NUTRITIONAL AND MICROBIAL QUALITY OF DIFFERENT BRANDS OF MOZZARELLA CHEESE AVAILABLE IN BANGLADESH

EMA SAHA

Roll No.: 0222/14

Registration No.: 1193

Session: 2022 (July - December)

A thesis submitted in the partial fulfillment of the requirements for the degree of Master of Science in Applied Human Nutrition and Dietetics

Department of Applied Food Science and Nutrition

Faculty of Food Science and Technology

Chattogram Veterinary and Animal Sciences University

Chattogram-4225, Bangladesh

December, 2024

Authorization

I hereby declare that I am the sole author of the thesis. I also authorize the Chattogram Veterinary and Animal Sciences University (CVASU) to lend this thesis to other institutions or individuals for the purpose of scholarly research. I further authorize the CVASU to reproduce the thesis by photocopying or by other means, in total or in part, at the request of other institutions or individuals for the purpose of scholarly research.

I, the undersigned, and author of this work, declare that the electronic copy of this thesis provided to the CVASU Library, is an accurate copy of the print thesis submitted, within the limits of the technology available.

Ema Saha

December, 2024

NUTRITIONAL AND MICROBIAL QUALITY OF DIFFERENT BRANDS OF MOZZARELLA CHEESE AVAILABLE IN BANGLADESH

EMA SAHA

Roll No.: 0222/14

Registration No.: 1193

Session: 2022 (July - December)

This is to certify that we have examined the above Master's thesis and have found that is complete and satisfactory in all respects and that all revisions required by the thesis examination committee have been made.

…………………………

Professor Goutam Kumar Debnath

Supervisor

Department of Dairy and Poultry Science

Faculty of Veterinary Medicine

…………………………

Assistant Professor Mohammad Mozibul Haque

Head of the department &

Chairman of the Examination Committee

Department of Applied Food Science and Nutrition

Faculty of Food Science and Technology

Department of Applied Food Science and Nutrition

Faculty of Food Science and Technology

Chattogram Veterinary and Animal Sciences University

Chattogram-4225, Bangladesh

December, 2024

Acknowledgement

With a deep sense of veneration, I express my gratitude to the supreme of everything, the Almighty God to whom all praises go, who has enabled me to complete the thesis for the degree of Master of Science (MS) in Applied Human Nutrition and dietetics. I express my sincere and deepest gratitude to Professor Dr. Mohammad Lutfur Rahman, Vice-Chancellor, Chattogram Veterinary and Animal Sciences University (CVASU) for giving a special opportunity and providing such research facilities. I feel the inadequacy of my diction to find a more suitable word for a wholehearted thanks to my major advisor and supervisor, Professor Goutam Kumar Debnath, Department of Dairy and Poultry Science, Faculty of Veterinary Medicine, Chattogram Veterinary and Animal Science University for his guidance and moral support. His knowledge, invaluable advice, unceasing encouragement, kind demeanor and constructive criticism significantly enhanced my experience. I am grateful to Professor Dr. Ferdusee Akter, Dean, Faculty of Food Science and Technology, CVASU for his kind cooperation and for providing the necessary facilities to carry out this research. I would also like to convey my gratitude to all of my respected instructors at CVASU's Faculty of Food Science and Technology for their assistance in completing my research work. I also want to thank all of the employees and members of the Departments of Applied Food Science and Nutrition, Dairy Science Laboratory, Animal Science and Nutrition, and Food Processing and Engineering for their and supportiveness in carrying out the research activities accurately. I'd like to thank the Ministry of Science and Technology of Bangladesh, as well as the Center for Advanced Studies and Research at CVASU, for funding this research.

Ema Saha

December, 2024

PLAGIARISM VERIFICATION

Title of Thesis: Nutritional and Microbial quality of different brands of mozzarella cheese available in Bangladesh.

Name of the Student: Ema Saha

Roll number: 0222/14

Reg. number: 1193

Department: Applied Food Science and Nutrition

Faculty: Food Science and Technology

Supervisor: Professor Goutam Kumar Debnath

This is to report that as per the check __19.25%___ of the content of the above thesis is stated to be plagiarized and is covered /not covered as per plagiarism policy and institutions issued from CASR, Chattogram Veterinary and Animal Sciences University.

The thesis may be considered for the evaluation.

____

(Supervisor)

Professor Goutam Kumar Debnath

Department of Dairy and Poultry Science

Faculty of Veterinary Medicine

Table of Contents

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

Abbreviation	Meaning
FAO	Food and Agricultural Organization
e.g.	Exempli gratia (example)
%	Percent
PDO	Protected Designation of Origin
EU	European Union
USDA	United States Department of Agriculture
ISO	International Organization of Standardization
gm	Gram
ml	Milliliters
Kg	Kilogram
ANOVA	Analysis of Variance
FDE	Fat in dry extract
DDE	Defatted dry extract
TDE	Total dry extract
AOAC	Association of Official Analytical Collaboration
TVC	Total Viable Count
TCC	Total Coliform Count
VRB	Violet Red Agar
PDB	Primary Dilution Blank
lbs.	libra pondo (a pound by weight)
IDF	International Dairy Federation
ADSA	American Dairy Science Association
IS	Indian Standard
PCA	Plate Count Agar
Avg.	Average
TBX	Tryptone Bile-X-Glucuronide Agar

Abbreviation	**Meaning**
SPC	Standard Plate Count
DM	Dry matter
MRS	Man, Rogosa and Sharpe Agar
SD	Standard Deviation
BCE	Before common Era
TS	Total Solids
CMA	Chattogram Metropolitan Area
OECD	The Organization for Economic Co-operation and Development
CVASU	Chattogram Veterinary and Animal Sciences University
PCV	Packed cell Volume
WBC	White Blood Cell
MC	Moisture Content

Abstract

Mozzarella cheese is one of the most widely consumed dairy products worldwide owing to its versatility, rich nutritional profile, unique texture. This study aimed to evaluate the nutritional composition, microbial quality, and sensory properties of selected mozzarella cheese brands available in Bangladesh to determine their overall quality and suitability for consumption.

Sensory evaluation based on color, body, texture, flavor, aroma, and appearance revealed that Lactima mozzarella received the highest overall acceptability scores, whereas Goodlife mozzarella scored lower in flavor and finish. Statistical analysis using Tukey's Honest Significant Difference (HSD) test confirmed Significant differences ($p \leq 0.05$) were found in body, texture, flavor, aroma, and appearance, though color differences were statistically insignificant ($p > 0.05$). Proximate analysis revealed significant differences ($p < 0.05$) in moisture, protein, fat, ash, and salt contents across the samples. The protein content of the samples ranged from 17.57% to 25.35%, while the fat content ranged from 13.99% to 22.36%. The study determined that the moisture content, titratable acidity, ash content, and salt content of mozzarella cheese ranged from 48.70 ± 0.54% to 52.98 ± 0.87%, 0.52 ± 0.17% to 1.07 ± 0.38%, 1.83 ± 0.08% to 3.18 ± 0.36%, and 0.39 ± 0.14% to 1.06 ± 0.07%, respectively. Compared to BSTI standards for processed cheese (minimum protein 16%, minimum fat 20%, moisture not exceeding 52%, and maximum salt 2.5%), most samples met or exceeded protein and moisture requirements, although Lactima fell short on fat content (13.99%). All samples complied with the BSTI salt limit. Microbiological analysis identified significant variations in total viable count (TVC) and coliform presence. While three brands of mozzarella cheese exhibited no coliform growth (NG), the remaining one had an average coliform count of 3.62×10^2 CFU/g within BSTI's acceptable range ($<10^3$ CFU/g), although this difference was not statistically significant ($p = 0.12$, NS). TVC results were statistically significant ($p < 0.05$), where the highest bacterial load was (1.86×10^5 CFU/g) and the lowest bacterial load was (5.6×10^3 CFU/g). Notably, one brand of mozzarella cheese showed no detectable bacterial growth (NG), suggesting superior microbiological quality.

Keywords: Mozzarella Cheese, Sensory Evaluation, Nutritional Quality, Microbiological Safety, BSTI Standards.

Chapter 1: Introduction

Dairy products encompass a wide range of commodities derived from the primary and secondary processing of milk obtained from mammals such as cattle, goats, and sheep. Among these, fermented dairy products are produced through the fermentation of milk by beneficial and non-pathogenic microorganisms, including Lactobacillus and Bifidobacterium species. Examples of such products include cheese, paneer, yogurt, cultured buttermilk, and sour cream. According to the Prevention of Food Adulteration (PFA) Act of 1976, cheese is defined as a dairy product obtained by coagulating milk using microbial or vegetable rennet, vinegar, or lemon, followed by the removal of whey. This process occurs under the influence of safe bacterial cultures, ensuring product safety and quality. Davis (1976) described cheese as a nutrient-dense dairy product produced through the coagulation of casein in milk, facilitated by rennet or similar enzymatic agents, in the presence of lactic acid generated by either introduced or naturally occurring microorganisms. The resulting curd undergoes moisture reduction through cutting, cooking, and pressing, after which it is shaped into molds and allowed to ripen under controlled temperature and humidity conditions. During cheese production, lactic acid bacteria (LAB) play a crucial role in acidification, as they contribute to the fermentation process by generating acid in situ (McElhatton and El Idrissi, 2016). Their metabolic activity not only influences the texture and flavor of cheese but also enhances its safety and stability by inhibiting the growth of undesirable microorganisms.

More than 1000 varieties of cheese persist globally (Sandine and Elliker, 1970). Among them, consumers prefer mozzarella cheese mostly. Mozzarella is a semi-soft non-aged cheese prepared by the pasta filata (stretched-curd) method with origins from southern Italy. The unique plasticizing and kneading process of the fresh fibrous curd in hot water, which gives the finished cheese its characteristic structure and melting and stretching characteristics, distinguishes pasta filata cheeses (Kindstedt, 1993). It is prepared not only from cow's milk but also from buffalo milk, taking the following names: 1) "Mozzarella fior di latte" or "mozzarella": cow's milk. 2) Mozzarella di bufala": Italian buffalo's milk. According to a Journal article published by the Bangladeshi Veterinarian (2010), There is slightly difference between cow cheese and buffalo cheese in terms of flavor and taste, finish and color. But a significant difference existed between cow cheese and buffalo cheese in terms of fat, protein, lactose and ash content. Cow cheese has higher amount of fat, protein, lactose content than those of buffalo cheese. Conventional Mozzarella cheese produced using water buffalo milk is one of the most exceptionally esteemed unripened pasta filata cheeses in Battipaglia, Italy certified with the European Protected designation of origin (Citro, 1981). In spite of its production in Italy, it is generally exported and also manufactured in other countries. The particular taste quality of mozzarella cheese chiefly emerges from the raw milk utilized, the area of creation, the ecological circumstances, the conventional tools and producers (Kindstedt and Fox, 1993; Mauriello *et al*., 2003).

In 2020, as the world barreled toward a health crisis triggered by novel coronavirus and people everywhere looked for ways to boost their immunity. Here dairy products like cheese have played an important role in the diet of Bangladeshi people. As cheese is partially digested by the bacteria. For this, it becomes easier to digest, especially for those people who are lactose-intolerant and have milk allergies. Now-a-days, people give more concern in fermented dairy products and maximum people prefer mozzarella cheese to eat. That's why understanding the nutritional and microbial quality of different brands of Mozzarella cheese in Bangladesh is very vital for the safety to the customers. Yet there is limited works have been done for cheese but no comprehensive research has been done in Bangladesh for the quality assessment of Mozzarella cheese. Thus, current study is undertaken to determine the nutritional and microbial quality of different brands of Mozzarella cheese available in Bangladesh for the interest of the people of the country.

Aims and Objectives of the study:

1. To determine nutritional profile of the Mozzarella cheese available in Bangladesh.
2. To determine microbial quality of Mozzarella cheese and know about the source of microbial hazard of cheese.
3. To compare the quality of different brands of mozzarella cheese available in Bangladesh.

Chapter 2: Literature Review

2.1 Introduction:

Cheese is one of the most important and most popular dairy products which has complex biological system. The word cheese comes from a Latin word called causes. This term is also the source of the modern word casein. According to Food Safety and Standards Regulations (FSSR)-2011, Cheese is defined as the ripened or unripened soft or semi-hard, hard and extra hard product, that may be coated with food grade waxes or polyfilm, and in which the whey protein/casein ratio does not exceed that of milk. Cheese is compared to milk from which it is obtained. It is comparatively more reliable, lighter weight and has longer shelf life than milk (Smith, 1995). Cheese is a nutritive product containing essential nutrients particularly proteins, bioactive peptides, amino acids, fat, fatty acids, vitamins and mineral (Walther *et al*., 2008). Cheese can be prepared from whole milk, partially skimmed milk and skimmed milk. This product is very essential for human especially where source of plant protein is not in an adequate quantity (Kosikowski, 1982). Actually, the commencement of cheese making has no authentic evidence. It is assumed that the origin of cheese making ranges from around 8000 BCE in Iraq when people started to propagate plants and animals as a source of food (Fox, 1993; McSweeny, 2007; Fox, 2011). Accidently the cheese making procedure was found out by the storage of milk from the stomach of an animal where the milk was converted to whey and curd by the enzyme rennet from stomach (Silanikove *et al*., 2015). In 2018, scientists examined a whitish residue discovered in the tomb of Ptahmes, an Egyptian official from the 13th century BC, and identified it as an ancient solid cheese. Analysis suggested that the cheese was made from a combination of cow, sheep, or goat milk. This finding represents the oldest known example of solid cheese, estimated to be around 3,200 years old (Razi and Greco,2018). East Asian Culture was unfamiliar to cheese. But, with the expansion of European and Euro-American culture and food, people over the world had come to know about cheese and ultimately cheese got its popularity. In 1815, Switzerland started a factory for the industrial production of cheese. By 2012 cheese was considered one of the most shoplifted dairy products from super shop worldwide (Barkham, 2012). Cheese is also known as functional food. So, people considered it as an ideal food due to its high nutrition, convenience, variety, availability and good taste (Bogue *et al*., 1999). Demand for cheese is increasing day by day. To meet up the consumer's demand, the production of cheese has increased and cottage type cheese industry has transformed into large scale cheese industry by itself (Legg *et al*., 2017). FAO estimated that global cheese production was approximately 18.43 million tons in 2005. According to estimation of International Dairy Federation approximately 23 million ton was produced globally in 2015 (IDF, 2016).

2.2 Classification of Cheese:

There are several types of cheese. Cheeses are classified into different types based on different criteria. These criteria include types of animal milk, species of animal used to produce milk, methods how the animals produce, country or region of origin of animals, extent of fermentation, consistency or texture, amount of fat, pH and calcium content, coagulation type, ripening and curing method, heat treatment, moisture or water content, curd preparation, filtration technique, firmness, microbial characteristics, eye development in cheese, whether rind is allowed to form or not etc. (Fox *et al*., 2000). Single or combined criteria may be applied to produce cheese and globally no single method is used. International Dairy Federation identified that around 500 kinds of cheese are produced (Burkhalter, 1981). Several efforts have been taken to classify cheese with logical and plausible reasons.

Finally, it is suggested that there are 18 remarkable types of natural cheese which are further divided into 4 families by moisture content. The four major families are- Very hard (<42% moisture) Cheese, Hard (43-50% moisture) Cheese, Semi-soft (45-55% moisture) Cheese, Soft (>55% moisture) Cheese.

Pieter Walstra proposed a classification of cheese based on primary and secondary starter with moisture percentage combinedly (Fox PF., 1993), When Walter and Hargrove (1972)) proposed a classification of cheese on the basis of preparation technique. Lawrence *et al.*, 1984 first indicated that cheeses could be divided with the help of two criteria. These Criteria includes pH content and calcium content. Lawrence and his colleagues (1984) also suggested that protein matrix properties, residual lactose content, level of acidity, buffering capacity of the curd influence the final pH of cheese which is ultimately helped in development of new classes of cheese.

Depending on pH, Cheese are two types. They are-

1) Cheese of low pH (Example: - Swiss cheese, Gouda Cheese, Cheddar Cheese, Mozzarella Cheese etc.),

2) Cheese of high pH (Example: - Cheshire Cheese, Ricotta Cheese, Brick Cheese etc.).

According to coagulation process, Cheeses are classified into different super-families and they are further grouped on the basis of principle ripening agents and characteristics technology (McSweeny, 2007). They are stated below-

- Acid coagulated cheese (e.g. Qurag cheese, cottage cheese, cream cheese, Queso Blanko cheese).
- Combined acid and heat coagulated cheese (e.g. Ricotta).
- Concentrated or crystallized cheese (e.g. Mysost cheese).
- Rennet coagulated cheese. This type of cheese is sub grouped into other types of cheese. Such as-
    a) Internal Bacterially Ripened,
    b) Mould Ripened,
    c) Surface Ripened- (Brick, Havarti, Limburger, Munster etc.)

