



## Effect of Sono-Ohmic Pasteurizer on Microbial, Physicochemical and Sensorial Properties of Milk

Hafiz Danish Butt<sup>1</sup>, Syeda Noor-ul-Ain Naqvi<sup>2</sup>, Muqadas Iqbal<sup>3</sup>, Hira Shair<sup>2,4</sup>, Abdul Rauf Rajput<sup>5</sup>, Muhammad Kashif Iqbal Khan<sup>1,2</sup>, Aman Arif<sup>6</sup>

<sup>1</sup>Department of Food Engineering, University of Agriculture, Faisalabad, Punjab, Pakistan

<sup>2</sup>National Institute of Food Science and Technology, University of Agriculture, Faisalabad, Punjab, Pakistan.

<sup>3</sup>Government College Women University, Faisalabad, Punjab, Pakistan

<sup>4</sup>Wheat Research Institute, Ayub Agricultural Research Institute, Faisalabad, Punjab, Pakistan

<sup>5</sup>Institute of Food Science and Technology, Sindh Agriculture University, Tandojam, Sindh, Pakistan.

<sup>6</sup>Department of Nutritional Sciences, Government College University, Faisalabad, Punjab, Pakistan.

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**Correspondence to:** Muhammad Kashif Iqbal Khan,

Department of Food Engineering, University of Agriculture, Faisalabad, Punjab, Pakistan/National Institute of Food Science and Technology, University of Agriculture, Faisalabad, Punjab, Pakistan.

Email: [kashif.khan@uaf.edu.pk](mailto:kashif.khan@uaf.edu.pk)

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### ABSTRACT

Milk is a perfect food that is nutritious, and essential part of the human diet. It is perishable, and consumers want to use it fresh. It is being processed to increase the shelf life for later use. Traditional thermal methods of pasteurizing and sterilizing foods are based on heat transfer. These methods cause color loss, flavor, and nutrients due to loss of protein, and volatile compounds. Milk samples were treated at different temperatures for different time duration and the best combination of both was observed. For this purpose, milk was treated at different temperatures (50°C, 60°C, 63°C) for 10, 15, and 20 minutes respectively. The study showed pH values were 6.36 to 6.67, and titratable acidity of 0.15 to 0.11, indicating that the sono-ohmic pasteurization technique slightly affected these parameters. The value of brix was 5.8 to 9.2, indicating a significant effect. However, the coliform had significantly decreased from 12.16 to 11.47 log<sub>10</sub> CFU/mL, and yeast mold value from 11.99 to 11.66 log<sub>10</sub> CFU/mL, exhibited significant impacts on milk quality. The total plate count, coliform bacteria, mold, and yeast have been linked to good milk quality and decreased risk of consumers health illnesses. The best treatment of milk was treated at 63°C for 20 minutes and the storage study was evaluated at refrigeration temperature. The storage study showed pH values, brix, titratable acidity, coliform and yeast mold, indicating that the sono-ohmic pasteurization technique had a significant effect ( $P<0.05$ ) on these parameters. Increased value of TPC, coliform bacteria, mold, and yeast had caused the deterioration of milk which is not good for health. Results showed that sono-ohmic pasteurization was an efficient process.

### INTRODUCTION

Food is thermally processed to extend the shelf life and maintain the product safety. Although food fortification can reduce the loss of nutrients and the standard thermal processes makes it difficult to maintain sensory qualities including texture, scent, flavor, and appearance. In order to meet consumers demand and provide high-quality processed food with a prolonged shelf life, novel thermal, and non-thermal processes based physical approaches for food preservation are needed (Goullieux and Pain, 2014). Milk is a healthy and extensively consumed dairy product produced by animals' mammary glands. Milk is an important source of nourishment, especially for babies, and it remains an important element of the diet for the entire human life (Willett and Ludwig, 2020).

Milk is well-known for its high concentration of essential elements such as proteins, carbohydrates, lipids, vitamins, and minerals. These nutrients make milk a significant source of energy and contribute to the human body's growth, development, and maintenance. Casein and whey proteins in milk supply amino acids that are required for tissue development and repair. Lactose in milk provides energy, while lipids present in milk give necessary fatty acids. Furthermore, milk is a natural supply of vitamins A, D, and B-complex vitamins, as well as minerals such as calcium, phosphorus, and potassium (Foroutan *et al.*, 2019). Milk is processed to guarantee safety and extend its shelf life due to its perishable nature. Pasteurization, which includes heating milk to kill dangerous bacteria, and ultra-high temperature

processing, which requires heating milk to a higher temperature for a shorter period of time to accomplish sterilization, are two of these techniques. These processing processes are critical in assuring the safety of milk, prolonging its shelf life, and maintaining its quality (Bezie, 2019).

Microbial safety and improved shelf life are two beneficial effects of milk pasteurization process. Pasteurization efficiently eliminates hazardous germs contained in raw milk, lowering the risk of foodborne illness. Pasteurization also helps to lengthen milk's shelf life by eliminating spoilage-causing bacteria, yeasts, and molds, and allowing it for better distribution and storage. The drawback of milk pasteurization process is the possible loss of heat-sensitive components such as some vitamins. However, the overall influence on nutritional availability is thought to be minor (Vroegindewey *et al.*, 2021).

Following conventional heat pasteurization techniques, milk often undergoes several common chemical changes, including protein denaturation, vitamin degradation, enzyme inactivation, the Maillard reaction, and the creation of lysin alanine. Heat during conventional pasteurization alters the chemical composition of proteins and water-soluble vitamins, such as denaturalizing proteins (Pegu and Arya, 2023). Ohmic heating is a technique that involves passing an electric current to heat food solids and liquids simultaneously. It uses electromagnetic techniques such as radiant heating, capacitive heating, and radiant dielectric. Its function is similar to microwave heating but with a different frequency. Ohmic heating has the benefit of heating items consistently. Additionally, it decreases the fouling of treated food surfaces during processing while also preventing the degradation of thermos-sensitive chemicals due to overheating. The main benefits of ohmic heating are to improve food quality, enhance sensorial attributes as well as inactivate the microorganisms, and enzymes (Gavahian *et al.*, 2019). Ohmic processing allows the material to heat at extremely fast rates (typically from a few seconds to a few minutes). Ohmic heating has numerous potential uses, such as in fermentation, evaporation, dehydration, and extraction (Knirsch *et al.*, 2010). Moreover, juice heat treatments are linked to quality loss due to vitamin and flavor component degradation. Heat can change the nutritional and organoleptic qualities of juice, process technology advancements are being pursued to reduce juice heat exposure (Sun *et al.*, 2008).

Juices with little processing currently hold a sizable market share. Heat exchange is a common method of pasteurization where energy is delivered into the food item through conductivity by steaming or warm water. The heat exchange is more or less efficient depending on the technology used and the method of pasteurization. Ohmic heating offers greater thermal efficiency, hence it is gradually gaining popularity and is now being used in various continuous facilities in the food business. With the use of electrodes, ohmic heating devices allow alternating electrical currents travel through food products that creates interior heat caused by resistance to electricity (Varghese *et al.*, 2014; Jaeger *et al.*, 2016). Due to a variety

of reasons, this method reduces the thermal harm to food. Furthermore, high-temperature/short-term treatments have a reputation for effectively eliminating germs and enzymes while retaining the food's nutritional and organoleptic properties. Thus, it has been observed that the fruit products that are processed with ohmic technology have improved organoleptic qualities (Achir *et al.*, 2016).

The use of ohmic heating is restricted due to electrode erosion at high temperatures and in acidic media (Cappato *et al.*, 2017). Enzyme activity and microbial spoilage are important aspects that must be considered while processing (Bhattacharjee *et al.*, 2022). Moreover, thermal practices to inhibit microbial growth and enzyme denaturation is a very old technique. Studies reveal many distinct losses in quality while enhancing the shelf time by using thermal treatment systems. A novel practice to increase the availability of products is ultrasound treatment which improves the quality. Ultrasonication is an innovative non-thermal technique, a part of the inhibition of microbes it also preserves liquefied foods. It is energy efficient and environmentally responsive as it produces fewer chemical and physical hazards (Mohideen *et al.*, 2015). The ultrasonic system creates high pressure and shear that disrupt the microbe cells as cavitation which is very effectively resistant against pathogenic microbes (Manzoor *et al.*, 2019; Zia *et al.*, 2019). Ultrasonication, when alone used, is not as effective as it can be used with a heating effect (thermo-ultrasound) that can inhibit microbes (Aadil *et al.*, 2015).

