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Cold Plasma Technology and its Effect on the Microbial Content of Raw Milk

Widian Ali Mohammed[1], Nahla Tariq Khalid[1]*, Kadhim A. Aadim[2]

[1]Department of Food Science, College of Agricultural Engineering Sciences, University of Baghdad, Baghdad, Iraq; [2]Institute of laser for Postgraduate studies, University of Baghdad, Baghdad, Iraq.

Abstract | This study investigated the effect of cold plasma technology on the microbial content of raw milk using a nitrogen plasma jet operated at 15 kV and low temperatures (<8°C) for varying exposure times and sample volumes. The results revealed a substantial reduction in both bacterial and fungal counts compared with untreated samples. Treatments with volumes 20 and 25 mL had the lowest microbial values which were 0 $\times 10^3$ CFU/mL for both bacteria and fungi, respectively. The results showed no significant differences between the treatment 2-minute with 20 mL was compared with pasteurized milk. There were no significant differences between the treatments 9 and 10 minutes with 5 mL and pasteurized milk. Increasing the exposure time up to 9 minutes further enhanced microbial inactivation without thermal effects. Compared to conventional pasteurization, cold plasma treatment resulted in a similar reduction in microorganisms, yet with a greater retention of sensory characteristics and nutrients in the milk. These results demonstrate that the use of cold plasma is a promising new approach in lieu of conventional heat treatment.

Keywords | Milk, Composition of milk, Cold plasma technology

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***Correspondence** | Nahla Tariq Khalid, Department of Food Science, College of Agricultural Engineering Sciences, University of Baghdad, Baghdad, Iraq; **Email:** nahla.tariq@coagri.uobaghdad.edu.iq
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INTRODUCTION

Raw milk is high in nutrients. It is rich in protein, fat, vitamins and minerals. According to the Food and Agriculture Organization (FAO), global per capita dairy consumption will increase 12.5% by 2025 (Joshi *et al*., 2019). However, its composition also causes rapid perishability and susceptibility to microbial contamination by dangerous microorganisms. Pathogenic organisms such as *Escherichia coli*, *Listerian monocytogenesse* and *Staphylococci aureus* are frequently associated with raw milk, making it a serious threat to human health. Further, spoilage microorganisms shorten raw milk's shelf life and bring down its quality causing losses for the dairy industry. Conventional heat treatments such as pasteurization and ultra-high-temperature (UHT) processing are effective in cutting the level of microbes (Amaral *et al*., 2017). But they may also lead to undesirable changes such as in taste and smell or to degeneration of heat-sensitive nutrients (Claeys *et al*., 2013). Consumers have searched for dairy products that are safe, nutritious, practical, minimally processed, environmentally friendly, healthy, appetizing, economical, but that also have a long shelf life (Nahla and Makarim, 2018; Khalid *et al*., 2021; Tariq *et al*., 2023). Thermal processing improves the microbiological safety (Misra *et al*., 2017), but they extensively damage sensory, nutritional,

and physicochemical properties (Barba *et al*., 2012; Sayel *et al*., 2023), resulting in non-enzymatic browning, loss of vitamins and volatile flavour compounds, freezing point depression, and flavour changes in dairy products (Gurol *et al*., 2012). In addition, they require high-energy consumption, which compromises with the final product value to guarantee the profitability of the industry (Barba *et al*., 2017).