Mould Ripened Cheese are two types.

i) Surface mould Cheese (e.g. Brie, Cambert)
ii) Internal mould Cheese (e.g. Roquefort, Gorgonzala)

Internal Bacterially Ripened Cheeses are 6 types.

i. Extra Hard Cheese (e.g. Grana Padano, Parmesan)
ii. Hard Cheese (e.g. Cheshire, Cheddar)
iii. Semi-hard Cheese (e.g.Caerphilly, Mahon)
iv. Cheese with eyes
    - Swiss type Cheese (e.g. Emmental)
    - Dutch type Cheese (e.g. Gouda)
v. High salt varieties Cheese (e.g. Feta, Domiati)
vi. Pasta -filata Cheese (e.g. Mozzarella, Provolone)

2.3 <u>Health Benefits of Cheese:</u>

Cheese is widely recognized as a highly nutritious food product due to its rich composition of essential nutrients. As a dairy-based product, cheese serves as an excellent source of high-quality milk proteins, calcium, phosphorus, fat-soluble vitamins, and concentrated energy (De, 2010). It also contains essential micronutrients such as magnesium, zinc, vitamin D, and vitamin K, which contribute to bone development, maintenance of bone health, and the prevention of osteoporosis (Ware, RDN and LD., 2017). Beyond its role in bone health, cheese consumption has been associated with several cardiovascular benefits. Studies have shown that it may help lower blood pressure (Hajjar *et al.,* 2003) and reduce the risk of heart disease by positively influencing plasma lipid profiles (Jacqmain *et al.,* 2003). Additionally, Van Mierlo *et al.* (2006) reported that cheese contains cardio-protective nutrients that protect against vascular calcification, thereby contributing to healthier blood vessels and reducing the risk of hypertension (Dietary Guidelines Advisory Community, 2014). Cheese whey, a byproduct of cheese production, has also demonstrated significant health benefits. An experimental study involving the inoculation of cheese whey in rats revealed an increase in packed cell volume (PCV), total white blood cell (WBC) count, as well as lymphocyte and monocyte counts (Adebolu and Olorunfemi, 2016). These findings suggest that cheese whey may play a role in addressing anemia by enhancing red blood cell production. Cheese is also a valuable source of vitamin A, which plays a crucial role in immune function. Vitamin A contributes to antibody production, hematopoiesis, and the proper functioning of T and B lymphocytes, natural killer (NK) cells, and neutrophils (Semba *et al.,* 1998). Additionally, McLaren and Frigg (2001) reported that vitamin A and its metabolites function as gene transcription modulators. Swiss cheese, in particular, is an excellent source of vitamin B12, an essential nutrient for red blood cell production, protein synthesis, and DNA formation, as well as for supporting cognitive functions and mental health (Miller, 2017). Moreover, the high calcium content in cheese has been linked to a reduced risk of colorectal cancer (Miller, 2017).

Dental health is another area where cheese has shown positive effects. A study conducted by dental professionals found that moderate cheese consumption increases the pH level in dental plaque, reducing its acidity and offering protection against cavities and dental caries (Ware, RDN and LD., 2017). Cheese's anti-caries properties are attributed to its ability to elevate calcium concentrations in dental plaque. Furthermore, a study published in the Journal of Translational Medicine (2022) reported that cheese consumption enhances salivary alkalinity, which, when combined with its nutrient composition, reduces cavity formation, prevents tooth demineralization, and promotes remineralization (Lorenzini *et al.,* 2022). Cheese also contributes to gut health due to its probiotic content. A study published in Probiotics and Antimicrobial Proteins (2019) identified Lactobacillus casei and Lactobacillus fermentum in mozzarella cheese, both of which promote intestinal health and strengthen the immune system (Souza *et al.,* 2019). Additionally, Kaushik *et al.* (2019) highlighted cheese's immunostimulatory properties, suggesting that regular consumption may enhance immune function in healthy individuals.

Overall, cheese is a nutritionally dense food with multifaceted health benefits, ranging from bone and cardiovascular health to immune and dental well-being.

2.4 Cheese Manufactured in Bangladesh:

There are endless varieties of cheese existed at all corner of the world. Such as- Feta, Mozzarella, Cheddar, French brie, Italian gorgonzola etc. Though Europe is the biggest collector of cheese, Asia is the biggest user or consumer of cheese. As Bangladesh is the part of Asia, it has its own share of delicious salted cottage cheese (Das, 2019). In last few decades the popularity of cheese was not up to the expected level. But The cheese is getting popularized day by day (Proma, 2016). They become very popular to the consumer because of their taste and quality (Proma, 2016). That means, cheese gradually takes a position in Bangladeshi household (Das, 2019). Due to meet up the upward want of cheese, some dairy processing companies have taken initiatives to produce cheese (Proma, 2016). Five variants of cheeses such as sliced, spread, cubes, Austagram and Shredded Mozzarella have been introduced in market by Arong Dairy. Mainly, Austagram cheese and Mozzarella cheese are produced in Bangladesh (Das, 2019).

2.4.1 Mozzarella Cheese

Mozzarella is a well-known cheese which is soft, smooth and flexible in nature. It is also a long-standing cheese having the quality to make dish more palatable. According to codex standard for cheese (Joint FAO/WHO, 2006), Mozzarella cheese possesses a fibrous protein structure and the structure is long stranded without presence of curd flecks. Traditionally, buffalo's milk is used to prepare mozzarella cheese and typically cow's milk is used to manufacture mozzarella cheese. Mozzarella cheese is made from pasteurized milk. It is fall into the category of unripened cheese since there is no ripening step related in its manufacture. It is lack of rind that means it is a fresh cheese and can be given different shapes. Depending on animal's diet, the color of mozzarella cheese may be white to light yellow color (Lambert, 2008) Based on moisture content, there are two types of mozzarella cheese. If there is high moisture content it is considered as soft cheese. As soft cheese has high moisture content, there is overlying layer on cheese which may create pockets filled with milky liquids. According to USDA specifications for mozzarella cheese, high moisture mozzarella cheese has more than 52% moisture content but not more than 60% moisture content. Mozzarella cheese which has low amount of moisture is known as hard cheese. According to USDA specifications for mozzarella cheese, low moisture mozzarella cheese has 45-52% moisture content. These cheeses have no holes and they can be used for shredding. Mozzarella cheese is one kind of pasta filata cheese. In pasta filata processing technique, the curd is heated with a particular pH and then the curd is kneaded and stretched until it becomes smooth and free of lumps.

Some brands of mozzarella cheese that are available in Bangladesh are stated below-

1. Pran Goodlife Mozzarella Cheese,
2. Pran Classic Mozzarella Cheese,
3. Hazrat Ali Mozzarella Cheese,
4. Aarong Dairy Shredded Mozzarella Cheese,
5. Danscorella Shredded Mozzarella Cheese,
6. Yummy Mozzarella Cheese,
7. Kraft Fat Free Mozzarella Shredded Cheese,
8. Arla Mozzarella Cheese Block,
9. Lactima Mozzarella Cheese,
10. Grand'Or Mozzarella Cheese,
11. Emborg Mozzarella cheese,
12. Quality Mozzarella Cheese,
13. Italian Mozzarella cheese,
14. Kraft Mozzarella String Cheese,
15. Floridia Mozzarella Cheese etc.

2.4.2 Process of Preparation of Mozzarella Cheese:

Figure 1. depicts the general processes taken throughout the manufacturing process of mozzarella cheese.

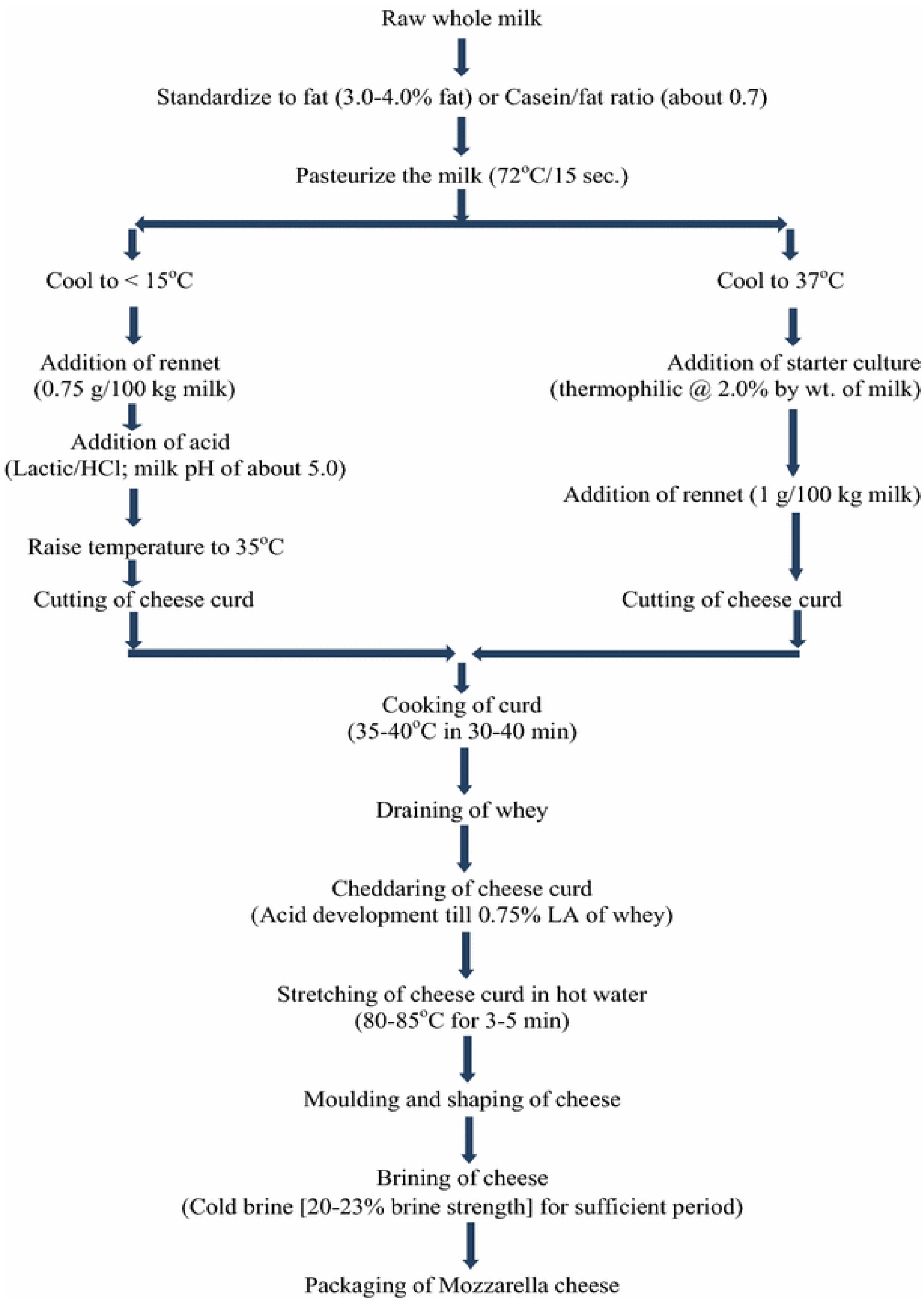


Figure 1: Flow chart for making mozzarella cheese.

Source: Patel *et al*. (1986); Ghosh and Singh (1996); Jana and Tagalpallewar (2017)

2.5 Assessment of cheese quality:

Quality assessment of cheese is defined as the evaluation of cheese quality to determine whether the cheese meets its ultimate quality or not. There is two dimension of quality assessment (objective and subjective dimension). Objective dimension refers to measure the physio-chemical properties of a product like protein, fat, ash, moisture, salt, vitamin, lactose content etc. Subjective evaluation of food depends on consumer expectations, perceptions and acceptance including color, flavor, texture of food. Cheese's quality characteristics can be divided into several groups such as microbial, physical, chemical etc. (Gunasekaran, 2016). Papetti and Carelli (2013) carried a study on composition and sensory analysis for quality evaluation of Italian Cheese. Chemical and sensory analysis of mozzarella cheese was conducted by Mijan *et al*. (2010). Aldrete *et al*. (2014) conducted microbial analysis of an Artisanal Mexican cheese which revealed a relationship between microbial flora and flavor of cheese.

2.5.1 Sensory evaluation of cheese:

The practice of using the five human senses such as smell, taste, touch, hearing and sight to evaluate the characteristics of food products is knowns as sensory evaluation. Several methods have been developed for the sensory evaluation but dairy industry use conventional method for sensory evaluation (Singh *et al*., 2003). In cheese research, one of the most powerful sensory tools is descriptive sensory analysis. In the literature, different types of descriptive sensory method have been reported. They are- texture profile method (Giongo, *et al*., 2019), quantitative descriptive analysis (Stone *et al*., 2004), quantitative flavor profiling (Kindstedt and Fox, 1993) etc. Fenelon *et al*., (2000) conducted a study on determination of sensory properties of low-fat cheddar cheese by descriptive analysis method. He found that the amount of fat content influenced the flavor characteristics of cheese. Adhikari *et al*. (2003) suggested that low fat cheeses are dry and crumbly where full fat cheese are more buttery, creamy and caramel like in nature. Virgili *et al.* (1994) conveyed a study on sensory-chemical relationships in Parmigiano-Reggiano Cheese. Another study was conducted by Bhattarai and Acharya (2010) on the quality evaluation of mozzarella cheese. In this study, the cheese samples were evaluated by nine judges using a nine-point hedonic rating scale where the used five attributes such as appearance, taste, flavor, texture and overall acceptability according to (Ranganna, 2000). Mijan *et al*., 2010 used a score card for the evaluation of cheese quality. American Dairy Science Association (ASDA) offered this score card for cheese to judge ((De, 2010). According to Indian Standard, there was an evaluating card for sensory evaluation of panner. The evaluation card was on the basis of four attributes including flavor, body, texture, color & appearance and Packaging. A panel of judge could give by using this evaluation card. Based on total score of evaluation card panner could be categorized into four grades. The Paneer an overall score of 90 or higher is considered excellent/A, 80–89 is considered good/B, 60–79 is considered fair/C, and 59 or lower is considered poor/D. Kumar et al. (2014) conveyed same procedure of sensory evaluation on paneer. For the determination of consumer responses, one of the most vital sensory attributes is flavor of cheese (Sameen *et al*., 2008). Eshetu & Asresie, 2019 introduced sensory analysis of Ethiopian cheese by using a questionnaire. Here they used some attributes such as taste, color, aroma, texture, appearance and overall acceptability for sensory evaluation. Pedro Bárcenas *et al.,* 2007 Carried a study on sensory evaluation of cheese using the sensory attributes and standard references. The attributes included surface roughness, surface moisture, elasticity, firmness, friability, adhesiveness, solubility and moisture in mouth. The main purpose of all studies was to evaluate whether there were any significant differences in terms of flavor, taste, body texture, colors. In short, sensory evaluation helps in making products the best the producers can be. This type of evaluation can be used to understand the defects, describe and quantify positive characteristics which increases liking of the products. Actually, the sensory methodology used to eliminate carryover and order of tasting bias (Muir and Hunter, 1991).

2.5.2 Review about chemical composition analysis of cheese:

El Owni and Osman (2009) conducted a study on evaluation of chemical composition of mozzarella cheese where two different methods of processing were used. They evaluated protein content(%), total solids content(%), fat content(%), moisture content(%), ash content(%), lactose content(%) of mozzarella cheese. In their study, it showed that mozzarella cheese contained fat(%) 27.25 ± 0.82; protein(%) 20.06 ± 0.65; total solids (%) 51.42 ± 1.32; moisture(%) 48.59 ± 1.32; ash(%) 2.25 ± 0.07; titrable acidity(%) 0.66 ± 0.02; lactose(%) 1.59 ± 1.35.

A study on the chemical composition assessment of buffalo mozzarella was conducted by Goncalves *et al*. 2017. They determined Protein content, acidity content, moisture content, ash contet, FDE, DDE, TDE of mozzarella cheese. Rudan *et al.* (1998) also determined pH, total nitrogen content, moisture content, salt content, fat content and calcium content of cheese. In their study they evaluated salt cncentration by Volhard test, fat content by Babcock test and calcium content by plexometric titration. Johnson and Olson (1985) determined the ammount of sodium chloride content present in cheese my applying Mohr's method. Mijan *et al.* (2010) tested the quality of mozzarella cheese. He calculated moisture (g/kg), TS (g/kg), lactose (g/kg), acidity (g/kg), fat (g/kg), protein (g/kg) and pH content of mozzarella cheese. Physical and chemical properties of mozzarella cheese analogue microwavable was determined by Abdul *et al.* (2016).