Sonication affects microorganisms in liquids in a hydrodynamic way, causing internal cavitation and the microstreaming phenomenon, as well as the production of radicals that damage cell structure. However, the use of ultrasound alone is insufficient to completely eradicate all bacteria. Furthermore, the nutritional value and sensory qualities of food might be negatively impacted by high ultrasonic power (Ferrario *et al.*, 2015). It offers high-quality food and is employed in a variety of processes, including pasteurization, sterilization, heating, thawing, fumigation, extraction, and fermentation (Kumar and Longhurst, 2018). In addition, it has less impact than conventional approaches on the sensory, functional, and synthetic qualities of food products (Lascorz *et al.*, 2016). Beyond ensuring microbiological safety, nonthermal technologies have benefits for the finished product. The obtained goods resemble fresh foods and suffer only minor losses in color, flavor, and nutrients. New non-thermal technologies including high pressure, pulsed electric fields, ultrasound, high-intensity light, and irradiation. The microbiological safety of the milk is guaranteed regardless of treatment, but the combined effects of the time-temperature procedures result in a slight reduction in milk quality (Bevilacqua *et al.*, 2018). High-power ultrasound has been demonstrated by some studies to be able to destroy germs, deactivate enzymes, and enhance the production of cheese or yogurt. The ability of high-power ultrasound to destroy bacteria raises the possibility of extending the shelf life of fluid milk (Jadhav *et al.*, 2023). The synergistic effect of heat and ultrasound was much higher for inactivating enzymes and reducing microbial load compared to ultrasound or heating alone. However,

thermosensation has been linked to the creation of off-odor and off-flavor in milk, a phenomenon that has been researched but is not fully understood (Priyadarshini *et al.*, 2022).

Ultrasound treatments can harm cell membranes causing them to collapse inward to varied degrees as well as wrinkle and shrink spores. *Bacillus* spores were successfully reduced in static testing employing batch ultrasonication in nonfat milk. However, ultrasonography has occasionally been used to promote bacterial growth. It has been discovered that the sensitivity to ultrasound does not depend on the size, the Gramme status, or the hydrophobicity of bacteria (Lim *et al.*, 2019). The ability to make items that resemble those that are fresh while also guaranteeing the microbiological safety of food remains a challenge for emerging technologies including high-pressure, pulsed electric fields, microwaves, and ohmic heating (Bermúdez-Aguirre *et al.*, 2009). Ultrasound combined with heat treatment cause the inactivation of microorganisms in juice, as well as to assess various quality factors like pH, acidity, and color. The inactivation of microorganisms under this novel technique was more effective than using sonication alone (Zou *et al.*, 2017).

## MATERIALS AND METHODS

### 2.1 Procurement of raw materials

Samples of raw milk were gathered from (Gulam Abad,) Faisalabad. Glass containers were sterilized, temperature of the sample was monitored, and bottles were labeled with the date and time of sample collection. After that, the samples were put in the ice box to be transported to the lab for examination. All the analysis were conducted at Dairy lab of National Institute of Food Science and Technology, University of Agriculture, Faisalabad.

### 2.2 Pasteurization

The milk sample was treated with conventional method for 15 minutes at 72°C. To check the effect of thermal treatment physicochemical and microbial analysis of sample was performed.

### 2.3 Microbial Testing

#### 2.3.1 Sterilization of glassware

Before sterilizing clean glassware thoroughly with soap and distilled water to remove dirt and debris, and dried in an oven to prevent contamination. Ensure that the glassware was properly wrapped before sterilization to prevent contamination during storage. Once it is sterilized, it is stored in a dry and clean place until it is ready to be used.

#### 2.3.2 Media preparation

Three types of agars were used for total plate count, coliform, yeast, and mold which were Volatile red agar, Nutrient Agar, and Potato dextrose agar, respectively. For preparing nutrient agar, Nutrient 2.8 grams of agar was dissolved in 100 mL of distilled water (Sirichan *et al.*, 2022). For preparing Potato dextrose agar 4.2 grams of agar was dissolved in 100 mL distilled water (Carrillo *et al.*, 2020; Wang *et al.*, 2018). Similarly, volatile red agar used for coliform growth takes 100 mL of distilled water in a conical flask and adds 3.85 grams of volatile red agar as mentioned on the label of a box that volatile red agar 38.5 grams for 1 liter of distilled water (Olushola *et al.*, 2021).

All the prepared media were sterilized in an autoclave at 121°C for 15 minutes and after sterilization, transferred in a laminar flow hood. Media were poured in petri plates up to 15 mL and left to solidify at room temperature in a laminar flow hood.

#### 2.3.3 Serial dilution

Test tubes were filled with 9 mL distilled water and covered with aluminum foil. Test tubes were sterilized in an autoclave at 121°C for 15 minutes and afterward left to cool down in a laminar flow hood. After cooling, treated milk samples were serially diluted following a 9:1 mL serial dilution method up to 9 folds, separately.

#### 2.3.4 Inoculation

After completing media preparation and serial dilutions, the spread plate method was followed to inoculate the last dilutions of each treated sample on the respective media. For inoculation, 1 mL of each last dilution of separate sample was poured on solidified media plate and spread carefully with a disposable spreader, and labeled.

#### 2.3.5 Incubation

Incubation typically refers to providing controlled conditions for the growth of microorganisms. After inoculation, all the petri plates were placed in an inverted position, in an incubator for 24-48 hrs. at 37°C for bacterial growth and at 25°C for 5 days for yeast and mold growth.

#### 2.3.6 Colony counting

When the incubation period is completed then the plates were removed from the incubator and colonies were counted under the colony counter  $\log_{10}$  CFU/mL was calculated applying the  $\log_{10}$  formula.

## 2.4 Physio-chemical Analysis of Milk Samples

### 2.4.1 pH Determination

An electronic pH meter was used for the determination of the pH of the milk sample as described by (Pham *et al.*, 2020; Tripathy *et al.*, 2019). A 10mL sample was taken in a beaker. Electrodes of pH meter were rinsed with distilled water then it was a dip in buffer solution of pH 4. Electrodes were again rinsed with distilled water and dipped in a buffer solution of pH 7. After calibration, electrodes were cleaned with distilled water, dried, and dip in the beaker having the sample. pH was checked on the meter.

### 2.4.2 Brix determination

A digital refractometer was used for the determination of brix. First, we calibrate the refractometer with distilled water by adding a few drops of water to the lens of the refractometer and then closing the lid the reading should be zero after calibration add a few drops of milk on the lens then close the lid and note the reading from a screen (Safari and Varaminian, 2019; Rincon *et al.*, 2020).

### 2.4.3 Titratable acidity

The titration method was used for the determination of titratable acidity as described by AOAC (2019). Phenolphthalein was used as an indicator as it is colorless with acid but gives pink color to the basic solution. A burette was filled with N/10 NaOH and the initial volume of NaOH was noted. A 10mL sample was poured into a flask and 2-3 drops of phenolphthalein were added. Mixing of

the sample was followed by titration of a sample against 0.1N NaOH. Titration was continued till the appearance of light pink color. Titration was stopped and the volume of NaOH used was noted. Values were put in the following formula to calculate acidity

#### Titratable acidity

$$= \frac{\text{Volume of } 0.1 \text{ N NaOH used} \times 0.009}{\text{Weigh of sample}} \times 100$$

#### 2.4.4 Color and sensory evaluation

The color quality of treated samples was calculated through a colorimeter and the parameters of color were  $L^*$ ,  $a^*$ , and  $b^*$ . To check the color quality, firstly 20mL of each milk sample was poured into a petri dish and then placed under the lens of the colorimeter at a minimum distance (1-2cm), button was pressed and let the light reflected on the product and readings were noted down (Milovanovic *et al.*, 2021).