Plasma is the fourth state of matter, an ionized gases mixture, composed primarily of photons, ions, and electrons as neutral atoms with a simultaneous electric interaction between particles and atoms in its excited state (Olschewski, 2010; Khamsen *et al*., 2016; Pankaj and Keener, 2017; Park and Ha, 2018). It is commonly seen as bright fluorescent light; Langmuir (1928), coined the word "plasma" after seeing oscillations in ionized gas (Langmuir, 1928) and describing it as an "area having equilibrium charges of ions and electrons." Plasma status can naturally be found in aurora borealis, stars, fluorescent or neon gaslight, etc., Various materials can be prepared using the plasma jet method and the plasma laser method (Jamal *et al*., 2020; Mohammed *et al*., 2022). Non-thermal processes can meet microbial food safety standards and improve the physical, nutritional and sensory characteristics of the products, preserving unstable bioactive compounds and modulating enzyme activity (Amaral *et al*., 2017). These methods include ohmic heating (Cappato *et al*., 2017), high hydrostatic pressure (Barba *et al*., 2012), pulsed electric field (Da Cruz *et al*., 2010), pulsed-light technology (Abida *et al*., 2014), ultrasound (Ashokkumar *et al*., 2010), supercritical carbon dioxide technology (Amaral *et al*., 2017), and irradiation (Odueke *et al*., 2016). However, scientific knowledge on the utilization of cold plasma as an emerging non-thermal technology for dairy products is scattered and none of the previous studies reviewed these aspects. Consumer preferences are shifting towards foods that retain a sense of freshness and undergo minimal processing.

Therefore, there has been a growing interest in non-thermal methods of preservation. Cold plasma, also known as non-thermal plasma in some instances, is one of the promising techniques that are presently under consideration. This new technology presents a distinct opportunity to eliminate unwanted microorganisms in a way that does not involve high-temperature processing. Cold plasma is a distinctive substance consisting of a partially ionized gas abundant in ROS and RNS species, as well as UV photons, in addition to a large quantity of free electrons, ions, and UV photons. This combination of those components gives rise to a powerful antimicrobial property. In simple words, these agents cause damage to the microorganism's membrane, proteins, as well as the DNA itself, a process that ultimately leads to the inactivation of microorganisms (Misra *et al*., 2011; Thirumdas *et al*., 2018). It is a notable benefit of this technology that it is capable of operating near the environment's temperature in a way that allows microorganism inactivation with minimal change in the nutritional properties of milk.

Recent studies established promising results regarding the application of cold plasma in milk conservation. For example, Abbas *et al*. (2024) indicated that the cold plasma treatment effectively reduced microbials in raw milk, slowed acidity, as well as wholesome oxidative stability in comparison to pasteurization. Additionally, Nicol *et al*. (2020) illustrated effective, large-scale reduction of *Escherichia coli* and *S. aureus* in raw milk treated with atmospheric stress plasma jets. These results demonstrate the efficiency of cold plasma in enhancing the product's shelf life with optimal maintenance of product first-class qualities (Lee *et al*., 2024). Cold plasma treatment efficiency can be influenced by variables including plasma gas type, voltage treatment time, as well as raw milk composition. Moreover, certain microbials display higher resistance to plasma treatment, particularly spore-forming microbials that require extensive consideration. Moreover, despite primary studies that demonstrate minimal alterations in nutritional compositions, a thorough examination across long-term plasma impacts is expected to formulate a comprehensive determination of proteins, fats, vitamins, as well as bioactive components (Ekezie *et al*., 2017; Kanca and Avşar, 2023).

Cold plasma technological application is presently hindered in industries due to a range of unprecedented questions that require evident consideration. Moreover, despite successful results in inactivating microbials in raw milk, few studies evaluate long-term functional, nutritional, as well as sensory statuses of milk treated with this technology. Hence, a thorough investigation is presently obligatory in undertaking a comprehensive analysis towards the impacts of cold plasma in the microbials of raw milk, as well as formulating optimal efficiency parameters that hold a balance between achieving microbial safety in conjunction with maintaining wholesome milk qualities. This study aims to assess the effect of cold plasma on the microbial content of raw milk and its potential as a non-thermal maintenance technique.

MATERIALS AND METHODS

Preparation of milk samples

Whole milk from the campus dairy farm, a component of the College of Agricultural Engineering Sciences, University of Baghdad, was used in this investigation. Before treatment, the samples were placed on ice until they were delivered to the laboratory, where they were processed immediately.