2.5.3 Review about Microbial quality of cheese:

Dairy products, particularly cheese, serve as an optimal medium for microbial proliferation due to their nutrient-rich composition. The microbial profile significantly influences the quality and safety of mozzarella cheese, with milk being the primary source of microbial contamination. Contamination may occur at various stages, including milk collection, processing, handling, and post-production storage. To ensure consumer safety, a comprehensive microbiological assessment is essential, with a specific focus on Escherichia coli (E. coli) concentration and total viable count (TVC). The physicochemical properties of cheese directly affect microbial growth; therefore, imposing rigid quantitative microbiological standards is often impractical (Ardic, 2003). Losito *et al.* (2014) conducted a study to assess the microbiological safety and quality of mozzarella cheese, evaluating three key parameters: total viable count (TVC) using plate count agar, coliform count using Tryptone Bile-X-Glucuronide Agar (TBX), and lactic acid bacteria concentration using the plate count method on De Man, Rogosa, and Sharpe (MRS) agar. Kumar *et al.* (2014) also employed standard plate count and coliform count techniques to analyze the microbiological quality of cheese. Similarly, Kunova *et al.* (2015) utilized violet red bile agar for the detection of coliform bacteria in cheese samples and applied 10^{-3} and 10^{-4} dilutions to quantify microbial presence. Furthermore, Nur *et al.* (2021) employed the pour plate method to determine the total viable bacterial count (TVBC) and total coliform count (TCC) in milk and dairy products, including cheese. Dina and Elsherif (2015) conducted a comprehensive microbiological evaluation of mozzarella cheese, examining TVC, TCC, yeast and mold counts, and the presence of E. coli, Staphylococcus aureus, and Salmonella. Their study reported an average total bacterial count of 3.84×10^6 CFU per gram of cheese, highlighting the need for stringent microbiological monitoring in dairy products.

2.6 Conclusion:

The cheese industry in Bangladesh is experiencing significant growth due to the increasing demand for cheese. As the sector expands, it is essential to assess the nutritional and microbiological quality of cheese available in the local market. This study aims to enhance understanding of the nutritional composition of cheese and its potential therapeutic applications in addressing malnutrition-related health conditions in Bangladesh. Additionally, this investigation will evaluate the extent to which domestic cheese producers comply with the standards and regulations set by the Bangladesh Standards and Testing Institution (BSTI), ensuring product quality and consumer safety.

Chapter 3: Materials and Methods

3.1 Site of the study:

The experiment was conducted in the three different laboratories of Chattogram Veterinary and Animal Sciences University. Chemical analysis of cheese was carried away in Dairy Science Laboratory and Animal Science and Nutrition Laboratory. Microbial analysis of cheese was performed in the Department of Microbiology and Veterinary Public Health Laboratory. From January 1 to June 30, 2024, a six-month experiment was run.

3.2 Collection of Sample:

Four different brands of mozzarella cheese samples were used in the study. All the samples were obtained from superstores of Chattogram Metropolitan area (CMA) such as Basket, Agora, Utsav, Swapno, Khulshi mart, Mina Bazar. Two of the brands of cheese came from renowned dairy producers in Bangladesh. Another two brands of cheese were from foreign dairy producers. For improved results, three batches of each sample were run.

3.3 Labelling of Sample:

The study analyzed four different brands of Mozzarella cheese, labeled as Cm_1, Cm_2, Cm_3, and Cm_4. Among them, Cm_2 and Cm_3 were sourced from two distinct cheese manufacturing companies in Bangladesh, while Cm_1 and Cm_4 originated from internationally recognized cheese manufacturers. Specifically, Cm_1 represents Grand Or Mozzarella Cheese, Cm_2 corresponds to Good Life Mozzarella Cheese, Cm_3 refers to Hazrat Ali Mozzarella Cheese, and Cm_4 denotes Lactima Mozzarella Cheese.

3.4 Sensory Evaluation of Cheese:

Sensory evaluation of mozzarella cheese was done by using a score card. All the manufacture labels were taken off. The samples were served on plate to judge and they were labelled with an exclusive identification code to eliminate testing bias as there was no possibility to know the brand or company of cheese. The evaluation was carried by the two renowned instructors of CVASU while the temperature was kept at room temperature. The honorable instructors rinsed their mouth with plain water before testing each sample and marked the samples in a provided organized score card. The attributes that were included in the scorecard for sensory evaluation were color, body, texture, finish and appearance, flavor and aroma.

<u>Score Card</u> (Source: De, 2010)

Terms	Marks
Color	10
Body	15
Texture	15
Flavor and aroma	45
Finish and appearance	15

3.5 <u>Chemical Analysis of Cheese:</u>

3.5.1 Preparation of Sample

By following AOAC method No. 955.30 (Bradley, 2023), the cheese sample were prepared. Firstly, the outer part (rind, smear, or moldy surface layer) of the cheese was taken aside. The thickness of the removed rind was equivalent with thickness of the rind that is normally removed before consuming cheese. For taking the sampling portion, the sample was mixed, grinded properly by stirring and kneading. By using a spoon or spatula, a segment of well mixed cheese was taken out. The sample that could not tested after pre-treatment was stored in an air-tight container at a temperature of (10-12) °C until further experiment or test.

3.5.2 Determination of Dry matter

Dry matter of cheese sample was determined according to AOAC method No. 926.08 (Bradley, 2023). The Petri dishes were washed, cleaned and dried properly for use. Each petri dish was weighted and marked to ensure accuracy. 10 gram of cheese sample from each brand was weighted and placed in each petri dish. The sample was dried in hot air oven to remove all the moisture from cheese sample at 105°C for 24 hours. The time was properly maintained. After drying each sample completely, the petri dishes were transferred to desiccator for cooling down properly. Sample was weighted again and the loss of weight as moisture was recorded. The process was repeated every 6 hours till constant weight were attained. Then, the final weight of each petri dish containing dried sample was recorded.

The following formula was then used to get the dry matter content.

$$\% \text{ of Dry matter } = \frac{W_d - W_e}{W_s} \times 100$$

W_s = Weight of sample

W_e = Weight of empty petri dish

W_d = Weight of petri dish containing dried sample

3.5.3 Determination of Moisture content

Moisture content of cheese sample was determined according to AOAC method No. 926.08 (Bradley, 2023). The Petri dishes were washed, cleaned and dried properly for use. Each petri dish was weighted and marked to ensure accuracy. 10 gram of cheese sample from each brand was weighted and placed in each petri dish. The sample was dried in hot air oven to remove all the moisture from cheese sample at 105°C for 24 hours. The time was properly maintained. After drying each sample completely, the petri dishes were transferred to desiccator for cooling down properly. Sample was weighted again and the loss of weight as moisture was recorded. The process was repeated every 6 hours till constant weight were attained. Then, the final weight of each petri dish containing dried sample was recorded.

The following formula was then used to get the moisture content.

$$\textit{\% of moisture content.} = \{1 - \frac{W_d - W_e}{W_s}\} \times 100$$

Ws = Weight of sample

We = Weight of empty petri dish

Wd = Weight of petri dish containing dried sample

3.5.4 Determination of fat content

Fat content of cheese sample was done by following Gerber method as it is a primary, historic, speedy and accurate chemical test to determine fat of milk and milk products.

3.5.4.1 Requirement

Butyrometer, 1ml and 10ml pipette, water bath, centrifuge machine, hot water, H_2SO_4 (specific gravity 1.82), amyl alcohol.

3.5.4.2 Procedure

Each butyrometer was cleaned and dried properly. 1gram of each brand of sample was taken in each butyrometer with the of spatula or glass rod. Then, 10.75 ml of hot water (temp. 60°C - 70°C) was added to the butyrometer and Shaked. After that, 10ml of H_2SO_4 to dissolve cheese fat and 1ml of Amyl alcohol to break the emulsion were added to the butyrometer. Cork or stopper was inserted to close the butyrometer properly. The mixture of the butyrometer was inverted for few times by applying gentle swirling motion. Then, the butyrometer was transferred in a hot water bath at 70°C for 10-minute with periodical removal and inverting. The Butyrometer was centrifuged in centrifugal machine at 100 r.p.m for 5 minute. The reading of fat column was recorded.

$$\text{Fat (\%) of Cheese} = \frac{\text{Observed reading} \times 11.25}{\text{Weight of cheese}}$$

3.5.5 Determination of Crude Protein

According to Kjeldahl method narrated in IDF20-1 (2014), the crude protein content of cheese sample was calculated.

3.5.5.1 Requirement

Kjeldahl digestion unit, Conical flask, burette, pipette, 98% H_2SO_4, catalyzer, distilled water, 0.1N HCl, 40% NaOH, 4% boric acid, methyl red indicator.

3.5.5.2 Procedure

3.5.5.2.1 Protein Digestion

2gram of cheese sample from each brand was weighed and placed them in Kjeldahl digestive tube. Then 20ml of 98% H_2SO_4 and Kjeldahl catalyst (K_2SO_4, $CuSO_4$) were added into the digestive tube. Digestive tube was transferred to Kjeldahl digestion unit and digested with heating for 3 hours with occasional shaking. Then, the mixture was allowed to cool at room temperature.

3.5.5.2.2 Protein Distillation

After cooling, 50ml of distilled water was added to the digested mixture. After adding water, 10ml of mixed indicator was put into a conical flask. Here, the mixed indicator was made by adding 10ml of Boric acid solution with 2 drops of methyl red indicator. Then, digestive tube (containing diluted digested mixture) and conical flask (holding mixed indicator) were transferred to distillation unit. Before using distillation unit, it was steamed for 15 minutes. 40% NaOH was added mechanically to the digestive mixture from the flask containing NaOH connected to distillation unit. The distillation vapor was collected in conical flask which liberated Ammonia by chemical reaction and distillation.

3.5.5.2.3 Titration

Titration of the distillate was done against 0.1N HCl solution and the result was recorded.

It is essential to calculate N2 content of the sample for finding out the crude protein content. Following formula was used to derive N2 content.

$$\text{Total Nitrogen (\%)} = \frac{14.007 \times (T - B) \times N \times 100}{W_s}$$

Where,

W_s = Weight of sample

N = Normality of Standard HCl

B = Volume of Standard HCl (in ml) used in titration for blank determination

T = Volume of Standard HCl (in ml) used in titration for test sample

Protein content was determined by multiplying the nitrogen content by a factor of 6.38.

$$\text{Crude Protein (\%)} = \text{Total nitrogen (\%)} \times 6.38$$

3.5.6 Determination of ash content

Determination of ash content in cheese sample was carried out following AOAC method (1965). The crucible was cleaned and dried properly. Weight of empty crucible was taken. (2 – 5) gram of sample from each brand of cheese were weighed accurately and taken them in each crucible. The crucibles with samples were heated in an oven at 100°C to evaporate sample moisture. Then, the crucibles were placed into the pre-heated muffle furnace at 600°C. It was continued till white or light color, moisten with few amounts of moisture to liquefy salt, then the crucibles were dried in an oven. The process was repeated. The crucibles were transferred into a desiccator to cool the sample. The final weight of crucible with dried sample was taken.

The formula for determination of ash content is stated below-

$$\text{Percentage of ash (dry basis)} = \frac{(W_3 - W_1) \times 100}{W_2 - W_1}$$

Where,

W_1= initial weight of empty crucible

W_2= weight of crucible containing original sample before ashing

W_3= final weight of crucible containing dried sample after ashing

3.5.7 Determination of acidity content

Th acidity content of cheese sample was calculated by using AOAC (1995) method 920.124.

3.5.7.1 Requirement

Conical flask, 0.1N NaOH, 0.5% phenolphthalein, glass rod, burette, pipette, distilled water.

3.5.7.2 Procedure

Conical flask was cleaned and dried properly. Two grams of cheese sample was weighed accurately and transferred into conical flask. 20 ml distilled water (temp. 60°C - 70°C) was added to conical flask. After adding water, a fine paste was prepared by using glass rod. Then, 2 drops of 0.5% phenolphthalein indicator were added to the fine paste. Titration of the mixture was done against 0.1N NaOH solution and stirred the mixture till faint pink color persisted. The burette reading was noted down.

Acidity (%) was calculated according to following formula-

$$\text{Acidity (\%)} = \frac{0.009 \times V \times 100}{Ws}$$

Ws= Weight of sample

V= Volume of 0.1N NaOH required for titration

3.5.8 Determination of salt content (NaCl) of cheese

By applying Mohr's method (IS: 3507 – 1966), salt content of cheese was performed.

3.5.8.1 Requirement

Conical flask, burette, pipette, Potassium chromate, distilled water, Silver Nitrate.

3.5.8.2 Procedure

Conical flask was cleaned and dried properly. (3 – 5) gram of cheese sample was weighted accurately and put them into conical flask. 50ml distilled water was added to conical flask and Shaked it. Shaking of conical flask continued for 30 minutes. After swirling, 1ml of potassium chromate as an indicator was added to the mixture. The mixture was titrated against 0.1N Silver Nitrate solution. Orange coloration indicated the end point of titration which existed for 30 seconds. The burette reading was taken.

Na Content was calculated according to given formula-

$$\text{NaCl (\%)} = \frac{5.844 \times C \times (V_2 - V_1)}{Ws}$$

Where,

W_s= weight of sample

V_1= Volume of Silver Nitrate solution used in titration for blank determination

V_2= Volume of Silver Nitrate solution used in titration for test sample

C= Concentration of silver nitrate solution (in mol/L)

3.6 Microbial Analysis of Cheese

Total Coliform Count (TCC) and Total Viable Count (TVC) were done to determine microbial quality of cheese sample. Plate count agar was used for Total Viable Count Total coliform count was done in VRB agar. A temperature of 37°C for 24hour is used for incubation in the incubator.

3.6.1 Preparation of Butterfield's Phosphate Diluent

As a part of microbial analysis, Butterfield Phosphate Diluent was used to provide a standardized medium for the preparation of microbiological sample dilution blanks.

3.6.2 Procedure for the preparation of stock solution

500ml of purified water was taken. 34gram of potassium di hydrogen phosphate was added into the water and dissolved. 1N NaOH was prepared and then 175ml of 1N NaOH solution was mixed with KH_2PO_4 solution. More distilled water was added until the volume of the solution would be 1000ml. pH paper was used to ensure the optimum pH (7.2) of the solution. The solution was sterilized at 121°C for 15 minute and stored in refrigerator.

3.6.3 Preparation of Dilution Blank

1.25ml of above stock solution was taken and added to 1ml of distilled water. Falcon tubes and Dilution blanks were properly cleaned and dried. 90ml diluents was dispensed in each dilution blank and 9ml diluents was dispensed in each falcon tubes. The falcon tubes and dilution blanks were capped loosely before sterilization so that a portion of diluent was lost during sterilization. Then, the dilution blank and falcon tubes were sterilized at 121°C for 15minute.

3.6.4 Preparation of agar media

Following the manufacturer's direction, the media were prepared. For preparation of plate count agar media, 23.5-gram plate count agar was suspended in 1L sterilized distilled water. In case of preparation of VRB agar media, 41.53-gram VRB agar was suspended in 1L sterilized distilled water. The media ware heated to boiling to dissolve.

Then, they were sterilized in autoclave at 121°C for 15sec at 15lbs pressure. After sterilization, the media were transferred to hot water bath at (45 -50) °C until used. They were swirled well and poured into sterile petri plates. After that, the petri plates were kept to solidify. It is recommended that the temperature of hot water bath should not exceed 50°C.

3.6.5 Calculation of TVC

- 10 gram of cheese sample was weighted and homogenized in 90ml diluents and made suspension in a beaker. It was considered as Primary or Original Dilution Blank (PDB).
- From that, 1ml of PDB was transferred to falcon tube no 1 mixed finely.
- From first falcon tube 1ml of sample was moved to second falcon tube and it was continued up to last one and 1ml was discarded from the last falcon tube. That means, the sample was diluted by a factor of 10, 100, 1000, 10000 etc.
- For each tube, three petri dishes were taken.
- 1ml of each dilution mixture was transferred from each falcon tube to petri dishes. One pipette was used for one tube. Tips of test tube was touched gently to the media.
- Each petri dishes were poured with plate count agar and they had been cooled to 45°C to 50°C temperatures. For proper mixing, the plates were revolved and kept them to solidify.
- Dilution factor, sample no and date were labelled on each petri dish. The petri dishes were incubated at 37°C for 24 hours at inverted position to facilitate viable bacterial growth.
- After (1 – 3) days interval of incubation period, the colonies were observed.
- Colony counter was used to calculate the number of colonies per plate.
- Calculation: Total No. of bacteria = average No. of colonies × Dilution factor.

3.6.6 Enumeration of total coliform count

- 10 gram of cheese sample was weighted and homogenized in 90ml diluents and made suspension in a beaker. It was considered as Primary or Original Dilution Blank (PDB).
- From that, 1ml of PDB was transferred to falcon tube no 1 mixed finely.

- From first falcon tube 1ml of sample was moved to second falcon tube and it was continued up to last one and 1ml was discarded from the last falcon tube. That means, the sample was diluted by a factor of 10, 100, 1000, 10000 etc.
- For each tube, three petri dishes were taken.
- 1ml of each dilution mixture was transferred from each falcon tube to petri dishes. One pipette was used for one tube. Tips of test tube was touched gently to the media.
- Each petri dishes were poured with VRB agar and they had been cooled to 45°C to 50°C temperatures. For proper mixing, the plates were revolved and kept them to solidify.
- Dilution factor, sample no and date were labelled on each petri dish. The petri dishes were incubated at 37°C for 24 hours at inverted position to facilitate viable bacterial growth.
- After 1 day interval of incubation period, the colonies (dark red colonies) were observed.
- Colony counter was used to calculate the number of colonies per plate (Dark-red colonies with reddish zone of precipitated bile having an estimated area of 0.5mm or more were considered as coliform colonies). Plates in which colony growth were not observed; they were not calculated.
- Calculation:

No. of coliforms per gram = average No. of coliform colonies × Dilution factor

3.6.7 Confirmation of presence of *E. coli* in cheese sample

Some petri dishes containing EMB agar were prepared. Subsequent colonies were taken from VRB agar and streaked on the petri dishes. The petri dishes were incubated at 37°C for 24 hours. Presence of E. coli in cheese sample was confirmed by observing metallic sheen on bacterial colony in the petri dishes.