$\Delta E = [(L^* - L_0)^2 + (a^* - a_0)^2 + (b^* - b_0)^2]^{1/2}$

All treated samples were organoleptically evaluated for color, flavor, taste, aroma, and overall acceptability by a panel of 20 experts from faculty NIFSAT, UAF. All the samples were given codes to avoid data biasing, and experts were asked to sip water between each sample to avoid the taste of the previous sample (Mbye *et al.*, 2020).

#### 2.5 Storage study

Pasteurized product was stored at refrigerator temperature for 16 days and the above-mentioned analysis were performed after 1, 4, 8, 12, and 16 days of storage.

#### 2.6 Statistical analysis

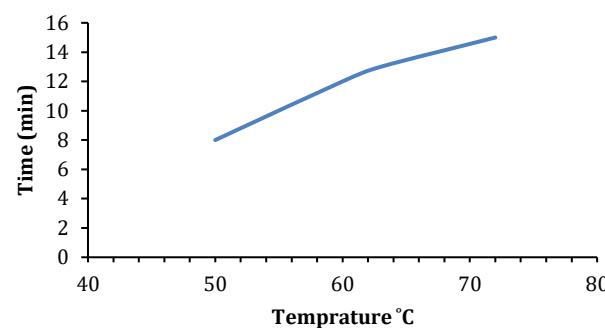
All the analyses were performed in triplicate and obtained data were subjected to statistical tools (statistics 8.1) (Montgomery, 2017). The analysis of variance (ANOVA) was checked at a confidence interval of  $\alpha = 0.05$ .

### RESULTS AND DISCUSSION

Sono-ohmic pasteurization was used in this research to pasteurize milk and prevent the loss of its beneficial nutrients. Milk was pasteurized at various temperatures (50, 60, and 63°C) and sonication periods (10, 15, and 20 minutes) with constant voltage (200V). The pasteurized milk was stored and analyzed for total plate count, coliform, yeast and mold, pH, brix, and color analysis were all examined in this study. In this study, time and temperature combination play a key role for milk pasteurization. As shown in figure 1, the time taken to reach at specific temperature 50, 60, 63°C was 5, 10, 12 minutes, respectively. The milk sample was treated with conventional method for 15 minutes at 72°C, to check the effect of thermal treatment on physicochemical and microbial attributes. The study showed pH value 7.5, titratable acidity 0.2, and the brix value was 5 after 7 days of storage at refrigeration temperature. The coliform value was  $11.90 \log_{10} \text{CFU/mL}$ , and yeast mold value  $11.80 \log_{10} \text{CFU/mL}$ , exhibited significant impacts ( $P < 0.05$ ) on milk quality. The total plate count value was  $11.95 \log_{10} \text{CFU/mL}$ .

**Figure 1**

*Relationship between time and temperature*

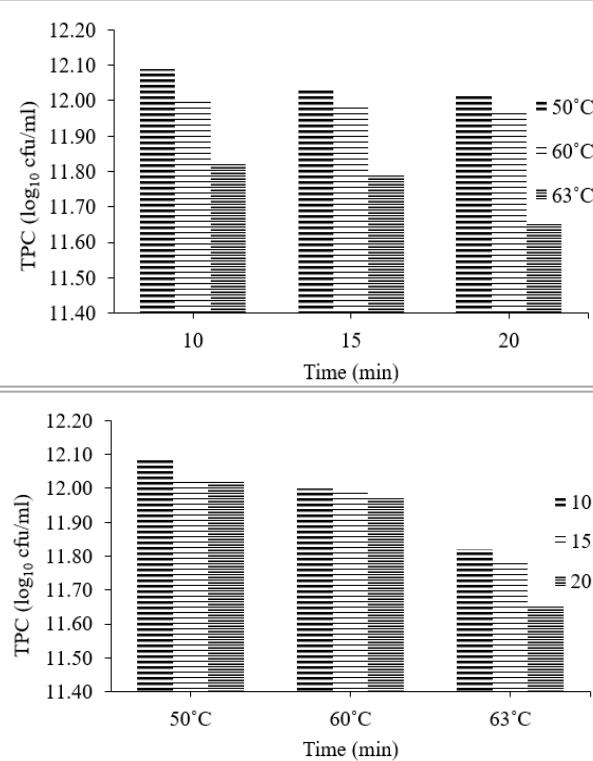


#### 3.1 Total Plate Count

Results indicated that the temperature has significantly ( $P < 0.05$ ) reduced the microbes during the sono-ohmic pasteurization process. At the temperature increase from 50°C to 63°C, the TPC value was reduced from  $12.09 \log_{10} \text{CFU/mL}$  to  $11.82 \log_{10} \text{CFU/mL}$  respectively. The decrease was 0.83% more when compared with 60°C and 2.5% more than 50°C. Likewise, the result indicated that at 50°C sonication for 10 and 15 minutes, the TPC value was decreased from  $12.09 \log_{10} \text{CFU/mL}$  to  $12.03 \log_{10}$ . Moreover, an increase in temperature and time has significantly reduced the CFU/mL value. The increase in sonication time has reduced TPC up to 3-fold. Similarly, the number of colonies reduced to  $11.66 \log_{10} \text{CFU/mL}$  when sonication time increased from 10 to 20 minutes at 63°C as shown in figure 2. The studies showed that the decreased in the colonies of TPC because of high temperature and sonication time in treated samples (Eazhumalai *et al.*, 2022).

**Figure 2**

*Total plate count of pasteurized milk applying sono-ohmic pasteurization*

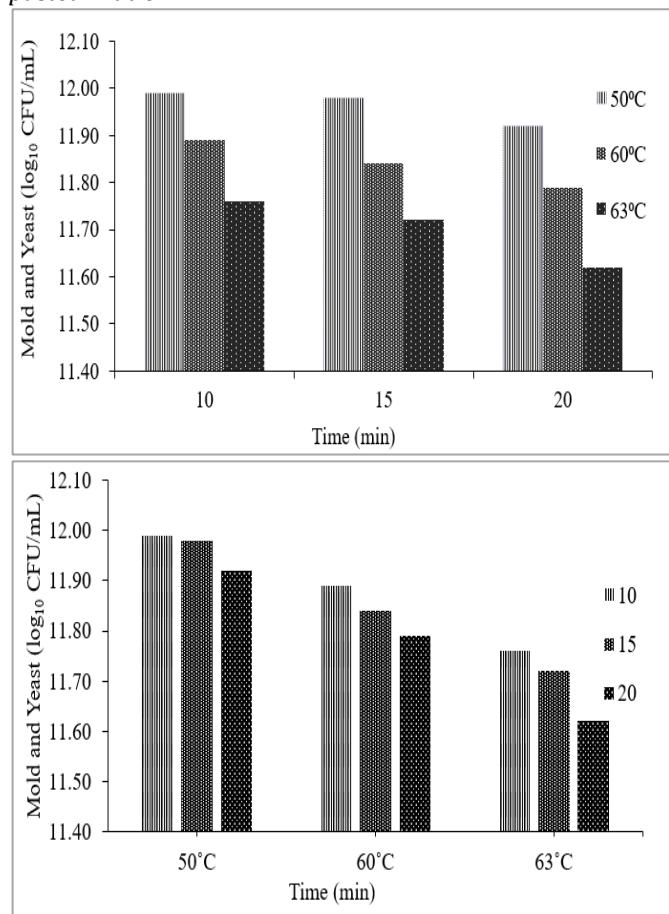


### 3.2 Mold and Yeast

Both mold and yeast are fungus forms yet they have different traits and uses (Awasti and Anand, 2020). The sono-ohmic pasteurization process results showed that the temperature considerably decreased the number of microorganisms. The mold and yeast values were  $11.99 \log_{10} \text{CFU/mL}$  at  $50^\circ\text{C}$ , and  $11.89 \log_{10} \text{CFU/mL}$  at  $60^\circ\text{C}$ , and further decreased to  $11.76 \log_{10} \text{CFU/mL}$  as the temperature rise to  $63^\circ\text{C}$ . The drop was 0.58% more than  $50^\circ\text{C}$  and 0.84% greater than  $60^\circ\text{C}$ . The outcome showed that sonication for 10 and 20 min at  $50^\circ\text{C}$  had dropped from  $11.99 \log_{10} \text{CFU/mL}$  to  $11.98 \log_{10} \text{CFU/mL}$ . A temperature rise ( $50\text{--}60^\circ\text{C}$ ) also considerably decreased the CFU/mL number as indicated in Figure 3. Associated with the CFU/mL value, the temperature rise caused the value to drop dramatically from  $11.99 \log_{10}$  to  $11.62 \log_{10} \text{CFU/mL}$ . Mold and yeast have decreased by up to three times because of the longer sonication period. When the sonication period rises from 10 to 20 minutes at  $63^\circ\text{C}$ , the number of colonies decreased to  $11.62 \log_{10} \text{CFU/mL}$ . Previous studies showed that the colonies of mold and yeast decreased because of change in electric field, temperature and sonication time in treated samples (Rocha et al., 2022).