The plasma jet electrode

The plasma jet electrode it is a locally made jazz found in the physics laboratories of the University of Baghdad, represents one of the connected electrodes in the high-voltage power supply, which is a cathodic electrode. This electrode resembled a large needle. The inner diameter of the needle was 3 mm, and it was made of stainless steel. The length of this needle is 4 cm, and it is connected directly through the electrical connection wires in the electrode cathode of the power supply as shown in Figure 2. The end of the plasma needle represents the flow of nitrogen gas and its transformation into cold plasma through a series of gas-ionization processes. In contrast, the beginning of the plasma needle represented a direct contact point with the connecting tube connected to the nitrogen gas flow regulator, as the inner diameter of the gas delivery tube corresponded to the size of the plasma needle starter, which was tightly connected to prevent nitrogen gas leakage Figure 3 shows the image and design for a plasma jet used in plasma generation and synthesizing metal nanoparticles with all the accompanying processes during generation and the flowing of plasma (Hussain, 2015; Naz *et al*., 2021; Abbas, 2023; Ahmed and Humud, 2025). The plasma jet electrode it is a locally made jazz found in the plasma physics laboratories for Postgraduate studies, Department of physics, college of science, University of Baghdad, represents one of the connected electrodes in the high-voltage power supply (Figure 1).

Figure 1: The plasma jet found in the plasma physics laboratories for Postgraduate Studies, Department of Physics, College of Science, University of Baghdad.

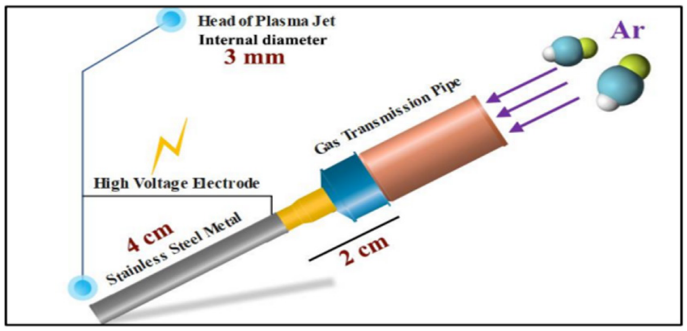

Figure 2: 3D image design of the plasma jet (Abbas, 2023).

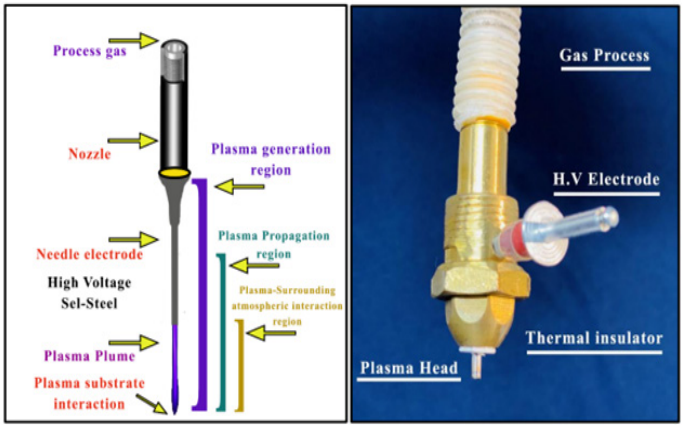

Figure 3: Design and image of the Plasma Jet represent all the accompanying processes during the generation and the flowing of plasma (Abbas, 2023).

Sample preparation and plasma treatment

Plasma application

The upper electrode, which was sterilized after each application, was rotated using an adjustable DC motor, and a motorized stirrer was attached to maintain application homogeneity (Kadhem, 2021; Muhsen *et al*., 2025; Jassim *et al*., 2025).

The temperature was monitored throughout the discharge application using a thermometer, and the system was shut Low-temperature plasma was applied to the contaminated milk solution by applying a high voltage between the upper electrode tip and the liquid surface (in which the other electrode was immersed) down whenever the temperature reached the allowed maximum temperature (35°C). The current of nearly 90 mA in the plasma corona resulted in nearly 10 A/cm^2. To cool the system, it was lowered to contact the surface of an icepack flow rate 2.5L/min.