3.6.8 Statistical Analysis

To conduct additional statistical analysis, all types of data were entered into Microsoft Office Excel 2013 version spread sheet. Sensory evaluation, physical and chemical composition of cheeses were evaluated using one-way ANOVA procedures in order to generate a 95% interval for the degree of significant variances.

Chapter 4: Results

4.1 Sensory Evaluation

Sensory evaluation is a crucial aspect of determining the quality of mozzarella cheese. This process involves assessing various attributes, including color, body, finish and appearance, texture, and flavor/aroma. Trained panellists evaluate these attributes using a standardized scoring system, where higher scores indicate better quality. This evaluation provides insights into the impact of different formulations or processing conditions on the sensory characteristics of mozzarella cheese.

4.1.1Sensory Attributes and Statistical Analysis

The sensory evaluation of mozzarella cheese samples revealed significant differences in body, finish & appearance, texture, and flavor & aroma, whereas color remained statistically non-significant. Cm_4 consistently outperformed the other samples, suggesting that its formulation or processing method may contribute to superior sensory attributes. The sensory attributes of mozzarella cheese samples (Cm_1, Cm_2, Cm_3, and Cm_4) were analyzed, and the mean scores with standard deviations are presented in Table 4.1. The statistical significance of differences among the samples was determined using p-values.

Table 1: Sensory Evaluation Scores (Mean ± SD) of Mozzarella Cheese

Variables	Full score	Sample				P Value	Level of Sign.
		Cm_1(A)	**Cm_2(B)**	**Cm_3(C)**	**Cm_4(D)**		
Color	10	7.867 ± 0.321^{a}	8.000 ± 1.000^{a}	7.767 ± 0.462^{a}	8..737± 0.251^{a}	0.247	NS
Body	15	11.333 ± 0.444^{bc}	10.153 ± 0.194^{c}	12.277 ± 1.109^{ab}	13.360 ± 0.445^{a}	0.002	**
Texture	15	13.133 ± 0.188^{a}	10.500 ± 0.250^{b}	12.270 ± 0.200^{a}	11.850 ± 1.008^{ab}	0.002	**
Flavor and aroma	45	36.030 ± 1.840^{b}	30.917 ± 0.520^{c}	37.380 ± 2.300^{ab}	41.680 ± 1.850^{a}	0.001	***
Finish and Appearance	15	12.920 ± 1.377^{a}	7.657 ± 1.469^{b}	11.310 ± 0.637^{a}	13.277 ± 0.254^{a}	0.001	***

Sensory Evaluation Scores of Mozzarella Cheese

Legends: Values in the same row with the same superscripts but different Means ± SD are not statistically significant (P≤0.05)

**Statistically significant at P≤0.01, ***Statistically significant at P≤0.001, NS = statistically not significant, SD = Standard Deviation

4.1.1.1 Color:

The color scores ranged from 7.767 to 8.737, with no statistically significant differences ($p = 0.247$), indicating that all samples had comparable color attributes.

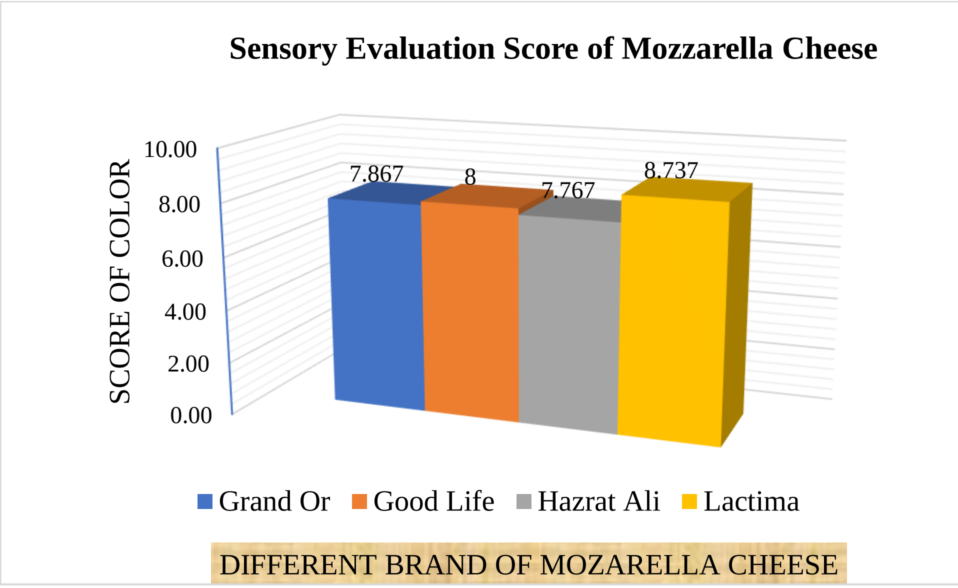


Figure 2: Sensory Evaluation of Mozzarella Cheese (Score of color)

4.1.1.2 Body:

A significant difference ($p = 0.002$) was observed, with Cm_4 having the highest score (13.360), suggesting a firmer and more desirable body.

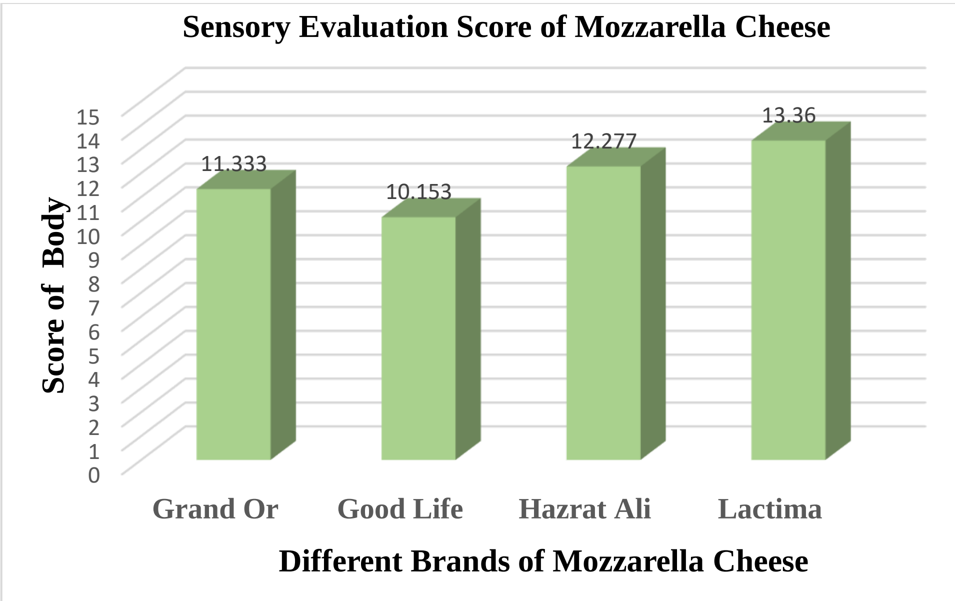


Figure 3: Sensory Evaluation of Mozzarella Cheese (Score of body)

4.1.1.3 Texture:

A significant variation ($p = 0.002$) was observed, with Cm_1 scoring the highest (13.133), implying better texture characteristics.

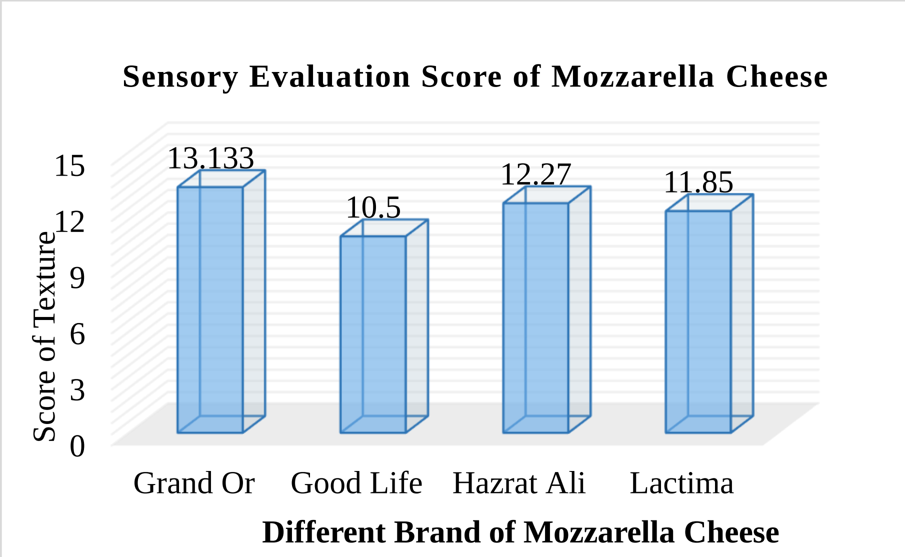


Figure 4: Sensory Evaluation of Mozzarella Cheese (Score of Texture)

4.1.1.4 Flavor & Aroma:

The most critical attribute, flavor & aroma, was highly significant ($p = 0.001$), with Cm_4 achieving the highest score (41.680), indicating superior sensory quality.

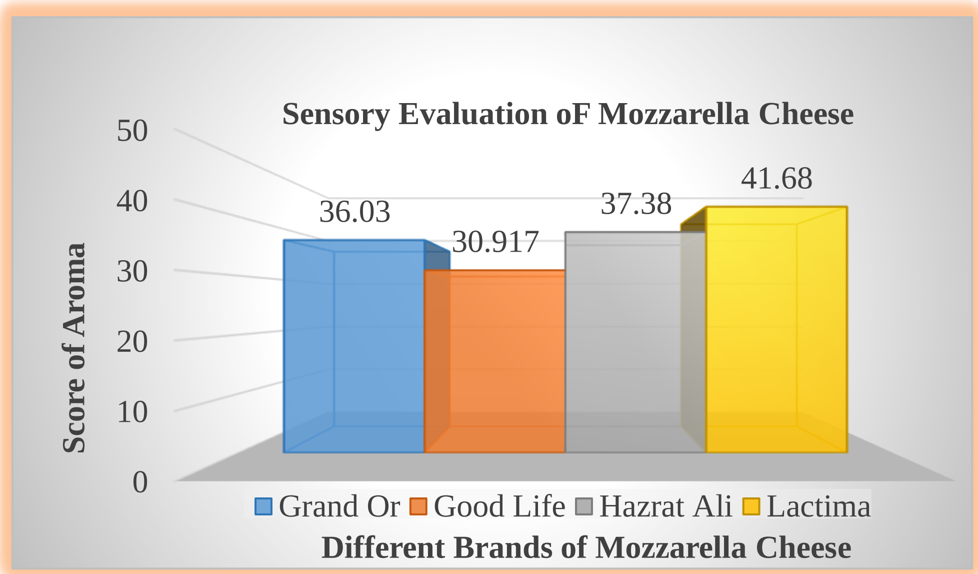


Figure 5: Sensory Evaluation of Mozzarella Cheese (Score of flavor and aroma)

4.1.1.5 Finish & Appearance:

This attribute showed a highly significant difference ($p = 0.001$), where Cm_4 had the best finish and appearance (13.277), while Cm_2 had the lowest (7.657).

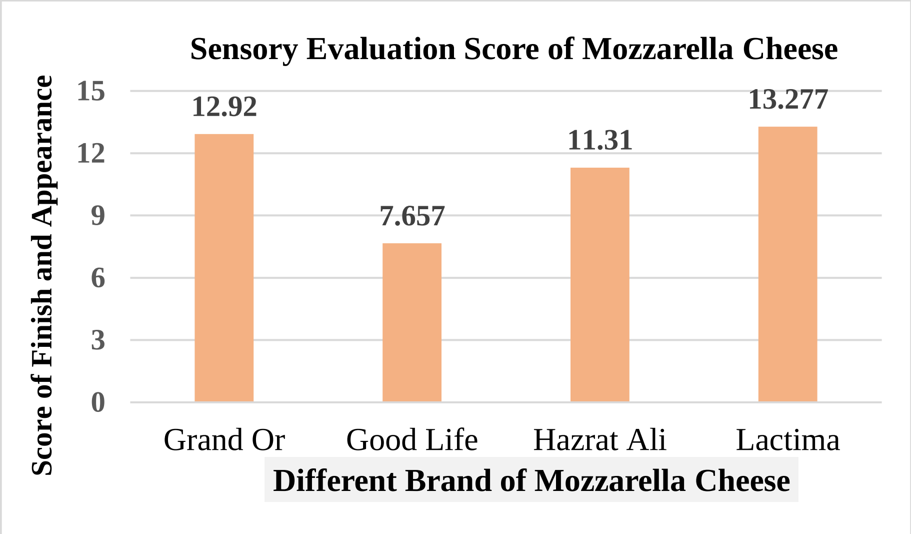


Figure 6: Sensory Evaluation of Mozzarella Cheese (Score of finish and appearance)

4.2 Nutritional attributes

Nutritional qualities of mozzarella cheeses were evaluated by looking at its moisture, dry matter, protein, crude fat, acidity, fiber, ash contents. The results of proximate analysis of four different mozzarella cheese are illustrated on table 2-

Table 2– Proximate Analysis (Mean ± SD) of Mozzarella Cheese of Mozzarella Cheese

Variables	Sample				P Value	Level of Sign.
	Cm_1(A)	**Cm_2(B)**	**Cm_3(C)**	**Cm_4(D)**		
Dry matter	50.907 ± 0.752^{a}	51.300 ± 0.542^{a}	50.920 ± 1.78^{a}	$47.017 \pm .0.873^{b}$	0.004	**
Moisture	49.093 ± 0.752^{a}	48.700 ± 0.542^{a}	49.080 ± 1.78^{a}	$52.983 \pm .0.873^{b}$	0.004	**
Protein	22.203 ± 0.758^{ab}	25.350 ± 2.210^{a}	21.383 ± 0.743^{b}	17.573 ± 0.611^{c}	0.001	***
Fat	22.363 ± 1.239^{a}	17.300 ± 1.026^{b}	16.58 ± 2.560^{b}	13.993 ± 0.990^{b}	0.001	***
Acidity	0.707 ± 0.127^{a}	0.520 ± 0.166^{a}	1.067 ± 0.379^{a}	0.527 ± 0.087^{a}	0.050	*
Ash	2.583 ± 0.382^{ab}	3.183 ± 0.355^{a}	1.833 ± 0.076^{c}	2.317 ± 0.126^{bc}	0.002	**
Salt	0.393 ± 0.137^{b}	0.910 ± 0.020^{a}	0.450 ± 0.304^{b}	1.060 ± 0.066^{a}	0.003	**

Proximate composition of different brands of mozzarella cheese

Legends: Values in the same row with the same superscripts but different Means ± SD are not statistically significant ($P \leq 0.05$)

*Statistically significant at $P \leq 0.05$ **Statistically significant at $P \leq 0.01$, ***Statistically significant at $P \leq 0.001$, NS = statistically not significant, SD = Standard Deviation.

4.2.1 Dry Matter:

Among the Four samples, the range of average percentage of dry matter was between (47.017 – 51.300) %. A one-way ANOVA revealed a statistically significant mean difference in dry matter content ($p = 0.004$). Cm_4 had a significantly lower dry matter content (47.017±.0.873) than others, which may suggest a higher moisture retention or altered composition in this condition. Cm_1 and Cm_3 had almost same amount of dry matter. This observation indicates that Cm4 may not be as effective in concentrating solid materials compared to Cm_1, Cm_2, and Cm_3, which could influence the product's final texture, stability, and shelf-life.