**Figure 3**

*Mold and yeast of pasteurized milk through sono-ohmic pasteurization*



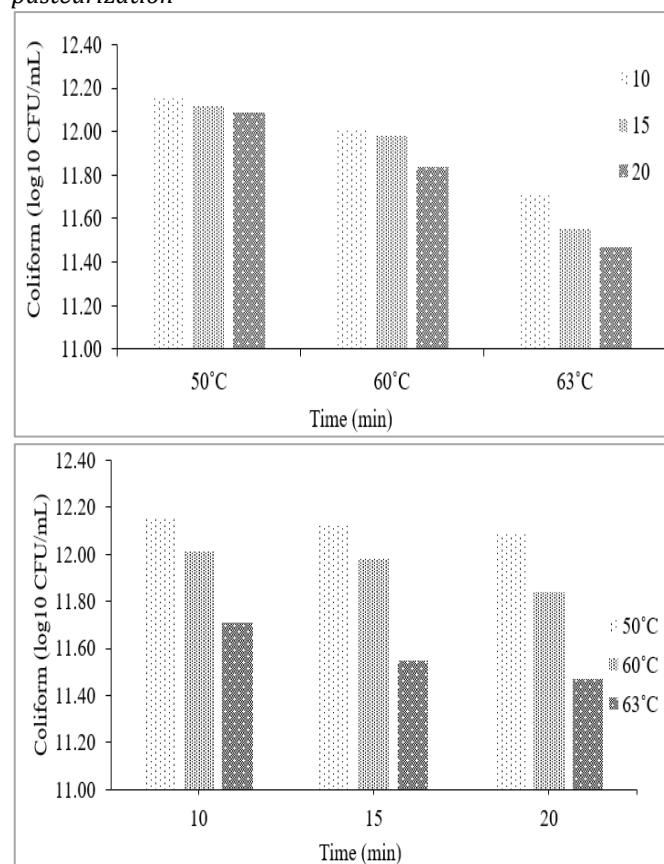
### 3.3 Coliform

A collective group of bacteria with similar traits is called coliforms and also known as facultative anaerobes (Martin et al., 2016). As indicated in figure 4, the sono-ohmic

pasteurization process results showed that the temperature considerably ( $P<0.05$ ) decreased the number of microorganisms. The coliform value was reduced from  $12.16 \log_{10} \text{CFU/mL}$  to  $11.01 \log_{10} \text{CFU/mL}$  at  $50^\circ\text{C}$  and  $60^\circ\text{C}$ , respectively. The outcome showed that sonication for 10 minutes at  $50^\circ\text{C}$  had  $12.16 \log_{10} \text{CFU/mL}$  and  $12.12 \log_{10} \text{CFU/mL}$  when the sonication period was extended to 15 minutes at  $50^\circ\text{C}$ . Similar to the CFU/mL value, the temperature rise caused the value to drop dramatically from  $12.01 \log_{10}$  to  $11.84 \log_{10} \text{CFU/mL}$ . The number of colonies decreased to  $11.47 \log_{10} \text{CFU/mL}$ , when the sonication period rises from 10 to 20 minutes at  $63^\circ\text{C}$ . Increased temperature and sonication time can put coliform bacteria under a lot of stress. When the temperature and sonication time exceeds their ideal range, leads to a decline in their number (Hashemi et al., 2019).

**Figure 4**

*Coliform of pasteurized milk through sono-ohmic pasteurization*



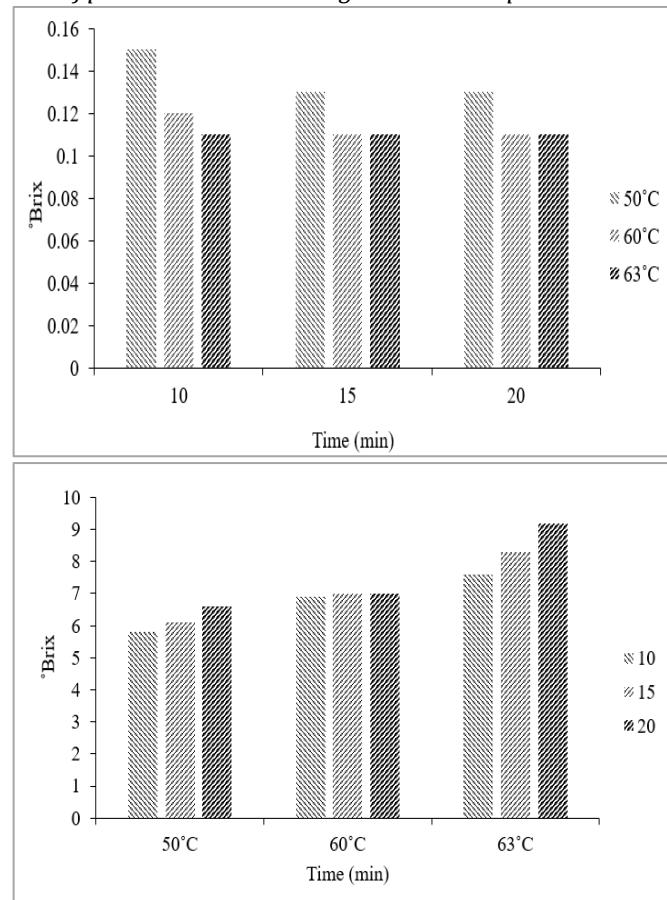
### 3.4 Brix

The sugar concentration of a fluid is, determined by the brix scale, typically expressed as a percentage by weight. The brix scale is not generally used to quantify milk. On average, milk has a 4-5% lactose level (Beldie and Moraru, 2021). The milk brix levels rise between 5.8 and 9.2 at various time intervals and temperatures of sono-ohmic pasteurization. The brix value was 5.8, 8.3 and 9.2 at  $50^\circ\text{C}$ ,  $60^\circ\text{C}$  and  $63^\circ\text{C}$ , respectively. The outcome showed that sonication for 10 minutes at  $50^\circ\text{C}$  had 5.8 brix. However, when the sonication period was extended to 15 minutes at the same temperature ( $50^\circ\text{C}$ ), the brix value rises to 6.1. Additionally, a temperature rise ( $50\text{--}60^\circ\text{C}$ ) considerably increased the brix level. Similar to the brix

value, the temperature rise caused the value to rise dramatically from 7 to 8.3 as shown in figure 5. The sonication period was extended from 10 to 20 minutes at 60°C. brix value had increased because of the longer sonication period. Similar to this, when the sonication period rises from 10 to 20 minutes at 63°C, the value of brix increases to 9.2. The outcomes of my research were comparable to those that were reported in the previous literature (Manzoor *et al.*, 2021).