Plasma-treated milk

The apparatus described above was placed in a laminar flow cabinet (Heal Force) to prevent contamination. Raw milk was treated using cold atmospheric pressure plasma using a plasma jet device at 15 kilovolts while maintaining a temperature below 8°C to avoid thermal effects on milk components.

Pasteurized milk

High Temperature Short Time (HTST) pasteurization was applied at 72°C for 15 seconds using a laboratory pasteurizer, followed by rapid cooling to 4°C.

Autoclaved milk

Milk was sterilized using an autoclave at 121°C and 15 psi for 15 minutes, with gradual cooling to prevent foam formation.

UHT milk (ultra high temperature)

UHT milk samples were obtained from the local market from different companies, which were treated at 135-150 °C for 2-4 seconds according to manufacturer standards.

Sample treatments

Prepare sterile 50 ml containers. 5, 10, 15, 20, 25, and 30 mL of raw milk were pipetted into containers and treated using a cold plasma apparatus for 1, 1.5, 2, 2.5, 3, 4, 5, 7, 9, 10, and 15 minutes. exposure time with constant stirring at room temperature. After treatment the cold, plasma-treated milk was stored in the refrigerator until subsequent analyses were performed. Experiments were carried out three times. Control and treated samples were stored at 8 °C prior to microorganisms analyses. To determine if the plasma treatment causes changes in milk microorganisms, analysis of milk bacterial, mold, and yeast colony counting.

Sample collection and preparation

Milk samples were collected in sterile 50 mL containers. Samples were transported to the laboratory in a cooling box at 4±1°C within two hours of collection. Detailed information about each sample was recorded including source, collection time, temperature, and treatment type.

Before analysis, samples were removed from refrigeration and allowed to reach room temperature for 30 minutes. Each sample was mixed with gentle circular motion 25 times to ensure homogeneous distribution of microorganisms, then left for 15 minutes to allow bacterial dispersion before starting analysis.

Serial dilution preparation

This study was conducted at the Al-Nahrain University Laboratories, Biotechnology Research, during the period from January 2024 to October 2024. This experiment sought to evaluate the microbiological effects of various treatment processes of milk based on bacterial, mold, and yeast colony counts. Serial dilutions were prepared using sterile peptone solution as a diluent. One milliliter of the original sample was taken using a sterile pipette and added to 9 mL of sterile peptone solution in a sterile test tube to obtain the first dilution (10^{-1}). The solution was mixed using a vortex mixer for 15 seconds at medium speed, then 1 mL from the first dilution was taken and added to 9 mL of sterile peptone solution to obtain the second dilution (10^{-2}). This process was repeated to obtain serial dilutions up to 10^{-6} as needed.

Total aerobic bacterial count

The pour plate method was used according to international standards. Plate Count Agar (PCA) medium produced by Oxoid UK (Code: CM0325) was used for total aerobic bacterial count. PCA medium was prepared and dissolved in water bath at 45±1°C. One milliliter of each dilution was poured into sterile petri dishes in triplicate for each dilution. Fifteen to twenty milliliters of molten PCA medium were added to each plate, taking care not to form air bubbles. The sample was mixed in circular motion clockwise and counterclockwise for 10 seconds to ensure that the microorganisms in the medium are evenly distributed. Plates stood at room temperature for 15 minutes to allow them to solidify, then the plates were inverted and kept in the incubator at a temperature of 30±1°C for a period of 48±2 hours. Bacterial colonies that formed visible colonies after the incubation period were measured using a colony counter (Mai *et al*., 2018).

Molds and yeasts count

Pour plate technique is done as per international standards. For mold and yeast count, Potato Dextrose Agar (PDA) medium obtained from Difco USA (Code No.: 213400) is employed. PDA medium is prepared after adding a concentration of 100 mg/L of the antibiotic chloramphenicol to inhibit bacterial proliferation. Medium is sterilized through autoclaving at a temperature of 121°C for a period of 15 minutes. The antibiotic was added to the medium after cooling to 45°C to avoid antibiotic decomposition by heat. One milliliter of each dilution was poured into sterile Petri dishes in triplicate for each dilution; 15-20 mL of prepared PDA medium was added.