From the data table: dry matter (%) of different brands of Mozzarella Cheese is represented graphically in Figure 7:

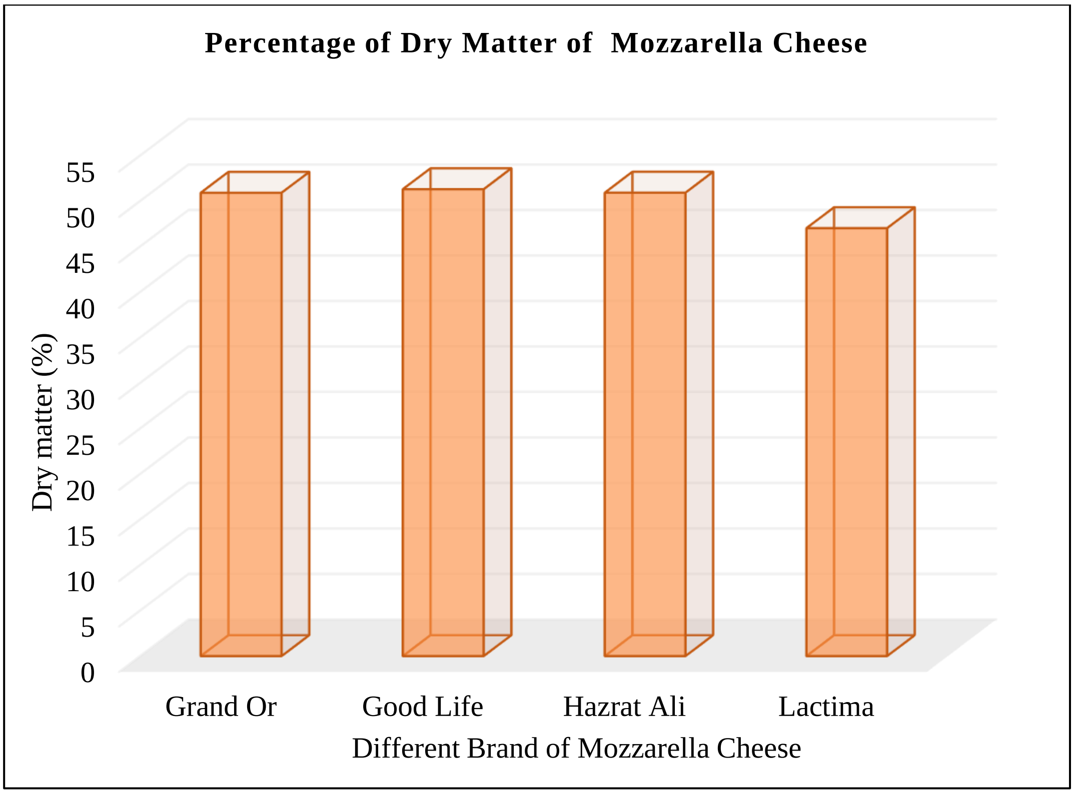


Figure 7: Percentage of dry matter of mozzarella cheese

4.2.2 Moisture Content:

Moisture content is inversely related to dry matter. Among the Four samples, the range of average percentage of moisture was between (48.700 – 52.983) %. Moisture content was found to differ significantly across conditions ($p = 0.004$). Cm_4 exhibited the highest moisture content (52.983±.0.873) among the samples, which is consistent with its lower dry matter value. Cm_2 has the lowest amount of moisture content (48.700±0.542). Cm_1 and Cm_3 have almost same amount of moisture content. This result suggests that Cm_4 may have retained more water during processing, potentially affecting the product's weight, texture, and preservation characteristics. In contrast, Cm_1, Cm_2, and Cm_3 exhibited relatively lower moisture levels, which could contribute to longer shelf-life and better texture stability in the final product.

From the data table: moisture (%) of different brands of Mozzarella Cheese is represented graphically in Figure 8:

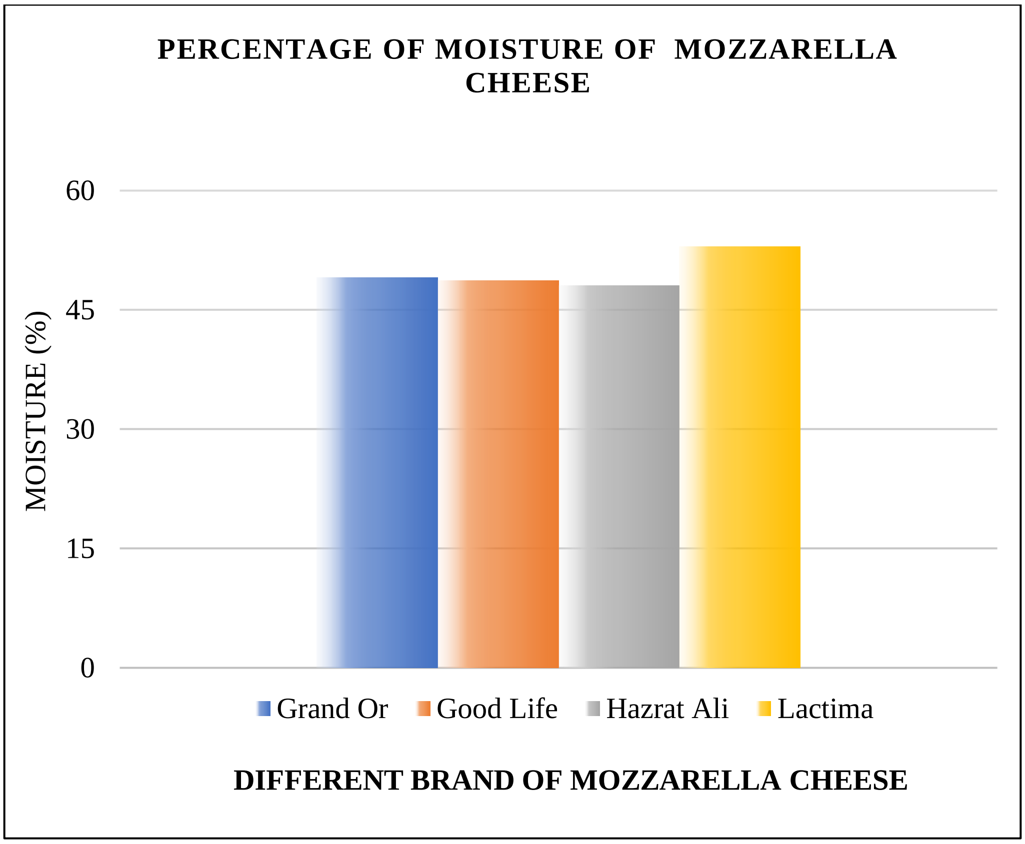


Figure 8: Percentage of moisture of mozzarella cheese

4.2.3 Protein:

Among the Four samples, the range of average percentage of crude protein content was between (17.573 – 25.350) %. The analysis indicated a significant difference in crude protein content ($p = 0.001$), with Cm_2 having the highest crude protein content (25.350±2.210) among the samples. This suggests that the conditions in Cm_2 were conducive to protein retention or enhancement. The significant drop in crude protein content observed in Cm_4 (17.573±0.611) could indicate the loss of protein during the process, possibly due to degradation or leaching into the medium. These findings suggest that Cm_2 may be particularly beneficial for applications requiring higher protein concentrations, such as in nutritional or dietary food products.

From the data table: crude protein content (%) of different brands of Mozzarella Cheese is represented via pie chart in Figure 9:

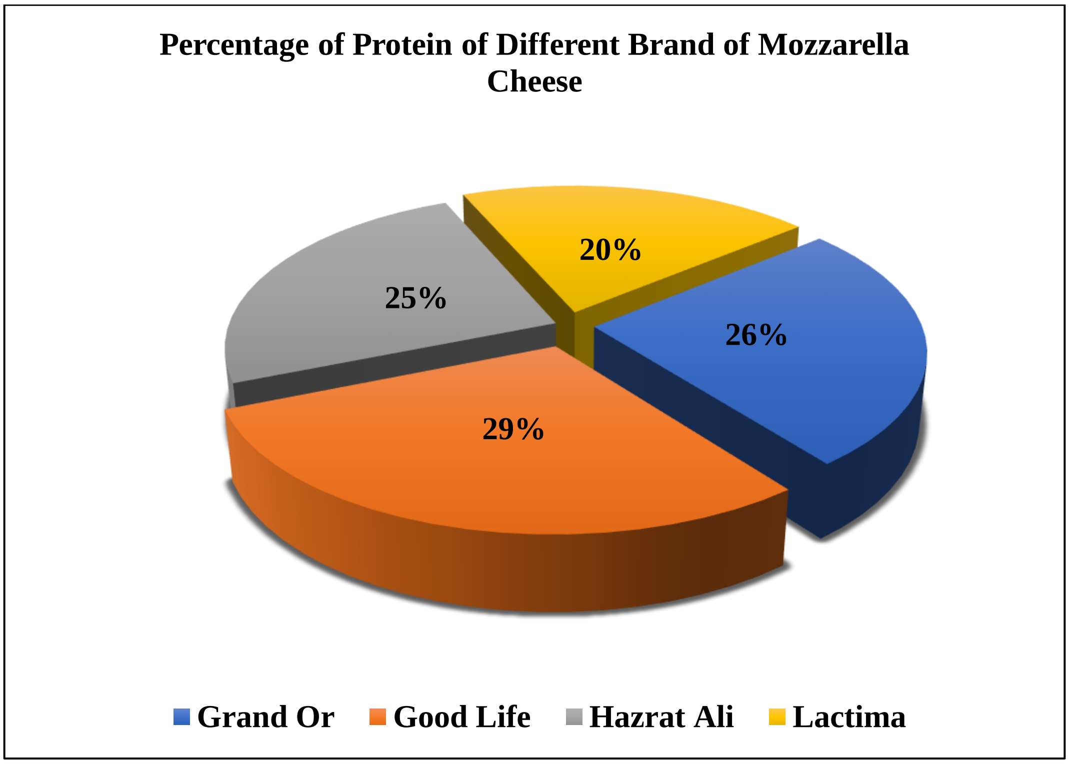


Figure 9: Percentage of crude protein of mozzarella cheese

4.2.4 Fat:

Among the Four samples, the range of average percentage of fat content was between (22.363 - 13.993) %. A significant difference in fat content was found ($p = 0.001$), with Cm_1 exhibiting the highest fat content (22.363±1.239), and Cm_4 showing the lowest (13.993±0.990). These results indicate that Cm_1 may have been the most effective at preserving fat, while Cm_4 might have led to the loss of fat content, potentially due to more aggressive processing or degradation. The variation in fat content is important, especially in food processing, as fat plays a crucial role in texture, flavor, and nutritional value.

From the data table: fat content (%) of different brands of Mozzarella Cheese is represented via bar graph in Figure 10:

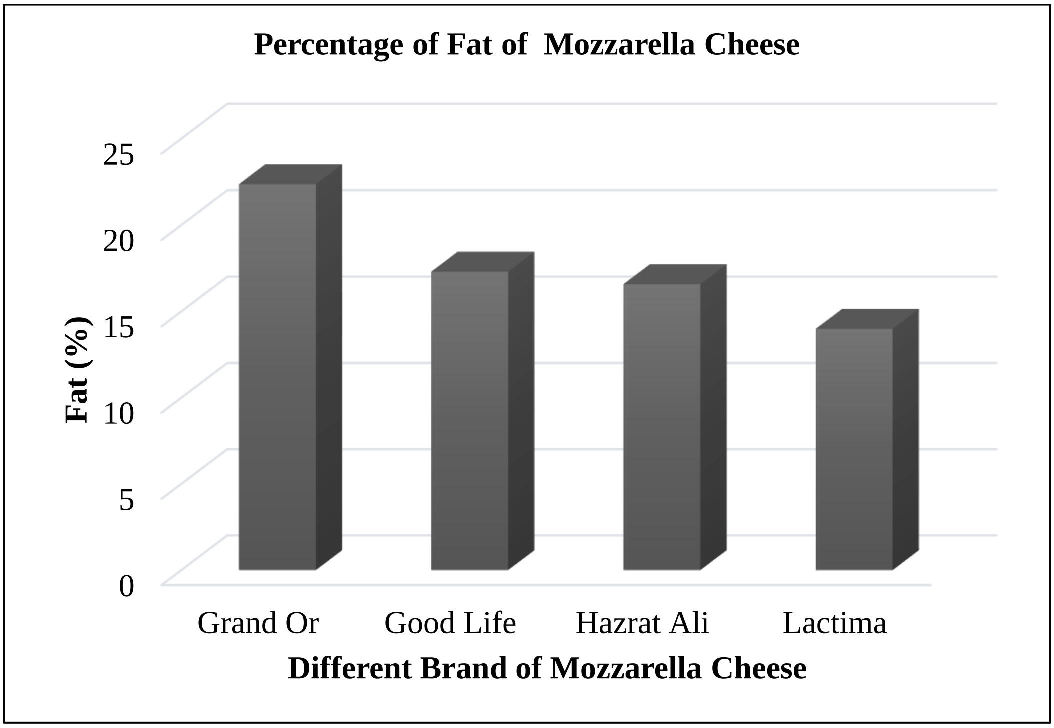


Figure 10: Percentage of fat of mozzarella cheese

4.2.5 Acidity:

Among the Four samples, the range of average percentage of acidity content was between (0.520 - 1.067) %. The results showed a significant difference in acidity levels ($p = 0.050$), with Cm_3 exhibiting the highest acidity (1.067±0.379). Increased acidity could influence the flavor profile of the product, making Cm_3 potentially more suitable for certain applications where a tangy or sour flavor is desirable. On the other hand, the lower acidity in Cm_1, Cm_2, and Cm_4 suggests that these conditions may produce less acidic products, potentially appealing to consumers seeking milder flavors.

From the data table: acidity (%) of different brands of Mozzarella Cheese is represented graphically in Figure 11:

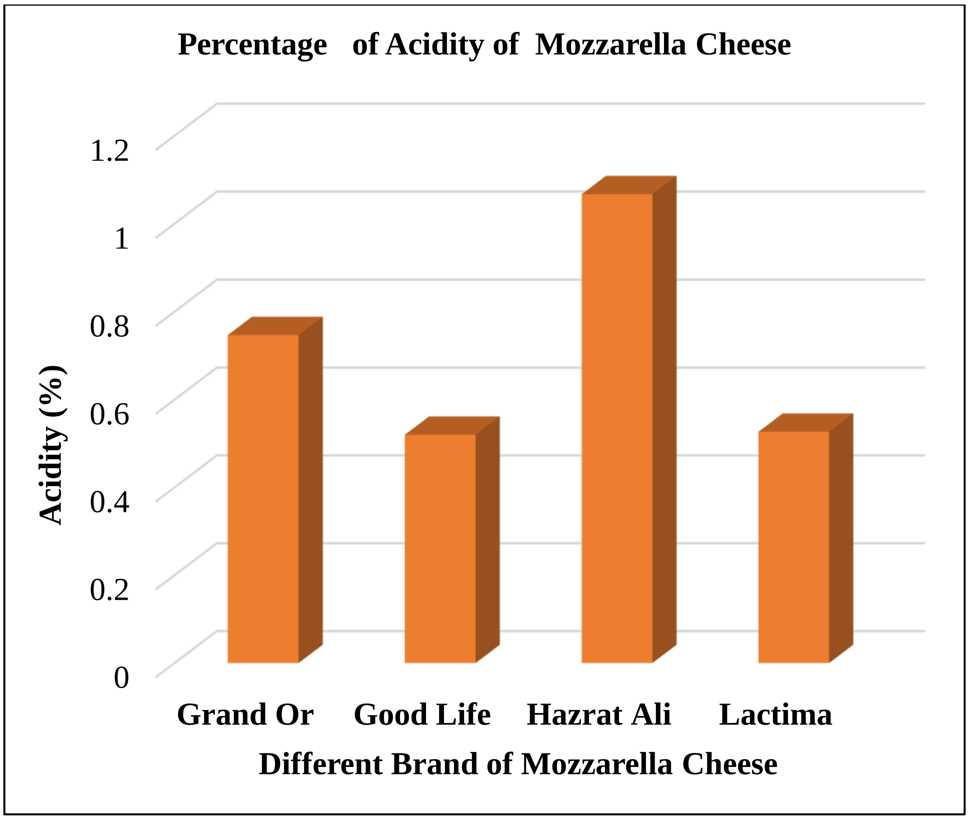


Figure 11: Percentage of acidity of mozzarella cheese

4.2.6 Ash:

Among the Four samples, the range of average percentage of ashcontent was between (1.833 - 3.183) %. A significant difference was observed in ash content ($p = 0.002$), with Cm_2 having the highest ash content (3.183±0.355). This suggests that Cm2 may have been more effective at retaining minerals, which could be beneficial in products where higher mineral content is desired, such as fortified foods. Cm_3, in contrast, exhibited the lowest ash content (1.833±0.076), indicating a potential loss of minerals during processing.