**Figure 5**

Brix of pasteurized milk through sono-ohmic pasteurization

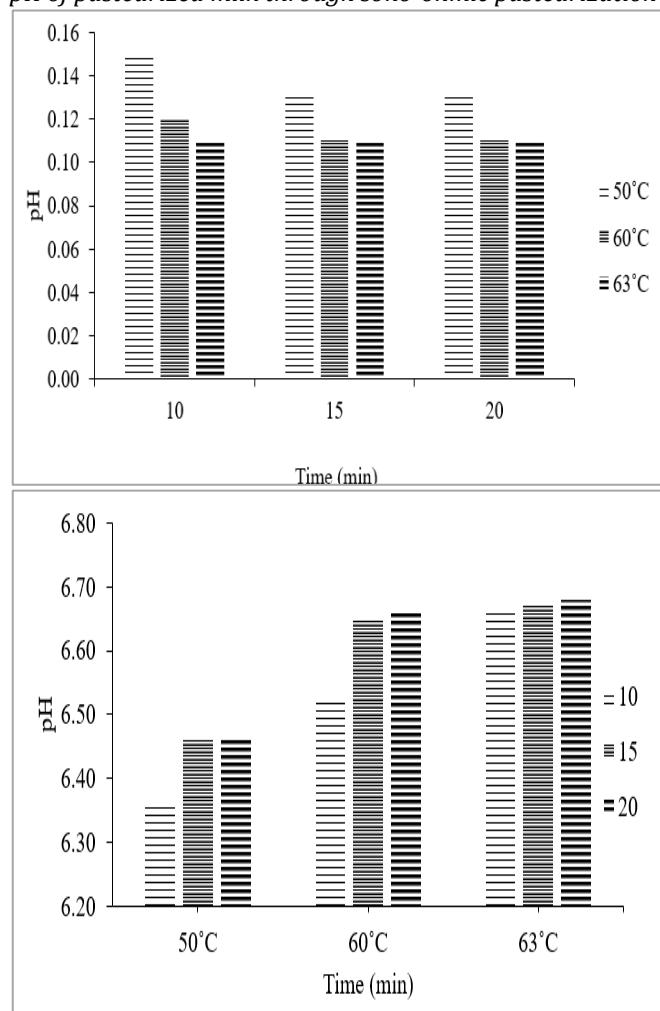


### 3.5 pH

The acidity or alkalinity of a solution is determined by its pH value. When pH value is below 7 it denotes acidity and above 7 denotes alkalinity (Marouf and Sara, 2018). When the temperature was 50°C, the pH value was 6.36, and it went up to 6.66 when the temperature was raised to 60°C. It then increased to 6.67 when the temperature was raised to 63°C. The results revealed that 6.36 pH and 6.46 pH was produced after 10 and 15 minutes of sonication at 50°C. Furthermore, the pH level was significantly raised by a temperature increase (50–60°C) that was 6.36 to 6.66 pH. Because of the prolonged sonication time, pH value climbed as shown in figure 6. Similar to this, the value of pH climbed to 9.2 when the sonication duration was increased from 10 to 20 minutes at 63°C. The outcomes of my research were comparable to those that were reported in the previous literature (Mennane *et al.*, 2007; On-Nom *et al.*, 2010). Previous studies showed that the increased in pH because of decreased titratable acidity in treated samples (Wibowo *et al.*, 2015).

**Figure 6**

pH of pasteurized milk through sono-ohmic pasteurization

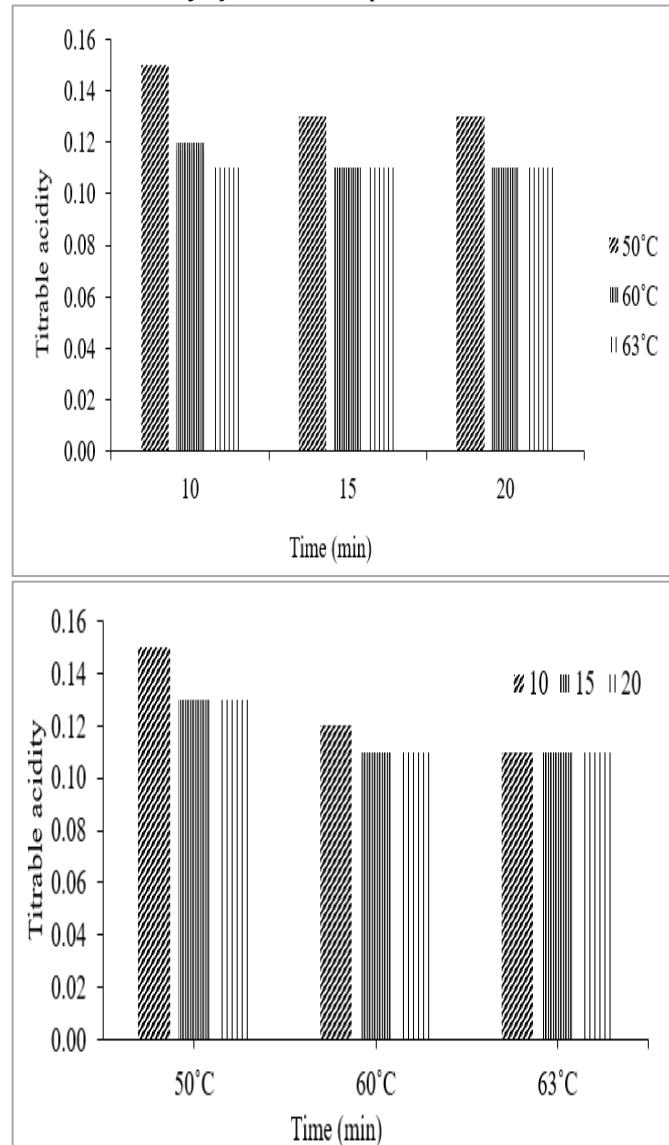
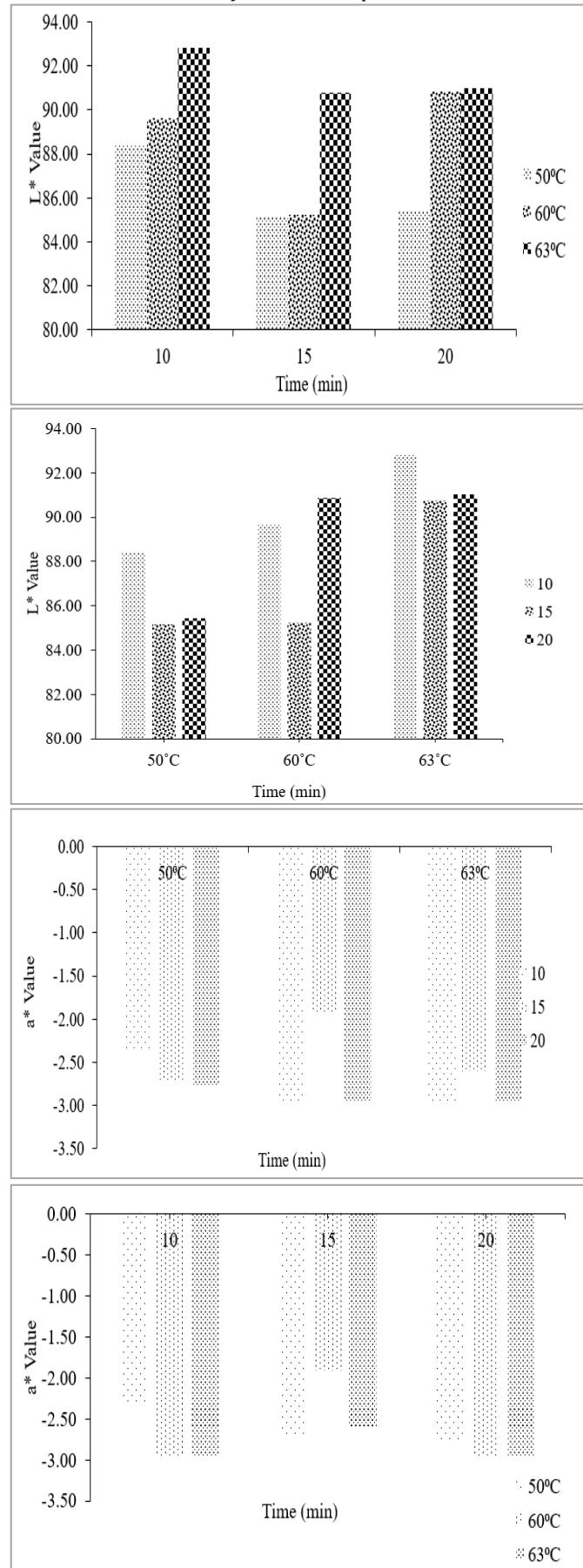


### 3.6 Titratable acidity

The amount of lactose acid generated in milk is an indicator of milk acidity. Yeast may thrive in acidic conditions and change acidic materials into nonacid ones (Toffanin *et al.*, 2015). Results indicated that the temperature has significantly ( $P<0.05$ ) reduced the titratable acidity during the sono-ohmic pasteurization process and ultrasonication is seen in Figure 7. The titratable acidity gradually decreased. The titratable acidity value was dropped from 0.15 to 0.11 when the temperature was increased from 50°C to 63°C. The results showed that after 10 minutes of sonication at 50°C, 0.15 titratable acidity was generated and 0.13 was generated when the sonication period is extended to 15 minutes at the same temperature (50°C). A temperature rise (50–60°C) also resulted in a considerable fall in acidity level. The sonication period was extended from 10 to 20 minutes at 60°C then the titratable acidity value dropped from 0.12 to 0.11 and it also dropped when the sonication time was increased from 10 to 20 minutes at 63°C. The outcomes of my research were comparable to those that were reported in the previous literature (Ziyaina *et al.*, 2018; Hassan *et al.*, 2009).