The mixture was stirred in a circular motion and left to form solids. Plates were turned upside down in an incubator at a temperature of 25±1°C for a period of 5±1 days. Daily checks of the plates were done to see if mold rotifers and yeasts had formed (Mai *et al*., 2018).

Sterilization efficacy of plasma

The total bacterial, molds and yeast count of the microbial indicators were used for evaluating the effect of plasma sterilizing. The total bacterial, molds and yeast count of the milk samples with the treatment of UHT, pasteurization, and plasma at different performing conditions were characterized according to Mai *et al*. (2018). All the samples were with 10^{-1}, 10^{-2} and 10^{-3} dilution. Then, 1 mL of each diluted sample was poured into a sterilized culture dish (diameter= 90 mm) and then dispersed with 20–25 mL PCA and PDA agar counting medium in the liquid state. The media were inverted once they were solidified, Incubation was done using an incubator at 37°C (for coliform), and 25°C (for yeast, mold) for 48 hr in a constant temperature incubator. The sterilization effect of milk with plasma treatment was evaluated. The experimental analysis was performed in triplicate, and the result was represented as log CFU/mL.

Quality control

Control samples

Positive and negative control samples were used for each

analysis batch. The positive control sample included a known contaminated milk sample, while the negative control sample included sterile saline solution. Empty control plates were used to test the sterility of culture media.

Media quality testing

Each batch of culture medium was tested to ensure sterility and growth suitability. Samples from each medium were incubated without adding microbial samples to ensure the absence of contamination in the medium. Media ability to support growth was also tested using reference bacterial strains.

Reading and calculations

Counting criteria

Plates containing 30-300 colonies of bacteria and 10-100 colonies of molds and yeasts were counted. A colony counter with appropriate lighting and a magnifying lens was used for accurate counting. Yeasts were distinguished from molds based on morphological characteristics (Mai *et al*., 2018).

Statistical calculations

Colony Forming Units (CFU/mL) were calculated using the equation:

$$CFU/mL = (\Sigma c / [(1.1 \times n_1) + (0.1 \times n_2)]) \times d$$

Where; Σc = sum of colonies counted in all calculated plates, n_1= number of plates counted in first dilution, n_2 = number of plates counted in second dilution, d = dilution factor corresponding to n_1

Statistical analysis

SPSS version 26.0 was used for statistical analysis. One-way ANOVA was applied to compare the means of microbial counts between different treatment types. Duncan's test was used for multiple comparisons to determine significant differences between groups at P≤0.05 significance level.

Data were transformed to decimal logarithm (log_{10}) before statistical analysis to ensure a normal data distribution. Mean and standard deviation were calculated for each group and results were represented in appropriate tables and graphs (de Mendiburu, 2023).

RESULTS AND DISCUSSION

Effect of volume factor on plasma exposed milk

The results in Table 1 show that applying cold plasma for one minute significantly reduced the number of both bacteria and fungi (both molds and yeasts) compared to the untreated control sample. This reduction in the number of microorganisms was observed to be more pronounced in smaller sample volumes exposed to plasma. The treatment involving 5, 15, and 25 mL of plasma had the least count of microorganisms, indicating that increased surface exposure to plasma improves the efficiency of sterilization.

Optimal treatment volume

It is important to mention that optimal microbiological reduction was obtained at a treatment volume of 15 mL at 1 min. as the lowest values in bacteria and fungi reduction were obtained, which were neglected, 0, and 25×103 CFU/mL, respectively.

The optimal volume required in a medium to obtain optimal plasma exposure as well as high efficiency of energy transfer is 15 mL.

This sharp reduction can be explained by the bactericidal properties of the reactive oxygen and nitrogen species (ROS, RNS), which are produced by plasma. Reactive species destroy the microorganisms' cell walls, as well as microorganism protein structures and DNA. This is in line with recent studies on plasma treatment (Yahaya *et al*., 2021; Wang *et al*., 2022).