From the data table: ash (%) of different brands of Mozzarella Cheese is represented via bar graph in Figure 12:

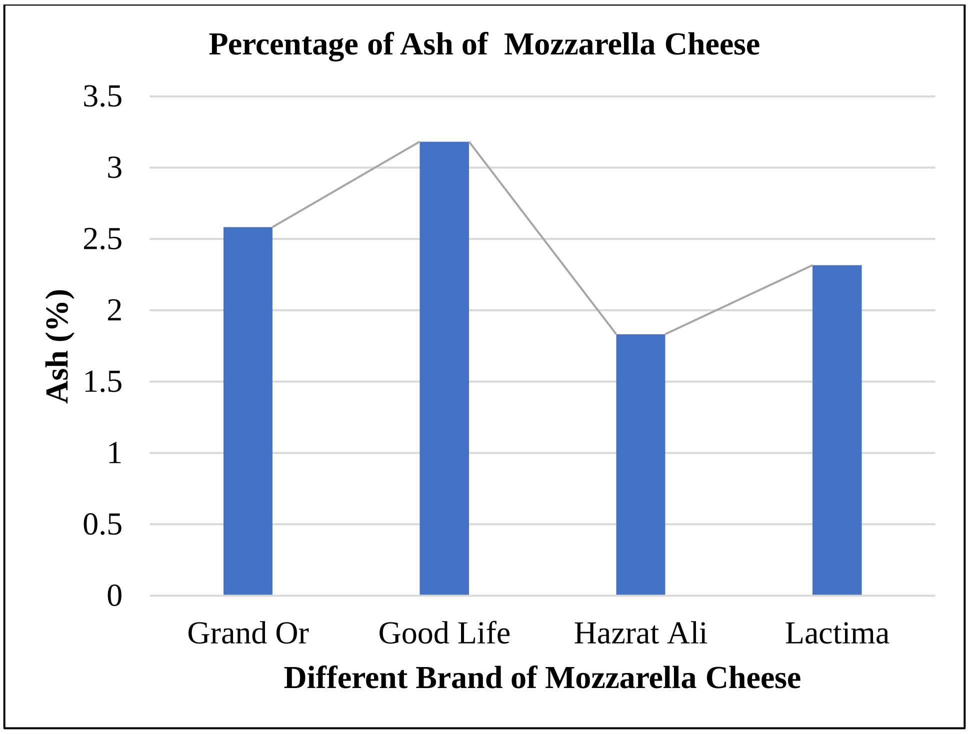


Figure 12: Percentage of ash of mozzarella cheese

4.2.7 Salt:

Among the Four samples, the range of average percentage of ash content was between (1.060 - 0.393) %. A significant difference in salt content was found ($p = 0.003$), with Cm_2 and Cm_4 showing the highest salt content. This could have implications for the preservation, flavor, and health aspects of the product. The higher salt content in Cm_2 and Cm_4 might be due to increased salt retention during processing or added as part of the treatment, which could influence consumer preferences depending on dietary needs.

From the data table: salt (%) of different brands of Mozzarella Cheese is represented graphically in Figure 13:

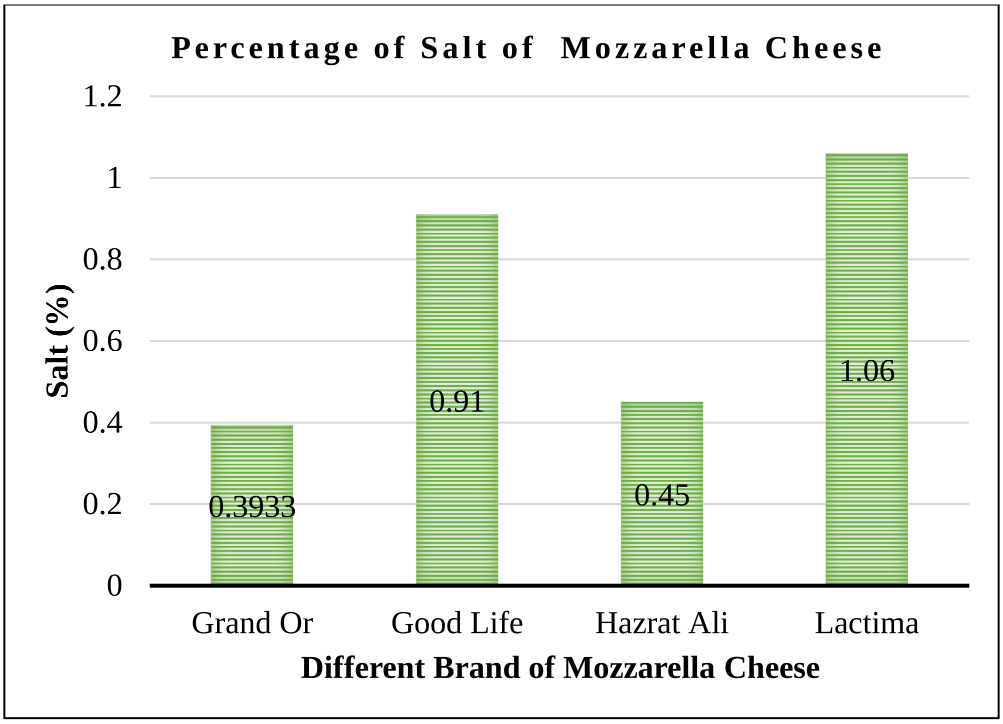


Figure 13: Percentage of salt of mozzarella cheese

4.3 Microbial Analysis of Mozzarella Cheese

Microbial analysis is crucial in evaluating the safety, quality, and characteristics of cheese. It helps identify microbial populations and their roles during cheese production, fermentation, and aging.

4.3.1 Coliform count

The presence or absence of microbial growth in different cheese samples (Cm_1, Cm_2, Cm_3, and Cm_4) was analyzed across three batches. Among four samples, 3 samples had no detectable microbial presence across all batches, indicating strict hygiene and effective quality control measures. But one sample (Cm_3) had microbial counts ranged from 9.00×10^1 to 1.00×10^2 CFU/g, with an average microbial load. The results are presented in Table 3.

Table 3: Coliform count (CFU/g) of Mozzarella Cheese

sample	Batch 1	Batch 2	Batch 3	Average	P Value	Level of sign
Cm_1	NG	NG	NG	0	0.12	NS
Cm_2	NG	NG	NG	0		
Cm_3	1.00×10^2	8.90×10^2	0.98×10^2	3.62×10^2		
Cm_4	NG	NG	NG	0		

Coliform count (CFU/g) of Mozzarella Cheese

Legends: NG = No Growth, NS = statistically not significant

4.3.2 TVC Count of Mozzarella Cheese

The Total Viable Count (TVC) analysis further confirms the presence of microbial load in three of the cheese samples. Samples Cm_1, Cm_2, and Cm_3 exhibited detectable microbial growth, with TVC values ranging from 8.5×10^2 to 1.92×10^5 CFU/g, in contrast, sample Cm_4 showed no detectable microbial growth across all batches. The results are presented in Table 4.

Table 4: TVC count (CFU/g) of Mozzarella Cheese

Sample	Batch 1	Batch 2	Batch 3	Average	P Value	Level of sign
Cm_1	8.5×10^2	9.0×10^3	7.2×10^3	5.6×10^3	0.03	*
Cm_2	1.87×10^5	1.80×10^5	1.92×10^5	1.86×10^5	0.01	**
Cm_3	1.25×10^5	1.22×10^5	1.30×10^5	1.26×10^5	0.02	*
Cm_4	NG	NG	NG	0	-	ND

Coliform count of Mozzarella Cheese

Legends: *Statistically significant at $P \leq 0.05$, **Statistically significant at $P \leq 0.01$,

NG = No Growth, ND = Not Detected.

Chapter-5: Discussion

In this study, a total of 12 cheese samples were examined, consisting of four brands each of Mozzarella Cheese, with three different batches from each brand. The physical and chemical properties of the samples were analyzed using standardized AOAC methods to ensure precision and consistency. Additionally, a detailed sensory evaluation was conducted using a structured scorecard, where trained panellists assessed key attributes such as color, texture, flavor, and overall acceptability. The study aimed not only to compare the quality variations between cheese types but also to identify potential factors affecting their characteristics, such as processing techniques, ingredient composition, and storage conditions. This comprehensive evaluation provides valuable insights for manufacturers and consumers regarding the quality and sensory appeal of these cheese varieties.

5.1 Analysis of sensory attributes

In our study, the sensory evaluation of mozzarella cheese yielded the following scores: color (7.767±0.462 to 8.737±0.251), body (10.153±0.194 to 13.360±0.445), texture (10.500±0.250 to 13.133±0.188), flavor and aroma (30.917±0.520 to 41.680±1.850) and finish and appearance (7.657±1.469 to 13.277±0.254). When compared to previous studies, Mizan *et al.* (2010) assessed similar sensory attributes, including flavor and taste, body and texture, finish, and color. Their reported scores—flavor and taste (40.1), body and texture (26.3), finish (12.5), and color (8.2)—were generally higher than those observed in our study. Rastogi *et al*. (1989) documented that the color score for cow milk cottage cheese ranged from 11.1 to 13.3 out of 15 (74–89%), which aligns with the findings of our study. Similarly, Dharam and Garg (1989) reported an appearance and color score of 7.9 ± 0.2, further supporting the consistency of our results. Similarly, Zedan *et al*. (2014) found that the appearance score of freshly prepared mozzarella cheese was almost identical to our findings. However, after 28 days of storage, their recorded scores were slightly lower. Additionally, minor discrepancies were noted in body & texture and flavor, which could be attributed to variations in processing techniques, ingredient composition, or storage conditions.

5.2 Nutritional Analysis:

The proximate analysis of the mozzarella cheese samples showed significant differences in their moisture, protein, fat, acidity, ash, and salt contents ($P \leq 0.05$). The results indicate that milk composition, processing techniques, and quality control measures have a substantial impact on the nutritional characteristics of the cheese.

5.2.1 Moisture Content dry matter:

The moisture content of the samples ranged from 48.70% (Cm_2) to 52.98% (Cm_4), with Cm4 exhibiting the highest moisture content, which is inversely related to its dry matter content ($P = 0.004$). This is consistent with findings from other studies, where higher moisture content correlates with lower dry matter. Mijan *et al.* (2010) reported a moisture content of 50.60% in mozzarella cheese made from fresh milk in Bangladesh. The moisture levels dry matter levels in our samples are within the range observed by Mijan *et al.*, indicating consistency with their study. Additionally, Mari *et al.* (2014) reported that mozzarella cheese made from buffalo milk was 55.15%. our moisture and dry matter contents were slightly lower on average than the findings of Mari *et al*. Zedan *et al*. (2014) investigated mozzarella cheese made from various milk sources and reported moisture content ranging from 48.17% to 49.65%, the moisture content observed in our study was slightly higher on average. Another investigation conducted by El-Owni and Osman (2009) stated that a moisture content of 48.59% in mozzarella cheese produced using Kosikowski's method, which is slightly lower than our result on average. As moisture (%) was lower in their study, DM (%) of examined sample would be higher than our observed value.

5.2.2 Protein Content:

Protein content significantly varied among the samples ($P = 0.001$), with Cm_2 exhibiting the highest protein content (25.35%) and Cm_4 the lowest (17.57%). This variation may be attributed to differences in milk source and cheese production methods. Protein content is a critical factor affecting the nutritional quality and functional properties (meltability and stretchability) of mozzarella cheese. Mijan *et al*. (2010) also reported a protein content of 22.10%, which is within the range observed in our samples. According to Mari *et al.* (2014), mozzarella cheese made from buffalo milk had a Protein Content of 25.28%. In contrast, our samples exhibited slightly lower protein contents on average which could be due to differences in milk source (cow vs. buffalo milk) and processing conditions. Similarly, Zedan et al (2014) found that the protein content of mozzarella cheese produced from different milk sources was between 22.53% and 26.36%. So, this finding is also within our observed value. El-owni and Oswan(2009) estimated that the protein content of mozzarella cheese was 20.06±0.65. our enumerated value of protein content was higher than the observation of El-owni and Oswan(2009).

5.2.3 Fat Content:

Fat content varied significantly among the samples (P = 0.001). Cm_1 had the highest fat content (22.36%). Cm_4 had the lowest (13.99%) that aligns closely with the buffalo milk mozzarella cheese profile described by Mari *et al.* (2014). Fat is essential for the flavor, texture, and mouthfeel of the cheese. Higher fat content is associated with superior melt and stretch properties, making it more suitable for culinary applications like pizza toppings. Similar to the findings of Mijan *et al.* (2010) and where the fat content was reported as 21.80%, our findings align with these values. Fat content of mozzarella cheese analyzed by Zedan *et al.* (2014) was between 17.9% and 21.7% and our study showed similar fat contents. El-Owni and Osman (2009) reported a fat content of 27.25% in their study, which is higher than the values observed in our samples on average.

5.2.4 Acidity:

Acidity levels varied significantly (P = 0.050), with Cm_3 having the highest acidity (1.067%) and Cm_2 the lowest (0.520%). Higher acidity typically results from extended fermentation or specific starter cultures, and it can influence the flavor profile and shelf life of the cheese. Mijan *et al.* (2010) did not report acidity directly, but variations in acidity may be influenced by the fermentation process and bacterial culture used. El-Owni and Osman (2009) found a titratable acidity of 0.66% in mozzarella cheese, which was slightly lower than our observed value on average.

5.2.5 Ash Content:

The ash content, which reflects the mineral composition of the cheese, varied significantly (P = 0.002). Cm_2 exhibited the highest ash content (3.183%), while Cm_3 showed the lowest (1.833%). The differences in ash content can be attributed to variations in milk feed and processing conditions. The findings of Mijan *et al.* (2010) and Mari *et al.* (2014) which reported an ash content of 3.50%, 3.42% respectively are comparable to our results. El-Owni and Osman (2009) reported an ash content of 2.25%, aligning closely with Cm_1 and Cm_4.

5.2.6 Salt Content:

Salt content in mozzarella cheese ranged from 0.39% (Cm_1) to 1.06% (Cm_4), showing a significant variation (P = 0.003). Salt is an essential ingredient in cheese production, contributing not only to flavor but also to microbial stability and texture. The salt content in our samples is comparable to the 0.70% reported by Mijan *et al.* (2010), demonstrating consistency between our study and their findings.

Another study conducted by Siddhant *et al*. (2024) on mozzarella cheese prepared from blends of cow and buffalo milk reported moisture content ranging from 49.00% to 53.00%, protein content from 18.00% to 24.00%, fat content from 14.00% to 21.00%, and ash content from 1.80% to 3.20%. The proximate composition of the current samples falls within these reported ranges, indicating consistency with existing literature.

5.3 Microbial Analysis:

5.3.1 Coliform count:

Samples Cm_1, Cm_2, and Cm_4 exhibited no coliform growth across all tested batches, indicating that proper hygiene measures were maintained throughout production and handling. However, sample Cm_3 showed coliform counts ranging from 1.00×10^2 to 8.90×10^2 CFU/g, with an average of 3.62×10^2 CFU/g. This suggests possible lapses in sanitation or post-processing contamination. Despite this, the P value of 0.12 indicates that the observed differences were not statistically significant. The coliform levels found in this study align with prior research findings. A study conducted by Ali and Elsherif (2015) published in the Assiut Veterinary Medical Journal reported that coliforms were present in 96% of mozzarella cheese samples, with concentrations ranging from 4×10^3 to 1.8×10^7 CFU/g and an average of 3.84×10^6 CFU/g. Similarly, another study observed that coliform counts in mozzarella cheese started at 6×10^2

CFU/g and increased to 10^3 CFU/g after five weeks of storage (Assiut Veterinary Medical Journal). Compared to these reports, the coliform counts detected in Cm_3 were lower, indicating a relatively better microbiological quality. Furthermore, research has shown that high levels of total and fecal coliforms, reaching up to 5×10^8 and 3×10^7 CFU/g (Mass *et al.* 1992), respectively, can contribute to spoilage in mozzarella cheese, leading to gas production and swelling of packaging. In contrast, the samples analyzed in this study did not exhibit any spoilage signs, supporting the conclusion that coliform contamination was minimal.

5.3.2 TVC Count:

Total Viable Count (TVC) is an essential microbiological indicator for evaluating the microbial load and safety of cheese. It measures the total number of living microorganisms, such as bacteria, molds, and yeasts, in a given sample. TVC plays a vital role in assessing the hygiene standards maintained during cheese production, storage, and handling. Monitoring TVC helps ensure product quality, detect potential contamination, and maintain compliance with food safety regulations. The Total Viable Count (TVC) analysis revealed significant variations among the mozzarella cheese samples, with a P-value indicating statistical significance ($P < 0.05$). Sample Cm_1 exhibited relatively low microbial counts, ranging from 8.5×10^2 to 9.0×10^3 CFU/g, with an average of 5.6×10^3 CFU/g. In contrast, Cm_2, and Cm_3 had considerably higher bacterial loads, with average counts of 1.86×10^5 CFU/g and 1.26×10^5 CFU/g, respectively. Notably, Cm_4 showed no bacterial growth across all batches, suggesting superior microbiological quality and possibly better processing and storage conditions. The results of this study are consistent with findings reported in previous research on the microbial quality of mozzarella cheese. A study conducted by El-Owni and Osman (2009) reported that TVC in traditionally produced mozzarella cheese ranged between 1.12×10^5 and 1.98×10^5 CFU/g, which closely aligns with the bacterial loads observed in Cm_2, and Cm_3 in this study. The relatively lower TVC in Cm_1 and the absence of growth in Cm_4 suggest that factors such as improved hygiene and better milk handling can significantly influence microbial quality. Similarly, Jana and Tagalpallewar (2017) analyzed the microbiological quality of mozzarella cheese and reported TVC counts between 10^3 and 10^5 CFU/g, depending on storage conditions and production methods. The presence of high microbial loads in Cm_2, and Cm_3 aligns with their findings, indicating that improper handling or prolonged storage can contribute to elevated bacterial counts. Another study conducted by Al-Otaibi & El-Demerdash, (2013) observed TVC counts in commercial mozzarella cheese samples ranging from 10^4 to 10^6 CFU/g, with the highest counts found in products stored under inadequate refrigeration. In this study, Cm_2, and Cm_3 fall within this range, suggesting that storage and processing conditions play a crucial role in maintaining microbial stability.