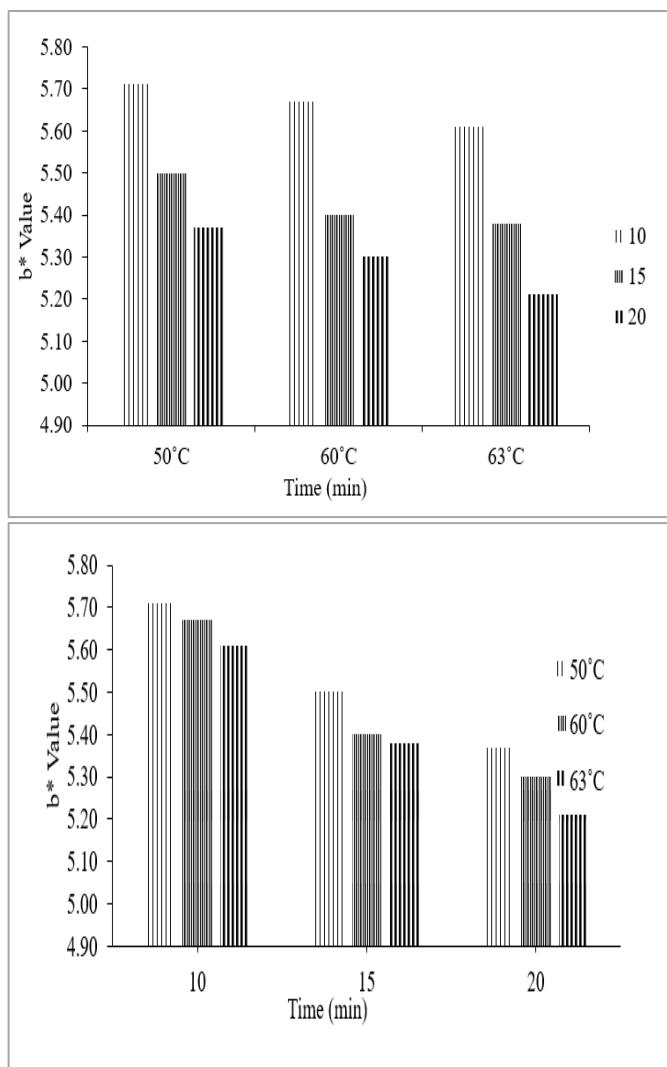
**Figure 7**

Titratable acidity of sono-ohmic pasteurized milk

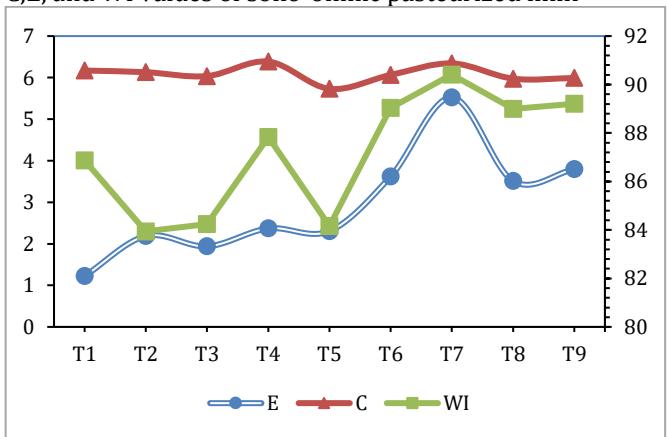
**Table**15  $L^*$ ,  $a^*$  and  $b^*$  value of sono-ohmic pasteurized milk

### 3.7 Color

One of the most important variables that immediately affects a customer is color. Determine the  $L^*$  value, which represents the product's brightness level, as well as values  $a^*$  and  $b^*$ , which represent the product's color coordinates. Color analysis can be carried out visually or using a colorimeter (Chudy *et al.*, 2020). The mean values for  $L^*$ ,  $a^*$ , and  $b^*$  are 4.7a, 4.8, 4.8a, and 4.9, 4.9a respectively. At 63°C and 20 minutes displayed a higher  $L^*$  value (92.81), whilst 60°C and 10 minutes displayed the lowest  $L^*$  value (85.16). All of the treatments had negative  $a^*$  values, which imply greenness. At 60°C and 15 minutes displayed the lowest  $a^*$  value (-1.92), while at 63°C and 20 minutes displayed the highest  $a^*$  value (-2.96). Every treatment had a positive  $b^*$  value, which denoted yellowness. The lower value for  $b^*$  (4.92) is at 60°C and 15 minutes whereas a higher value for  $b^*$  (5.77) is at 60°C and 20 minutes. According to (Chudy *et al.*, 2020)'s study, the  $L^*$ ,  $a^*$ , and  $b^*$  values of dairy products fell within the ranges of 69.6–95.46, -0.68–9.6, and 4.50–17.4, respectively (Cheng *et al.*, 2018).