It was also found that yeasts and mold had a greater relative resistance than bacteria. This is true since fungi have a higher complexity in their cell walls than bacteria, making them relatively resistant to the oxidative activity of plasma (Rahnavard *et al*., 2024).

These findings verify that the treatment of milk by cold plasma is a promising alternative to conventional technologies that use heat treatment to reduce the bacterial load in milk while maintaining the qualities of milk. This is in agreement with earlier studies that investigated the use of plasma in the sterilization of fluid (Kitsiou, 2023).

Data collected from Table 2 revealed that the increased exposure time of the sample to the cold plasma to two minutes caused a higher reduction in both bacterial and fungal (yeast and mold) numbers than those in the untreated sample (control).

Volumes of treatment (20 and 25 mL) marked the lowest values of microbe counts that stood at 0 ×103 CFU/mL both in bacteria as well as fungi, indicating the high efficiency of cold plasma technology in getting rid of microbe populations. This marked enhancement in results was contributed by the greater surface area as well as the uniformity of the molecules in the liquid substance as a result of increased treatment time as well as a small volume of the sample. Though not studied in this experiment, the data is conclusive that increased treatment time brings about efficiency in the reduction of microbe counts. This notion is well backed by Wang *et al*. (2022).

Table 1: Effect of Volume factor on plasma exposed milk/ 1 minute.

Dilution3 CFU/mL			Dilution 2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi		Bacteria	Treatments 1 min. Volume mL
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeast	Molds		
TNTC	Nill	100.0 a	TNTC	Nill	150 a	TNTC	Nill	200 a	TNTC	Nill	TNTC	Control
26.67 c	Nill	31.67 c	TNTC	Nill	58.33 c	TNTC	Nill	100 c	TNTC	Nill	283.3 ab	5
TNTC	Nill	50.00 b	TNTC	Nill	50.0 c	TNTC	Nill	150 b	TNTC	Nill	300.0 ab	10
25.00 c	Nill	n	TNTC	Nill	63.33 c	TNTC	Nill	91.7 c	TNTC	Nill	150.0 c	15
TNTC bbbc	Nill	58.33 b	TNTC	Nill	100 b	TNTC	Nill	200.0 a	TNTC	Nill	300.0 ab	20
26.67 c	Nill	58.33 b	TNTC	Nill	100 b	TNTC	Nill	150.0 b	TNTC	Nill	241.7 bc	25
10.27	-	16.37	55.90	-	22.87	-	-	20.97	-	-	107.4	LSD

n: neglected; TNTC: Too numerous to count.

Table 2: Effect of Volume factor on plasma exposed milk/ 2 minutes.

Dilution 3 CFU/mL			Dilution 2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL ml / colonies			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi (colonies)		Bacteria	Treatments 2 min. Volume mL
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeast	Molds		
6 a	Nill	178.33 a	14.33 a	Nill	259.7 a	TNTC	Nill	300 a	TNTC	400 a	TNTC	Control
Nill	Nill	100 c	Nill	Nill	150 c	Nill	Nill	200 c	TNTC	Nill	283.3 b	5
Nill	Nill	58.33 d	Nill	Nill	108.3 d	Nill	Nill	200 c	15.33 e	Nill	300 b	10
Nill	Nill	n	Nill	Nill	55 e	Nill	Nill	100 d	TNTC	Nill	150 c	15
Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	18.33 e	Nill	53.3 d	20
Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	n	TNTC	Nill	38.3 d	25
n	Nill	143.3 b	4.33 b	Nill	200 b	27.667 b	Nill	250 b	TNTC	Nill	300 b	30
0.66	-	18.43	3.54	-	28.92	4.17	-	46.82	12.93	33.09	42.38	LSD

n: neglected; TNTC: Too numerous to count

This drastic reduction can be explained by the destructive activity of ROS and RNS produced by plasma. This is due to the damage inflicted on the microorganism's membrane as well as DNA by ROS and RNS produced by plasma, which has been concluded in recent studies on the use of plasma against fungi (Ambrico *et al*., 2020; Xu *et al*., 2023; Barkhade *et al*., 2025).

