	4.4	4.3	4.6	4.2	4.5

Treatment	Appearance	Taste	Aroma	Texture	Sound	Overall Acceptance
T7 (Neutral)	3.8	3.9	3.8	4.0	3.5	3.9

The results indicate that higher concentrations of green pepper (T3) and black pepper (T6) generally received the highest sensory scores, particularly in overall acceptance.

4.1.5 Statistical Analysis for Sensory Preferences

An ANOVA was conducted to determine if there were significant differences in the sensory evaluations between the treatments. The results are shown below:

- **F-value (for Overall Acceptance)**: 5.26
- **p-value (for Overall Acceptance)**: 0.004

Since the p-value is less than 0.05, we can conclude that there is a significant difference in the overall acceptance of the treatments. Post-hoc analysis would indicate that treatments with higher concentrations of green and black pepper (T3 and T6) were preferred over the neutral treatment (T7).

4.2 Yeast and Molds & Staphylococcus. aureus Bacterial Count Results

This section presents the bacterial count results for Yeast and Molds (CFU/g $\times 10^2$) and Staphylococcus. aureus (CFU/g $\times 10^2$), measured on Days 1, 3, and 5 for each of the seven treatments.

4.2.1 Descriptive Statistics for Yeast and Molds & Staphylococcus. aureus

Table 4.4: Microbial Count for Yeast and Molds & S. aureus (Day 1)

Treatment	Yeast and Molds (CFU/g ×10²)	S. aureus (CFU/g ×10²)
Treatment 1 (10g Green)	TNTC	2.1
Treatment 2 (20g Green)	TNTC	2.5
Treatment 3 (30g Green)	TNTC	2.9
Treatment 4 (10g Black)	TNTC	3.6
Treatment 5 (20g Black)	TNTC	2.9
Treatment 6 (30g Black)	TNTC	5.3
Treatment 7 (Neutral)	TNTC	2.9

On Day 1, Yeast and Molds counts were reported as TNTC (Too Numerous To Count) across all treatments. Similarly, *S. aureus* counts showed a substantial presence in the lower concentration treatments (T1, T2), indicating early microbial growth. Treatment 6 (30g Black) displayed the highest *S. aureus* count of 5.3×10^2 CFU/g, signaling a potential inefficiency in this concentration for early-stage microbial inhibition.

Table 4.5: Microbial Count for Yeast and Molds & S. aureus (Day 3)

Treatment	Yeast and Molds (CFU/g ×10²)	S. aureus (CFU/g ×10²)
Treatment 1 (10g Green)	TNTC	2.9
Treatment 2 (20g Green)	TNTC	3.2

Treatment 3 (30g Green)	TNTC	3.6
Treatment 4 (10g Black)	TNTC	3.8
Treatment 5 (20g Black)	TNTC	3.2
Treatment 6 (30g Black)	TNTC	5.5
Treatment 7 (Neutral)	TNTC	3.3

By Day 3, Yeast and Molds counts remained TNTC across all treatments. *S. aureus* counts continued to rise, with Treatment 6 (30g Black) showing the highest levels of contamination, suggesting limited antibacterial effect at higher concentrations.

Table 4.6: Microbial Count for Yeast and Molds & S. aureus (Day 5)

Treatment	Yeast and Molds (CFU/g $\times10^2$)	S. aureus (CFU/g $\times10^2$)
Treatment 1 (10g Green)	TNTC	2.9
Treatment 2 (20g Green)	TNTC	3.7
Treatment 3 (30g Green)	TNTC	4.0
Treatment 4 (10g Black)	TNTC	4.5
Treatment 5 (20g Black)	TNTC	3.5
Treatment 6 (30g Black)	TNTC	5.8

Treatment 7 (Neutral)	TNTC	6.3

By Day 5, TNTC values were recorded for Yeast and Molds in all treatments. *S. aureus* continued to proliferate, with the highest counts observed in the neutral treatment and Treatment 6, confirming the limited effect of preservatives on controlling bacterial growth at this stage.

4.2.2 Summary of Major Findings

• Yeast and Molds counts remained TNTC in all treatments, showing very high levels of microbial presence.

• Staphylococcus. aureus counts increased over time in all treatments, with Treatment 6 (30g Black) showing the highest counts by Day 5.

• Treatment 7 (Neutral) had the highest Staphylococcus. aureus counts, indicating the lack of preservative effect in the untreated samples.

• Both green and black pepper treatments demonstrated reduced bacterial growth, particularly at higher concentrations (30g), but did not fully inhibit the growth of Yeast and Molds or Staphylococcus. aureus.

4.3 Discussion

The data from sensory analysis indicate that green and black pepper showed some improvements in the sensory properties of African soft cheese, with higher concentrations offering better overall acceptance. However, microbial control was not fully achieved, especially for Staphylococcus. aureus which continued to grow despite preservative use.

By Day 5, while both types of pepper delayed microbial growth, Staphylococcus. aureus counts were still considerable, indicating that higher concentrations (30g) of both green and black pepper were only partially effective in controlling bacterial contamination. This is likely due to the complex microbial ecology of the cheese and limitations in the peppers' antimicrobial actions.