5.4 Comparison of Nutritional Status of Mozzarella Cheese with BSTI Standards

According to the standards of BSTI (2008) Mozzarella cheese should contain moisture (max. 50%), protein (min. 22%), fat (min. 20%), salt (0.5–1.5%), acidity (0.4–1.0%), and ash (2–3.5%). Cm_1 emerges as the most balanced sample, meeting all criteria except for salt content, which is slightly below the minimum (0.393%). In contrast, Cm_4 falls short in protein, fat, and moisture content, despite demonstrating acceptable microbial and sensory quality. Cm_2 performs well overall but contains slightly less fat than the BSTI standard. Meanwhile, Cm_3 fails to meet the required protein, fat, and ash levels but exhibits the highest acidity among all samples.

5.5 Comparison of Mozzarella Cheese Microbial Analysis with BSTI Standards

According to the BSTI (2008) standards for Mozzarella cheese, the total viable count (TVC) should not exceed 5×10^5 CFU/g, and coliform bacteria must be absent. In this study, Cm_1, Cm_2, and Cm_4 complied with most of the BSTI microbial safety standards, indicating acceptable hygienic quality. However, Cm_3 exhibited the poorest microbial quality, as it exceeded the permissible limits for total viable count (TVC), and showed the presence of coliform bacteria making it unsuitable for safe consumption.

Chapter-6: Conclusion

This study provides a comprehensive analysis of the nutritional and microbial quality of different brands of mozzarella cheese available in Bangladesh. The findings highlight the importance of quality assessment in terms of chemical composition, microbial safety, and sensory characteristics to ensure consumer health and satisfaction. While global demand for mozzarella cheese continues to grow, maintaining high safety standards remains a challenge, particularly in developing countries. The findings indicate that there is considerable variation in the chemical composition of the tested cheese samples, particularly in terms of moisture, protein, fat, ash, acidity, and salt content. The observed differences suggest that factors such as milk source, processing techniques, and storage conditions influence the final quality of mozzarella cheese. From a microbial quality perspective, the presence of coliform bacteria in some samples raises concerns about hygienic practices during production, handling, and storage. While most samples fell within acceptable limits for microbial safety, the detection of coliforms in certain brands highlights the need for stricter quality control measures to ensure consumer safety. Comparing these findings with previous research, mozzarella cheese produced in Bangladesh shows both strengths and areas for improvement. While some brands meet the Codex and BSTI standards for cheese composition, others exhibit deviations that require attention from producers and regulatory authorities. Additionally, sensory evaluation results suggest that consumer preference is influenced by texture, flavor, and appearance, which can be further improved through standardization of production techniques. This study highlights the need for better regulatory oversight, improved production practices, and continued research efforts to enhance the quality of mozzarella cheese available in the market. By addressing these issues, the dairy industry in Bangladesh can produce high-quality cheese that meets both local and international standards, ensuring a safe and nutritious product for consumers.

Limitations

1. The study analyzed a specific number of mozzarella cheese brands and batches, which may not represent the full spectrum of cheese available in the Bangladeshi market.
2. Samples were collected from selected regions and markets, primarily in urban areas. Cheese quality may vary in different parts of the country, especially in rural and small-scale production facilities, which were not included in this study.
3. The study did not evaluate how storage temperature, packaging conditions, and shelf life impact the nutritional and microbial stability of mozzarella cheese. Future research should assess cheese quality over different storage durations and conditions.
4. Sensory evaluation was conducted based on specific attributes, but consumer preferences and acceptability were not extensively studied.

Chapter-7: Recommendation and Future perspectives

The Quality Evaluation for the different brands of mozzarella cheese were done in this thesis has provided new knowledge. Some recommendation and future perspectives for continued work in these issues are the following-

1)Number of samples should be increased in the study.

2) More detailed examination of the cheese is highly recommended in future.

3) For the identification of microbial quality molecular technique should be implemented.

4) Lactose content, Mineral content (Calcium), Vitamin content of cheese should be checked.

5) Determination of fat content by using Soxhlet apparatus is highly recommended in future.

References

Adebolu T, Olorunfemi O. 2016. Effects of fermented cheese whey on the cells of the immune system of apparently healthy albino rats. Journal of Immunobiology. 1(2):1-3. Available from: https://www.hilarispublisher.com/open-access/effects-of-fermented-cheese-whey-on-the-cells-of-the-immune-system-of-apparently-healthy-albino-rats-jib-1000109.pdf.

Abdul M, Hakim PL, Imam T, Herly E. 2016. Physical and chemical properties of mozzarella cheese analogue microwavable. International Journal of ChemTech Research. 9(7): 171-181.

Adhikari K, Heyman H, Huff HE. 2003. Textural characteristics of low fat, full fat and smoked cheeses: sensory and instrumental approaches. Food Quality and Preference. 14(3): 211-218. Available from: https://doi.org/10.1016/S0950-3293(02)00067-8.

Aldrete AT, Meyli, Escobar RM, Mark TL, Hernández IM. 2014. High-throughput sequencing of microbial communities in Poro cheese, an artisanal Mexican cheese. Food Microbiology. 44: 136-141. Available from: https://doi.org/10.1016/j.fm.2014.05.022.

Ali ND, Elsherif MAW. 2015. Microbiological evaluation of Mozzarella cheese. Assiut Veterinary Medical Journal. 61(144): 151-158. Available from: https://doi.org/10.21608/avmj.2015.170032.

Al-Otaibi MM, El-Demerdash, HA. 2013. Microbiological quality evaluation of some dairy products. African Journal of Microbiology Research. 7(50): 5641-5647.

Ardic M. 2003. Effects of pasteurization and different scalding temperatures on the quality of Urfa cheese (PhD Thesis). Selc`11uk University, Konya, Turkey.

Association of Official Analytical Chemists (AOAC). 1995. Official Methods of Analysis of AOAC International: 16th Edition.

Barcenas P, Elortondo FJ, Albisu M, Jacky M, Bivar RL, Francesca SM, Pierre L. 2007. An international ring trial for the sensory evaluation of raw ewes' milk cheese texture. International Dairy Journal. 17(9): 1139-1147.

Barkham P. January 10, 2012. "Why is cheese the most shoplifted food item in the world?". The Guardian. Retrieved April 10, 2022.

Bhattarai RR, Acharya PP. 2010. Preparation and quality evaluation of mozzarella cheese from different milk sources. Journal of Food Science and Technology Nepal. 6: 94-101. Available from: https://doi.org/10.3126/jfstn.v6i0.8268.

Bogue JC, Delahunty CM, Henry MK, Muarry JM. January 1999. Market-oriented methodologies to optimise consumer acceptability of Cheddar-type cheeses. British Food Journal. 101(4): 301-317. Available from: https://doi.org/10.1108/EUM0000000004561.

Bradley RL. 2023. Dairy Products. In: Latimer GW, editor. Official Methods of Analysis AOAC INTERNATIONAL, 22; online edn. New York: Oxford University press. Available from: https://doi.org/10.1093/9780197610145.003.213.

Burkhalter G. 1981. Catalogue of Cheese. In: Document 141, International Dairy Federation, Brussels, Belgium. pp. 15-33.

Citro V. 1981. A typical local product obtained from buffalo milk. Scienza-e-Tecnica-Lattiero-Casearia. 32: 263-273.

Das T. November 17, 2019. The popular Arong cheese. The Business Standard. Available from: https://www.tbsnews.net/feature/food/popular-aarong-cheese.

Davis JG. 1965. Cheese, Vol. 1, Basic Technology; Vol. 2, Bibliography. Churchill Livingstone, London.

Davis JG. 1976. Cheese, Volume III Manufacturing Methods, Churchill Livingstone Edinburgh. pp. 906–907.

De S. 2010. Outline of Dairy Technology. Oxford University Press. pp.224, 229, 263.

Dharam P, Garg FC 1989. Utilization of sour buttermilk in the manufacture of Paneer. Indian Journal of Dairy Science 42: 589-594.

Dina NA, Elsherif WM. 2015. Microbiological evaluation of Mozzarella cheese. Assiut Veterinary Medical Journal. 61(144): 151-158. Available from: https://doi.org/10.21068/avmj.2015.170032.

El-Owni OA, Osman SE. 2009. Evaluation of chemical composition and yield of mozzarella cheese using two different methods of processing. Pakistan Journal of Nutrition. 8(5): 684-687. Available from: https://doi.org/10.3923/pjn.2009.648.687.

Eshetu M, Asresie A. 2019. Chemical composition, mineral profile and sensory properties of traditional cheese varieties in selected areas of Eastern Gojjam, Ethiopia. East African Journal of Sciences. 13(2): 185-194.

Fenelon MA, Guinee TP, Delahunty CM, Murray J. 2000. Composition and sensory attributes of retail Cheddar cheese with different fat contents. Journal of Food Composition and Analysis. 13(1): 13-26. Available from: https://doi.org/10.1006/jfca.1999.0844.

Fox PF. 1993. Cheese: An overview. In: Fox, P.F. (Ed.), Cheese: Chemistry, Physics and Microbiology, Vol. 1, 2nd ed., London (UK): Chapman & Hall. pp. 1-36.

Fox PF. 2011. Cheese: Overview. In: Fuquay JW (Ed.), Encyclopedia of Dairy Science, 2nd ed., San Diego, Academic Press. pp. 533-543.

Fox PF, Guinee TP, Cogan TM, McSweeney PL. 2000. Principal families of cheese. In Fundamentals of Cheese Science, 2nd ed., pp. 388.

Ghosh BC, Singh S. 1996. A comparison of cow and buffalo milk Mozzarella cheese. Indian Journal of Dairy Science. 49(1): 38-41.

Giongo L, Ajelli M, Poncetta P, Ramos-Garcia M, Sambo P, Farneti B. 2019. Raspberry texture mechanical profiling during fruit ripening and storage. Postharvest Biology and Technology. 149: 177-186.

Goncalves RF, Ben-Hur, Grazielly DJ, Daniele GC, Silvio DE, Sibelli PB. 2017. Buffalo mozzarella chemical composition and authenticity assessment by electrophoretic profiling. Food Sciences. 38(4): 1841-1851. Available from: https://doi.org/10.5433/1679-0359.2017v38n4p1841.

Gunasekaran S. 2016. Cheese quality evaluation. In: Sun DW (Ed.), Computer Vision Technology for Food Quality Evaluation, 2nd ed., Academic Press. pp. 487-524.

Hajjar IM, Grim CE, Kotchen TA. 2003. Dietary calcium lowers the age-related rise in blood pressure in the United States: the NHANES III survey. Journal of Clinical Hypertension (Greenwich). 5(2): 122-126. Available from: https://doi.org/10.1111/j.1524-6175.2003.00963.x.

Indian Standard (IS). 1996. IS: 3507-1966. Indian Standard Methods of Sampling and Test for Butter.

International Dairy Federation (IDF). 2014. IDF 20-1. Milk and Milk Products - Determination of Nitrogen Content: Kjeldahl Principle and Crude Protein Calculation.

International Dairy Federation (IDF). 2016. Bulletin of the International Dairy Federation. 485/2016.

International Organization for Standardization (ISO). ISO 4832:2006(E). Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of coliforms – Colony-count technique.

Jacqmain M, Doucet E, Després JP, Bouchard C, Tremblay A. 2003, June. Calcium intake, body composition, and lipoprotein-lipid concentrations in adults. The American Journal of Clinical Nutrition. 77(6): 1448-1452. Available from: https://www.sciencedirect.com/science/article/pii/S0002916523058070?via%3Dihub.

Jana A, Tagalpallewar PG. 2017. Functional properties of Mozzarella cheese for its end-use application. Journal of Food Science and Technology. 54(12): 3766-3778. Available from: https://doi.org/10.1007/s13197-017-2886-z.

Johnson ME, Olson NF. 1985. A comparison of available methods for determining salt levels in cheese. Journal of Dairy Science. 68(4): 1020-1024. Available from: https://www.journalofdairyscience.org/article/S0022-0302(85)80924-3/pdf.

Kaushik R, Pawar N, Arora S, Padghan P. 2019. Cheese nutrition and health benefits. National Seminar. 57-63. Available from: https://www.researchgate.net/publication/353403479_CHEESE_NUTRITION_AND_HEALTH_BENEFITS.

Kindstedt PS, Fox PF. 1993. Effect of manufacturing factors, composition, and proteolysis on the functional characteristics of Mozzarella cheese. Critical Reviews in Food Science and Nutrition. 33(2): 2167-2187.

Kindstedt PS. 1993. Mozzarella and pizza cheese. In: Cheese: Chemistry, Physics and Microbiology. 2nd ed. Burlington (VT): Department of Animal Sciences, University of Vermont, USA. p. 337-362.

Kosikowski FV. 1982. Cheese and Fermented Milk Foods. 2nd ed. Ann Arbor (MI): Edwards Brothers, Inc.

Kumar S, Rai DC, Niranjan K, Bhat ZF. 2014. Paneer—An Indian soft cheese variant. Journal of Food Science and Technology. 51(5): 821-831. Available from: https://doi.org/10.1007/s13197-011-0567-x.

Kunova S, Kacaniova M, Juraj C, Peter H. 2015. Evaluation of microbiological quality of selected cheeses during storage. Potravinarstvo: Scientific Journal for Food Industry. 9(1): 143-148. Available from: https://doi.org/10.5219/463, https://www.researchgate.net/publication/277946804_Evaluation_of_microbiological_quality_of_selected_cheeses_during_storage.

Lambert P. 2008. Mozzarella cheese. Retrieved April 1, 2008.

Lawrence R, Heap H, Gilles J. 1984. A controlled approach to cheese technology. Journal of Dairy Science. 67(8): 1632-1645. Available from: https://doi.org/10.3168/jds.S0022-0302(84)81486-1.

Legg AK, Carr AJ, Bennett RJ, Johnston KA. 2017. General aspects of cheese technology. In: McSweeny Paul LH, Fox PF, Cotter PD, Everett DW, editors. Cheese: Chemistry, Physics and Microbiology. Vol. 2, 4th ed. Academic Press. pp. 643-675.

Lorenzini EC, Lazzari B, Tartaglia GM, Farronato G, Lanteri V, Botti S, Stella A. 2022. Oral ecological environment modifications by hard cheese: from pH to microbiome: a prospective cohort study based on 16S rRNA metabarcoding approach. Journal of Translational Medicine. Available from: https://doi.org/10.1186/s12967-022-03506-4.

Losito F, Arienzo A, Bottini G, Priolisi FR, Mari A, Visca P, Antonini G. 2014. Microbiological safety and quality of Mozzarella cheese assessed by the microbiological survey method. Journal of Dairy Science. 97(1): 46-55. Available from: https://doi.org/10.3168/jds.2013-7026.

Mari GM, Khaskheli M, Jatoi AS, Qazi IH, Sarnina M, Talpur A, Samad A. 2014. Compositional and rheological properties of Mozzarella cheese prepared from buffalo milk. Pakistan Journal of Nutrition. 13(9): 533-536. Available from: https://doi.org/10.3923/pjn.2014.533.536.

Massa S, Gardini F, Sinigaglia M, Guerzoni ME. 1992. Klebsiella pneumoniae as a spoilage organism in Mozzarella cheese. Journal of Dairy Science. 75(6): 1411-1414. Available from: https://doi.org/10.3168/jds.S0022-0302(92)77894-1.

McLaren DS, Frigg MM. 2001, January 1. Sight and Life Manual on Vitamin A Deficiency Disorders (VADD). Taskforce Sight and Life, Basel.

McSweeny PLH. 2007. Introduction (Principal families of cheese). In: McSweeny PLH, editor. Cheese Problems Solved. Cambridge (UK): Woodhead Publishing Limited. pp. 176-177.

McSweeny PLH. 2007. How did cheese originate? (Principal families of cheese). In: McSweeny PLH, editor. Cheese Problems Solved. Cambridge (UK): Woodhead Publishing Limited. pp. 184.

Mijan M, Hoque M, Habib M, Wadud M. 2010. Evaluation of quality of Mozzarella cheese. The Bangladesh Veterinarian. 27(1): 36-42.

Miller J. 2017, April 29. Health benefits of cheese, according to science (+8 delicious recipes). Retrieved from: Jen Reviews. Available from: https://www.jenreviews.com/cheese/.

Muir DD, Hunter EA. 1991. Sensory evaluation of Cheddar cheese: Order of tasting and carryover effects. Food Quality and Preference. 3: 141-145.

Nur IT, Ghosh BK, Urmi JN, Akter D, Ema EI. 2015. Microbiological quality assessment of milk and milk products along with their packaging materials collected from a food industry in the Dhaka Division. SVOA Microbiology. 2(2): 19-25.

Papetti P, Carelli A. 2013. Composition and sensory analysis for quality evaluation of a typical Italian cheese: Influence of ripening period. Czech Journal of Food Science. 31(5): 438-444.

Patel GC, Vyas S, Upadhyay AK. 1986. Evaluation of Mozzarella cheese made from buffalo milk. Journal of Dairy Science. 39: 394-403.

Proma A. 2016. Care for some cheese? The Daily Star. Available from: https://www.thedailystar.net/lifestyle/special-feature/care-some-cheese-1259203.

Ranganna S. 2000. Handbook of analysis and quality control for fruit and vegetable products. pp. 1038-1040, 1052-1054.