**Figure 8**  
C, E, and WI values of sono-ohmic pasteurized milk

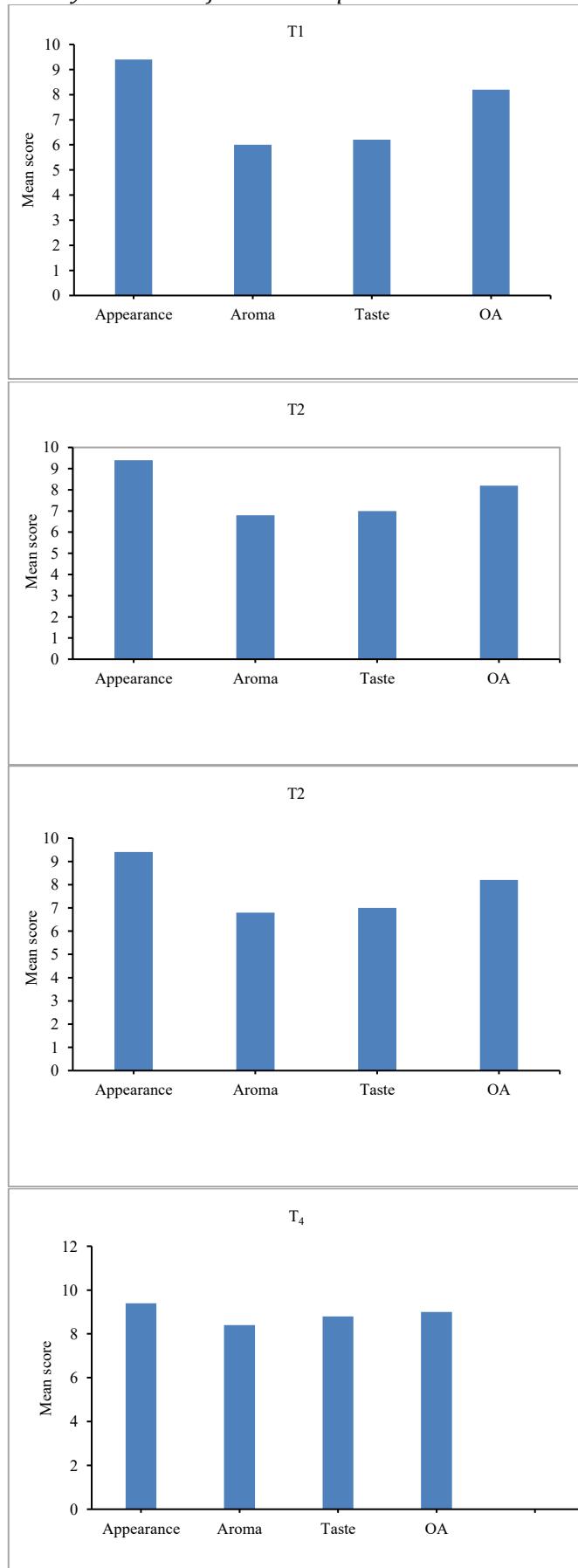


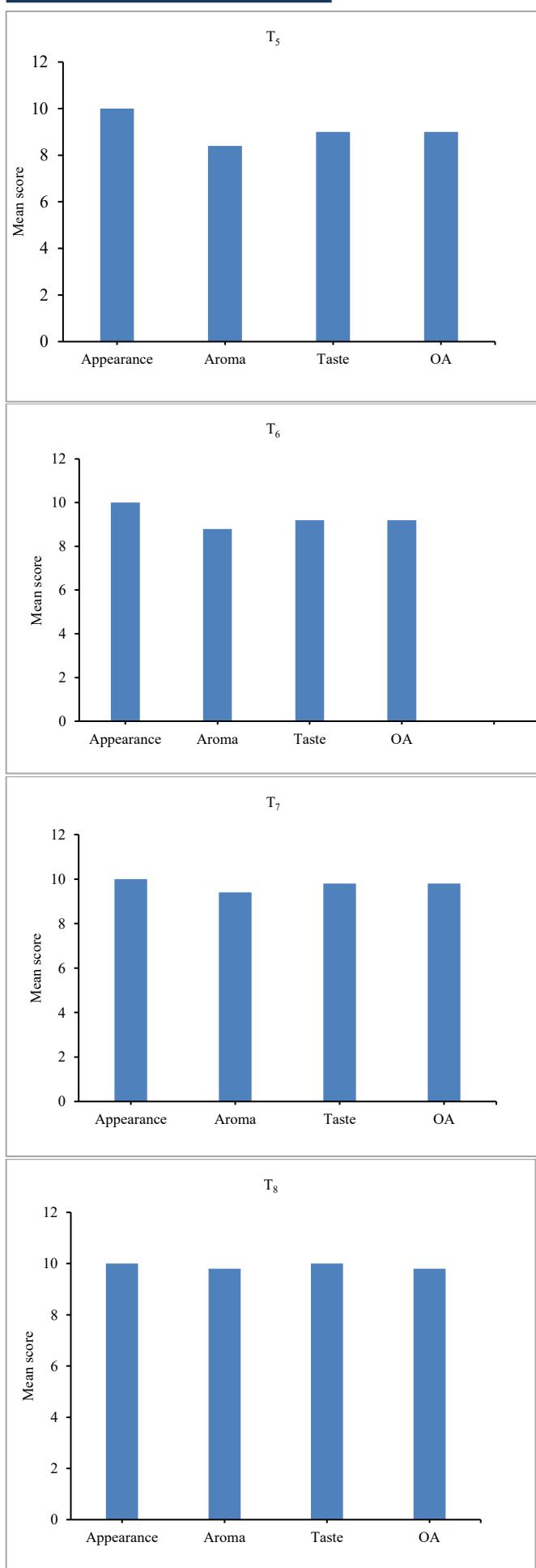
### 3.8 Sensory evaluation

The sensory assessment of pasteurized milk is critical in establishing its quality and customer acceptance. In this research, nine different milk samples labeled T<sub>1</sub> to T<sub>9</sub> were sensory analyzed to determine their characteristics. Participants (10) assessed the samples based on a variety of sensory criteria, including appearance, odor, taste, and overall preference. T<sub>9</sub> was the outstanding performer among these samples, receiving the highest ratings across all analyzed categories as indicated in Figure 9. The participants were delighted by its overall sensory profile,

making it the chosen option among the evaluated milk samples.

**Figure 9**  
*Sensory evaluation of sono-ohmic pasteurized milk*





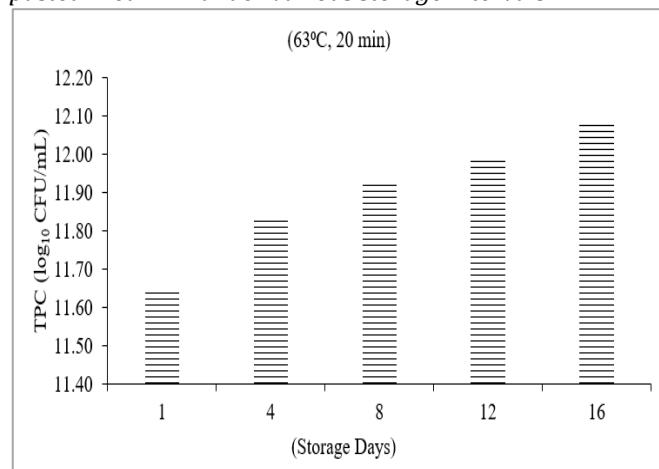
### 3.2 Storage Study

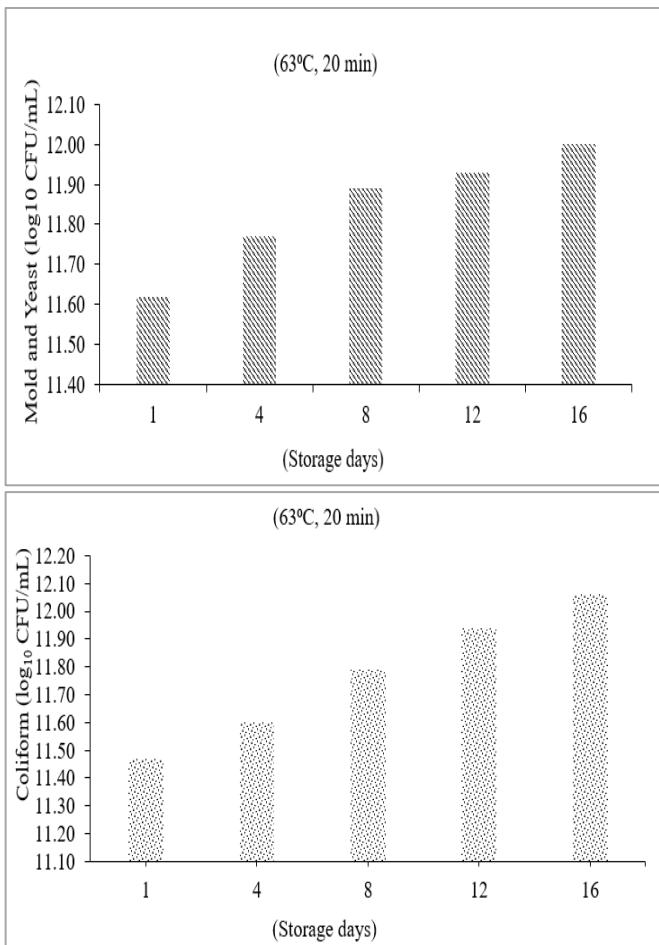
#### 3.2.1 Microbial Analysis

Sono-ohmic pasteurized milk storage was studied over a 16-day period to check the total plate count (TPC) value, with measurements being made on days 1, 4, 8, 12, and 16, at refrigeration temperature and the sample was treated for 20 minutes and the temperature was 63°C. For each of these days, TPC values were observed, ranging from 11.65 to 12.08  $\log_{10}$  CFU/mL. Similarly, storage research was done over 16 days to evaluate values of mold and yeast, with measurements recorded on days 1, 4, 8, 12, and 16 of the milk sample at refrigeration temperature. At 63°C, the range of mold and yeast measurements for these specific days was between 11.62  $\log_{10}$  and 12  $\log_{10}$  CFU/mL when treated for 20 min. This study was also evaluate the growth and viability of mold and yeast over time as compared with previous literature (Afzali *et al.*, 2020). During a 16-day storage review of coliform in sono-ohmic pasteurized milk, the coliform values taken on these specific days varied from 11.47 to 12.06  $\log_{10}$  CFU/mL. This study set out to evaluate the growth of coliform over time. The impact of storage conditions, potential sources of contamination, and necessary steps to maintain the quality and freshness of the milk might all be determined by monitoring coliform counts at various time intervals. The results of my study were comparable to previous literature (Sarkar, 2015).