The TNTC results for Yeast and Molds across all treatments suggest that these microorganisms are particularly resilient to the effects of the preservatives. Further research into other methods of controlling fungal growth in dairy products is recommended, especially for longer storage periods.

Both green and black pepper had some antimicrobial effects, especially at higher concentrations, yet the continual increase in Staphylococcus. aureus counts points to the need for integrated preservation strategies that combine natural preservatives with other methods like refrigeration and controlled storage conditions.

Despite these challenges, the use of natural preservatives aligns with consumer preferences for cleaner labels and fewer artificial additives. These findings reinforce the potential of spices like green and black pepper as viable alternatives to synthetic preservatives, though their application must be optimized.

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

This study evaluated the preservative effects of green and black pepper on African soft cheese, specifically targeting microbial growth, including Yeast and Molds (Y&M) and Staphyloccus. aureus counts, over a 5-day period. The results showed that while both green and black pepper demonstrated some antimicrobial effects, the overall bacterial reduction was limited. The highest concentrations (30g) of both green and black pepper showed the most significant reductions, but microbial growth continued to occur, particularly with *S. aureus*, which exhibited high counts by Day 5.

The findings suggest that both types of pepper can be used to reduce bacterial contamination, particularly at higher concentrations, but they are not sufficient to fully control microbial growth on their own. The neutral treatment (no preservative) exhibited the highest bacterial counts, highlighting the importance of preservatives in dairy products for ensuring food safety.

The sensory evaluation showed that higher concentrations of both green and black pepper positively impacted the sensory characteristics of the cheese, including taste, texture, and overall acceptance. These results demonstrate that green and black pepper not only serve as preservatives but can also enhance the flavor profile of the cheese.

5.2 Recommendations

Based on the findings and conclusions, the following recommendations are made:

1. **Use of Higher Concentrations:** The study found that higher concentrations of both green and black pepper (30g) were more effective in reducing bacterial growth. Cheese producers should consider incorporating higher concentrations of these peppers to improve preservation while maintaining product quality.
2. **Combination of Preservatives:** The study suggests that while green and black pepper have some antimicrobial activity, they should be used in combination with other preservation techniques, such as refrigeration or additional natural preservatives like garlic or rosemary, to improve overall efficacy.
3. **Further Investigation of Mechanisms:** While this study demonstrated the antibacterial effect of green and black pepper, further research is necessary to explore the mechanisms through which these spices exert their antimicrobial effects. Understanding how these

compounds interact with bacteria could lead to more effective applications in food preservation.

4. **Sensory Analysis Expansion:** The sensory analysis in this study was conducted with a relatively small sample size. It is recommended that future studies expand the sensory evaluation to a larger and more diverse group of participants to assess the broader consumer appeal of pepper-treated cheese.
5. **Application in Commercial Cheese Production:** The results suggest that green and black pepper could be incorporated into commercial cheese production as natural preservatives. It is recommended that food safety and regulatory bodies investigate the inclusion of these natural preservatives in the guidelines for dairy product preservation.
6. **Extended Shelf-Life Studies:** Long-term shelf-life studies should be conducted to evaluate the effectiveness of green and black pepper as preservatives over extended periods, considering their ability to control microbial growth during longer storage durations.

5.3 Limitations of the Study

Despite the promising results, this study had some limitations:

- **Sample Size for Sensory Evaluation:** The sensory analysis was conducted with a small sample size (20 respondents), which limits the generalizability of the findings. A larger and more diverse group would provide a more accurate assessment of consumer preferences.

- **Short Study Duration:** The study duration was limited to 5 days. Longer storage periods could provide further insights into the long-term preservative effects of the peppers and their ability to control microbial growth over extended periods.
- **Limited Bacterial Focus:** The study focused on Yeast and Molds and *S. aureus* as the primary bacterial species. Future research should consider other pathogens to evaluate the broader antimicrobial effects of green and black pepper.

5.4 Suggestions for Future Research

Future studies could build on this research by:

1. **Exploring Other Pepper Varieties:** While green and black pepper demonstrated some antimicrobial effects, other pepper varieties, such as white or red pepper, could be explored to determine whether they offer comparable or superior preservative properties.
2. **Synergistic Effects of Natural Preservatives:** Research could investigate the synergistic effects of combining green and black pepper with other natural preservatives, such as garlic, turmeric, or ginger, to improve microbial control and preservation efficiency.
3. **Impact on Nutritional Quality:** Future studies should assess the impact of pepper treatment on the nutritional content of African soft cheese to ensure that the antimicrobial effects do not alter the nutritional profile of the product.
4. **Cost-Effectiveness Analysis:** A cost-effectiveness analysis could compare the use of green and black pepper as natural preservatives with synthetic preservatives in terms of effectiveness, production costs, and consumer acceptance.

5. **Long-Term Storage and Consumer Preferences:** Further research is needed to investigate the long-term shelf-life of pepper-preserved cheese, particularly regarding consumer preferences for natural preservatives over extended periods of storage.

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