Rastogi MK, Panday RN, Tewari BD, Singh S. 1989. Comparative evaluation of lactic cultures of cottage cheese made from cow and buffalo milk. Indian Journal of Dairy Science. 42: 561-564.

Razi H, Greco E. August 17,2018. World's oldest cheese found in Egyptian tomb. Phys.org. Available from: https://phys.org/news/2018-08-world-oldest-cheese-egyptian-tomb.html.

Rudan MA, Barbano DM, Guo MR, Kindstedt PS. 1998. Effect of the modification of fat particle size by homogenization on composition, proteolysis, functionality, and appearance of reduced-fat Mozzarella cheese. Journal of Dairy Science. 81(8): 2065-2076.

Sameen A, Anjum FM, Huma N, Nawaz H. 2008. Quality evaluation of Mozzarella cheese from different milk sources. Pakistan Journal of Nutrition. 7(6): 753-756.

Semba RD, Lyles CM, Margolick JB, Gaiaffa W, Cohn SF. 1998. Vitamin A supplements and immunodeficiency virus load in injection drug users. Journal of Infectious Disease. 177: 611-616.

Sonwane SR, Thombre BM, Chauhan DS. 2024. Physico-chemical evaluation of Mozzarella cheese prepared from blends of cow and buffalo milk. International Journal of Advanced Biochemistry. 8(11): 46-49. Available from: https://doi.org/10.33545/26174693.2024.v8.i11a.2809.

Silanikove N, Leitner G, Merin U. 2015. The interrelationships between lactose intolerance and the modern dairy industry: Global perspectives in evolutionary and historical backgrounds. Nutrients. 7(9): 7312-7331. Available from: https://doi.org/10.3390/nu7095340.

Singh T, Drake M, Cadwallader K. 2003. Flavor of Cheddar cheese: A chemical and sensory perspective. Comprehensive Reviews in Food Science and Food Safety. 2(4): 166-189.

Smith JH. 1995. Cheese making in Scotland-A history. The Scottish Dairy Association.

Souza BM, Borgonovi TF, Casarotti SN, Todorov SD, Penna AL. 2019. Lactobacillus casei and Lactobacillus fermentum strains isolated from Mozzarella cheese: Probiotic potential, safety, acidifying kinetic parameters, and viability under gastrointestinal tract conditions. Probiotics and Antimicrobial Proteins. 11(2): 382-396. Available from: https://doi.org/10.1007/s12602-018-9406-y.

Stone H, Sidel J, Oliver AW, Singleton R. 2004. Sensory evaluation by quantitative descriptive analysis. Food Technology. 28: 11-24.

Van Mierlo L, Arends LR, Streppel MT, Zeegers MP, Kok FJ, Grobbee DE, Geleijnse JM. 2006, August. Blood pressure response to calcium supplementation: A meta-analysis of randomized controlled trials. Journal of Human Hypertension. 20(8): 571-580. Available from: https://doi.org/10.1038/sj.jhh.1002038.

Virgili R, Parolari G, Bolzoni L, Barbieri G, Mangia A, Careri M, Zannoni M. 1994. Sensory-chemical relationships in Parmigiano-Reggiano cheese. Food Science and Technology. 27(5): 491-495. Available from: https://doi.org/10.1006/fstl.1994.1098.

Walter HE, Hargrove RC. 1972. Cheeses of the world by US Department of Agriculture. Dover Publications, Inc., New York. pp. 2-3.

Walther B, Schmid A, Seiber R, Wehrmüller K. 2008. Cheese in nutrition and health. Switzerland: Dairy Science and Technology. 88: 389-405. Available from: https://www.dairy-journal.org/articles/dst/pdf/2008/04/dst88s4501.pdf.

Ware M. September 13,2017. Is cheese good or bad for you? Medical News Today. Available from https://www.medicalnewstoday.com/articles/299147#the-bottom-line.

Zedan IA, Abou-Shaloue Z, Zaky SM. 2014. Quality evaluation of Mozzarella cheese from different milk types. Alexandria Science Exchange Journal. 35(3): 162-177.

.

Appendix 1

SCORE CARD FOR EVALUATION OF SENSORY ATTRIBUTES OF MOZZARELLA CHEESE

Sample code:

Batch:

Judge:

Date of evaluation:

Attribute	Standards	Full Marks	Obtained Marks	Defects
Color	• Not artifically colored • Uniformed color	10		❖ Faded/bleached ❖ High/unnatural mottled ❖ Seamy ❖ Uneven/wavy

Attribute	Standards	Full marks	Obtained marks	Defects
Body	• Slightly elastic • Firm but not hard when crushed between fingers	15		❖ Softy body ❖ Crumbly body ❖ Greasy body ❖ Curdy body ❖ Watery body ❖ Dry/hard body

Attribute	Standards	Full marks	Obtained marks	Defects
Texture	• Compact • Continuous and homogenous • Free from openings, holes, cracks and fissures	15		❖ Mechanical holes ❖ Gas holes ❖ Yeast holes

Attribute	Standards	Full Marks	Obtained marks	Defects
Flavor and aroma	• Causes a pleasant sensation within mouth • Pleasing aroma • After swallowing taste resembling the flavor of sweet nuts • Mildy salted in taste	45		❖ Sour flavor ❖ Bitter flavor ❖ Mouldy flavor

Attribute	Standards	Full marks	Obtained marks	Defects
Finish and Appearance	• Smooth • Neat, clean, unbroken, appearance	15		❖ Misshapen surface ❖ Huffed surface ❖ Cracked surface ❖ Mouldy Surface

Appendix ii (Photo gallery)

Grand OR Mozzarella Cheese

Lactima Mozzarella Cheese

Hazrat Ali Mozzarella Cheese

Good Life Mozzarella Cheese

Collection of Sample

Collection of Sample

Sample at Superstore

Sample at Superstore

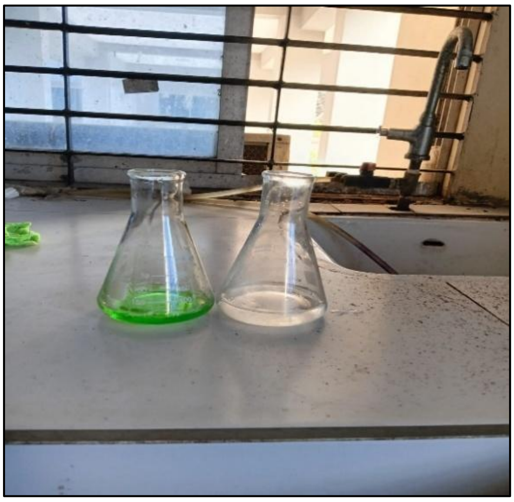
Sample for Sensory Evaluation

Sample for Sensory Evaluation

Moisture and Dry matter Determination

Moisture and Dry matter Determination

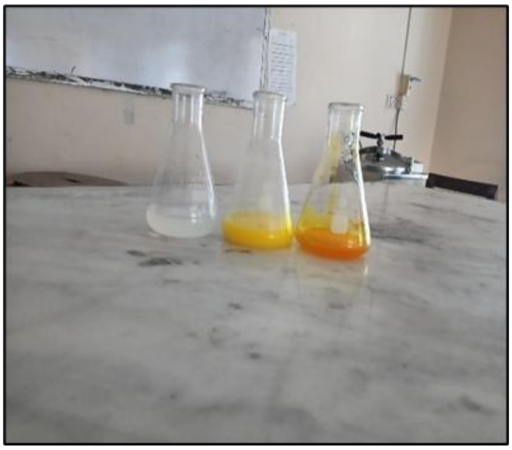
Protein Determination



Protein Determination

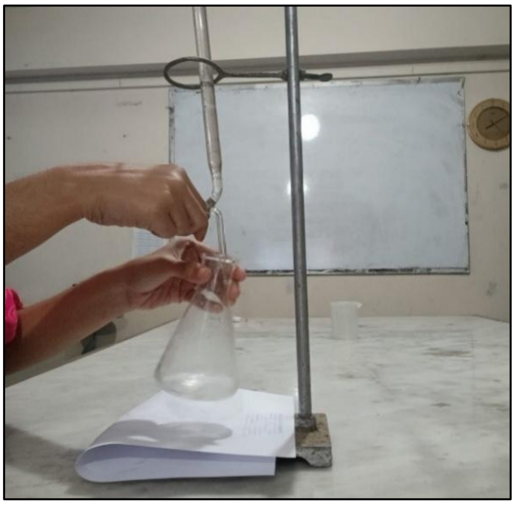
Fat Determination

Fat Determination



Salt Determination

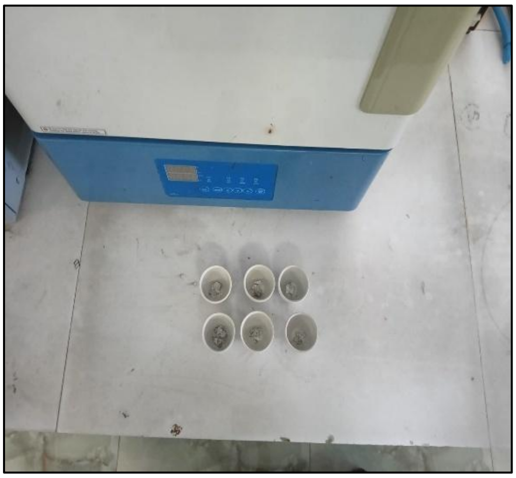
Salt Determination



Acidity Determination

Acidity Determination

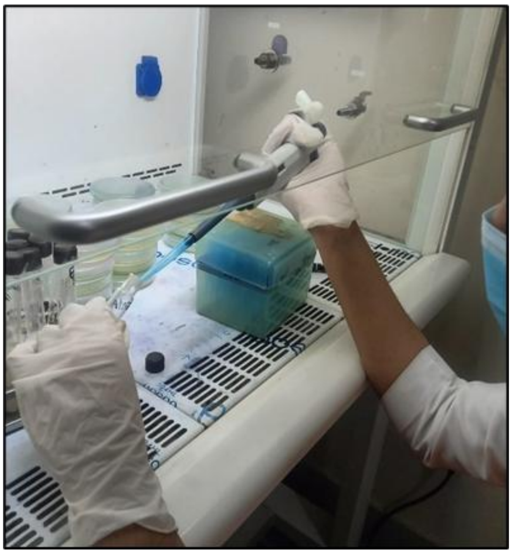
Ash Determination



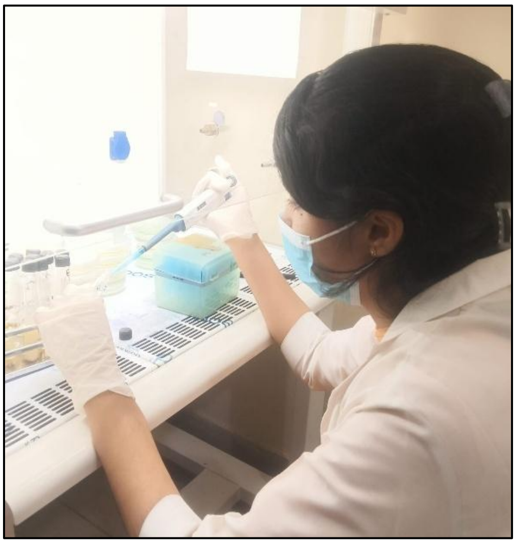
Ash Determination

Dilution of Sample

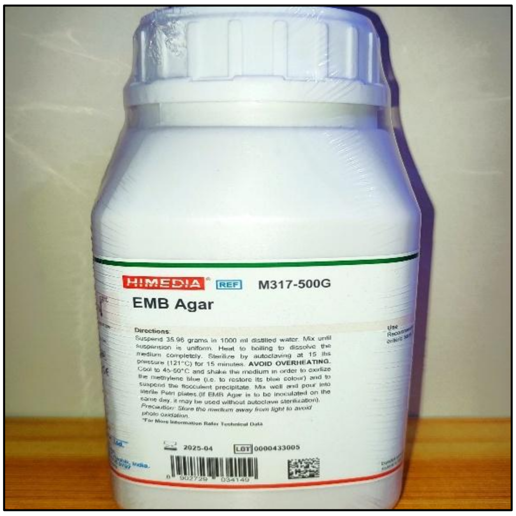
Dilution of Sample



Pouring of media



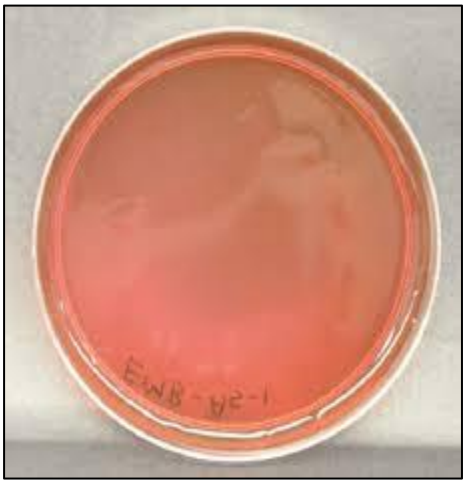
Pouring of media



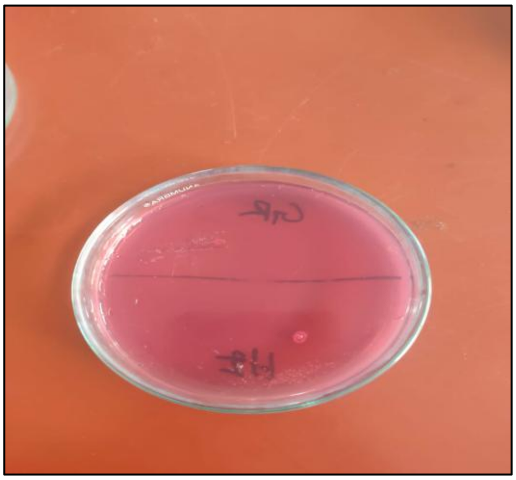
EMB Agar

Plate Count Agar

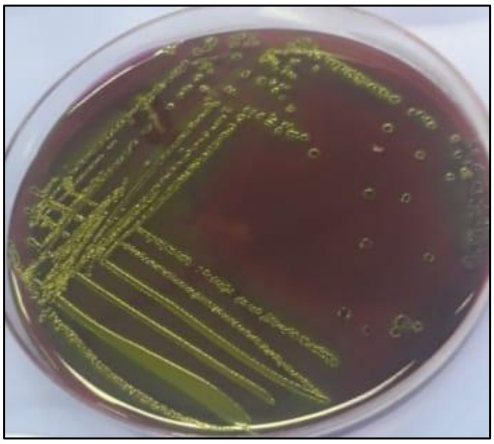
VRB Agar



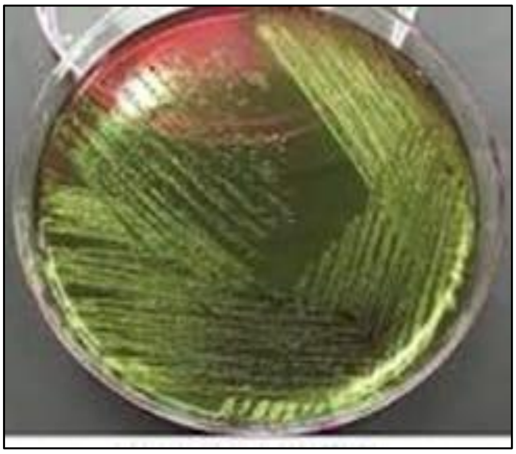
No Bacterial Colony growth in media



No Bacterial Colony growth in media

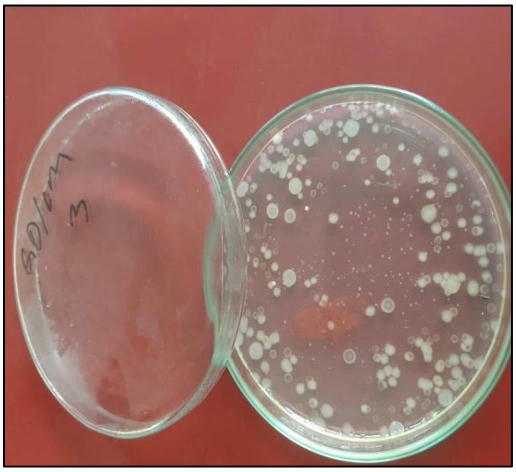


Metallic sheen in EMB



Metallic sheen in EMB Agar

Bacterial Colony growth in media



Bacterial Colony growth in media

Biography

I am Ema Saha, the proud daughter of Kanak Saha and Mondira Saha, born in a serene village at Rangunia Upazilla in Chattogram, Bangladesh. My academic journey began at Rangunia Majumderkhill High School, where I earned my S.S.C. in 2013, followed by my H.S.C. from Kapasgola City Corporation Mohila College, Chattogram in 2015. Passionate about food science, I pursued my undergraduate degree in Food Science and Technology at Chattogram Veterinary and Animal Sciences University (CVASU), graduating in 2020.Currently, I am engaged in postgraduate studies at the Department of Applied Food Science and Nutrition, Faculty of Food Science and Technology, CVASU, where I am refining my expertise and expanding my research horizons. My ultimate goal is to drive advancements in food technology that improve both industry standards and public health.