Moreover, the data revealed that yeasts and mold had a greater relative resistance than bacteria. This is in agreement with findings by various researchers that pointed out that the higher thickness of fungal cell walls and higher chitin content give fungal cells a higher ability to resist oxidative stress caused by plasma treatment (Los *et al*., 2020; Ribeiro *et al*., 2022).

A greater reduction of microorganisms occurred when a 20 mL sample of milk was treated for a period of 2 minutes This volume balances plasma exposure and energy transfer. It ensures the highest sterilization efficiency. Thus, this treatment was compared with pasteurized milk. Milk samples were exposed to plasma for 2 minutes, while pasteurized samples were treated at 63°C for 30 minutes.

These results strongly support the findings of contemporary studies on the effectiveness of cold plasma treatment as a non-thermal alternative for reducing the microbial load in milk while preserving its nutritional components and sensory properties without negative impact (Kitsiou, 2023).

Effect of time factor on plasma exposed milk

The results listed in Table 3 demonstrate that increasing the duration of milk with 5 mL exposure to cold plasma resulted in a gradual and continuous decrease in the numbers of both bacteria and fungi (molds and yeasts), compared to the untreated sample.

Treatment periods ranging from 1 to 10 minutes significantly reduced the microbial load, with the 10-minute period being the most effective which was neglected, achieving the lowest values for bacteria tested,

Table 3: Effect of time factor on plasma exposed milk/ volume 5 mL.

Dilution3 CFU/mL			Dilution2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi (colonies)		Bacteria	Treatments 5ml Time (min.)
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeasts	Molds		
TNTC	Nill	243 a	TNTC	Nill	300 a	TNTC	Nill	TNTC	TNTC	Nill	TNTC	Control
n	Nill	63.3 bc	TNTC	Nill	143.3 b	10.33 e	Nill	200 b	26.7 e	Nill	300 b	1
Nill	Nill	68.3 bc	Nill	Nill	133.3 b	Nill	Nill	200 b	25 e	Nill	300 b	2
Nill	Nill	100 b	Nill	Nill	150 b	Nill	Nill	200 b	TNTC	Nill	300 b	3
TNTC	Nill	50 c	TNTC	Nill	100 bc	TNTC	Nill	200 b	TNTC	Nill	300 b	5
50 b	Nill	46.7 c	TNTC	Nill	75 c	TNTC	Nill	150 b	TNTC	Nill	250 c	7
18.7 bc	Nill	48.3 c	26.67 c	Nill	65 c	TNTC	Nill	141.7 b	TNTC	Nill	200 d	9
8 c	Nill	n	20 c	Nill	45 c	TNTC	2.67 a	197.3 b	TNTC	6.67 a	250 c	10
40.58	-	49.01	45.72	-	58.03	59.72	1.27	86.1	40.67	1.87	40.48	LSD

n: neglected; TNTC: Too numerous to count.

while the counts of yeast were 8× 10^3, and molds was 0. In studies of exposure times of between 30 seconds to 10 minutes, results indicated that higher plasma treatment times result in a noticeable difference.

It is important to mention that from the results, continued exposure of over 9 minutes (10 minutes) contributed towards a significant reduction. This is believed to be because of achieving a point of balance between the rate of production of effective molecules as well as their stability in the liquid phase, as revealed by recent findings in plasma treatment of milk. This is in agreement with the findings that increasing exposure time leads to increased efficiency in the inactivation of microbes by plasma treatment, where this efficiency is expected to slow down over time. A recent finding suggests that treatment time of 10 minutes increased the total plate count reduction, with a consequent increase in the shelf life of milk to 15 days without affecting nutritional values (Lee *et al*., 2024).

It is important to note that the results indicated that exposure for periods of over nine minutes (10 minutes) led to a significant reduction as well. A comparison between the treatment methods and pasteurized milk necessitated the use of milk pasteurized at a temperature of 63 degrees for a period of 30 minutes, as well as milk exposed to plasma at a volume of 5 milliliters for a period of nine minutes and ten minutes.