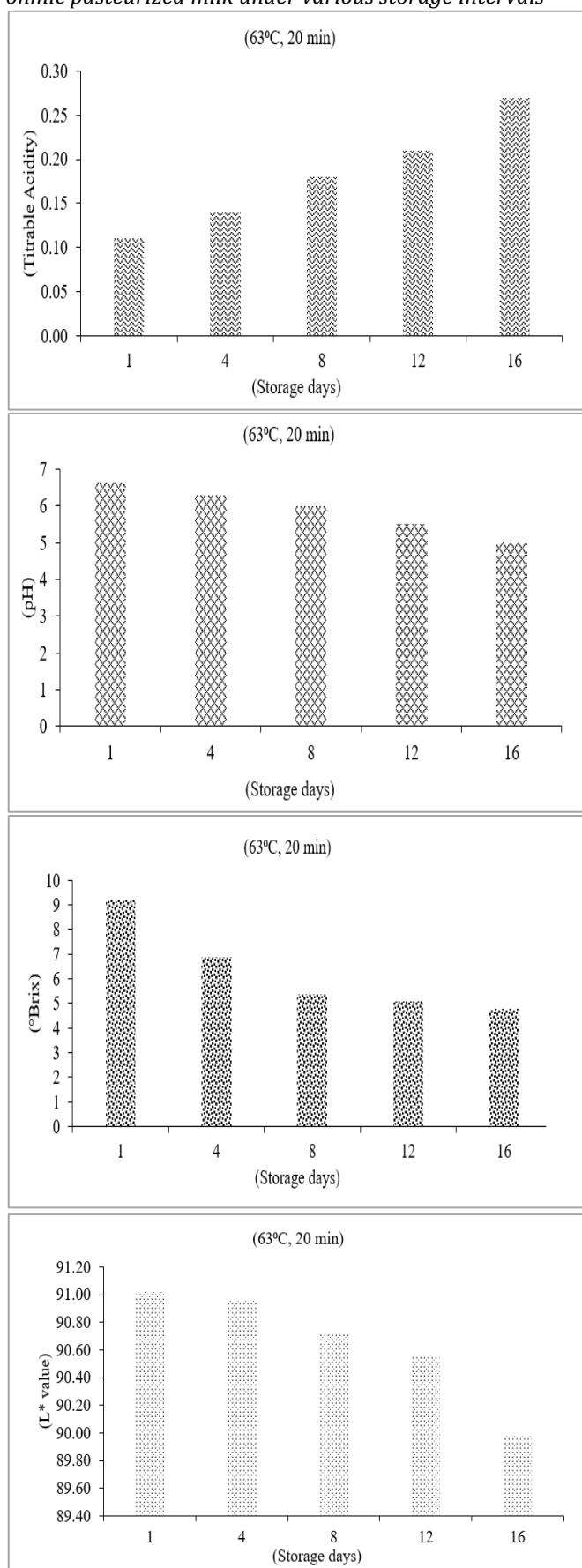
**Figure 10**

*TPC, Mold, yeast, and Coliform dynamics of sono-ohmic pasteurized milk under various storage intervals*



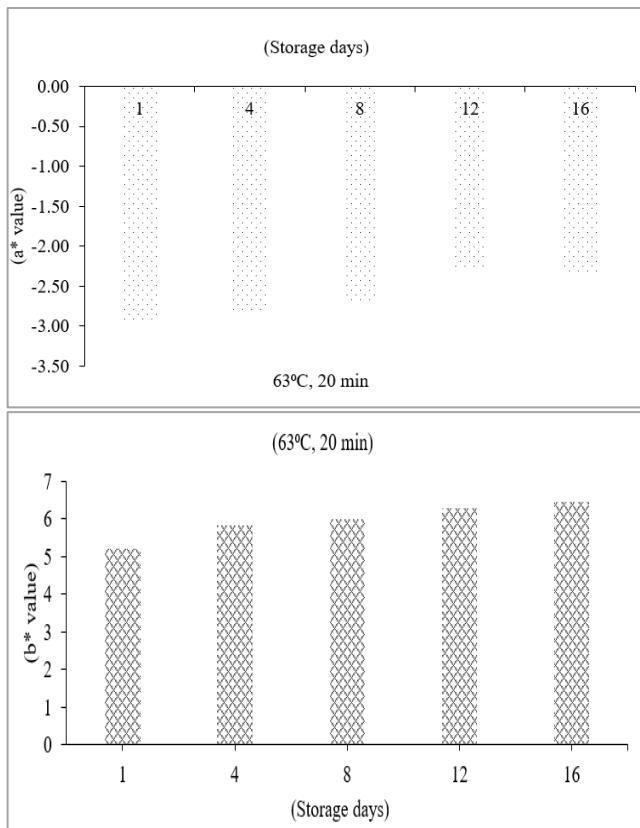
**Figure 11**

*Titratable acidity, pH, Brix, and Color dynamics of sono-ohmic pasteurized milk under various storage intervals*



### 3.2.2 Physicochemical analysis

A 16-days storage study of sono-ohmic pasteurized milk's titratable acidity was carried out, with data being obtained on 1, 4, 8, 12, and 16 days. The titratable acidity data made for each of these days varied from 0.11 to 0.27. This study looked at how the acidity levels of milk altered. The steady accumulation of acids over time, possibly caused by microbial activity and the emergence of spoilage organisms, is shown by the observed increase in titratable acidity with time. The results of my study were comparable to previous literature (Ziyaina *et al.*, 2018). Similarly, the pH value at first was 6.6 and steadily dropped during the next 4, 8, 12, and 16 storage days. The pH value decreased to 5 after 16 days of storage. The pasteurized milk's initial brix value was reported as 9.2 and decreased to 4.8 throughout the period of 1, 4, 8, 12, and 16 storage days. The results of this study were comparable to previous literature (Lemos *et al.*, 2023). Similarly, research was carried out to look into the variations in the color of pasteurized milk over time. The study focuses on three colour characteristics,  $L^*$ ,  $a^*$ , and  $b^*$ . The milk's initial  $L^*$  value was 91.02, which rapidly declined to 89.98 after 16 days at refrigeration temperature. Similarly, the  $a^*$  value dropped from -2.96 to -2.34, suggesting a decrease in redness, while the  $b^*$  value increased from 5.21 to 6.46, showing an increase in yellow tone. These colour attribute variations give useful insights into the colour stability and possible degeneration of pasteurized milk as comparable to previous studies (Ding *et al.*, 2023).



## CONCLUSION

The milk samples were treated with ohmic heating and sonication at constant voltages (200V) for three different stay periods of 10, 15, and 20 minutes at 50°C, 60°C, and 63°C. The results of all treatments show highly significant impact on total plate count, mold and yeast, and coliform. Whereas physiochemical tests of ohmic heating and sonication-treated samples were not highly significant effect on the pH, titratable acidity, and color. The brix was indicating significant effect. The best treatment of milk was treated at 63°C for 20 minutes and the 16 days storage study was evaluated at refrigeration temperature. Moreover, the results indicated that the treatment that was treated at 63°C, for 20 minutes was the best among all treatments. The storage study shows pH values, brix and titratable acidity indicating significant effect ( $P<0.05$ ). Increased value of total plate count, coliform bacteria, mold, and yeast had caused the deterioration of milk which is not good for health. Sensory evaluation of sono-ohmically treated milk samples was done to determine which treated milk sample has the best consumer acceptability. The sample that was treated at 63°C for 20 minutes has the best consumer acceptability according to the report. The storage life of sono-ohmically treated milk was increased compared it with the other techniques.

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