Evidence shows various mechanisms in this process, as Mumtaz *et al*. (2023) identified that higher plasma on-time resulted in a marked increase in the level of ROS/RNS, thereby affecting cell viability and ATP. It is evident that higher irradiation time leads to greater DNA damage, as observed by Sulaiman and Sulyman (2021).

These findings verify that time is a key factor in increasing the microbe-killing power of the cold plasma technology process in milk, making this technology promising in elevating milk safety as well as in the maintenance of milk qualities (Kitsiou, 2023).

From the results in Table 4, the exposure time of the milk to the plasma with a volume of 20 mL is a key factor that affects the reduction of microorganism counts. There is a marked reduction in the counts of bacteria, mold, and yeast as the exposure time increases, with the greatest efficiency obtained at an exposure time of 4 minutes. This efficiency of plasma technology can be linked to the generation of oxidative stress in microorganisms that prevents their growth.

As indicated in Table 4, the optimal treatment time of milk with plasma is 4 minutes. This time span did not record any bacterial or mold and yeasts' growth. There existed no significant difference between this treatment time and the treatment time of 2 minutes. Bacterial count = 0, Mold and Yeast count =0 after dilution factor 3.

However, the values beyond the 4-minute mark fluctuated to a certain extent, which could be ascribed to either the adaptation of microorganisms to the environment or secondary contaminants that came into play during the treatment process. These values agree with recent studies that revealed the efficiency of Cold Plasma technology in food processing (Ribeiro *et al*., 2022).

This results in severe damage to the cell wall as well as the destruction of the genetic material, thereby resulting in a reduction in the number of living cells. Moreover, Puligundla and Mok (2017) stated that plasma is effective in eliminating the microbic load in dairy products in particular,

Table 4: Effect of time factor on plasma exposed milk/ volume 20 mL.

Dilution3 CFU/mL			Dilution2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi (colonies)		Bacteria	Treatments 20 ml Time (min.)
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeast	Molds		
TNTC	Nill	50 bc	TNTC	n	100 abc	TNTC	n	216.7 ab	TNTC	n	283.3 a	Control
8.3 bc	Nill	41.7 cd	25 b	Nill	94 abc	TNTC	Nill	166.7 abc	TNTC	Nill	250 ab	1
Nill	Nill	n	Nill	Nill	55 cd	Nill	Nill	100 bcd	Nill	Nill	150 cd	1.5
Nill	Nill	n	Nill	Nill	38.3 cd	Nill	Nill	60 cd	5 e	Nill	110 d	2
Nill	Nill	n	Nill	Nill	59.3 bcd	Nill	Nill	93.3 cd	Nill	Nill	135 cd	2.5
Nill	Nill	90 a	Nill	Nill	163.3 a	Nill	Nill	226.7 a	Nill	Nill	268.3 a	3
Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	N	4
10 bc	Nill	n	TNTC	Nill	101.7abc	TNTC	Nill	216.7 ab	TNTC	Nill	300 a	5
20.7 b	Nill	n	25.3 b	Nill	75 abcd	TNTC	Nill	150 abc	TNTC	Nill	200 bc	10
4.7 bc	Nill	58.3 b	TNTC	Nill	150 ab	TNTC	Nill	233.3 a	TNTC	Nill	283.3 a	15
16.69	-	15.7	41.07	0.54	90.8	54.66	0.54	122.6	35.32	-	66.3	LSD

n: neglected, TNTC: Too numerous to count.

Table 5: Effect of temperature factor on plasma exposed milk.

Dilution3 CFU/mL			Dilution2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi (colonies)		Bacteria	Treatments 20 mL ^{o}C 2 min
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeast	Molds		
Nill	Nill	n	Nill	Nill	60 b	Nill	Nill	100 c	TNTC	n	216.7 b	Control
Nill	Nill	n	Nill	Nill	50 b	18	Nill	108.3 bc	15 c	n	200 b	15
Nill	Nill	n	Nill	Nill	60 b	n	Nill	150 b	TNTC	16.33 a	300 a	37
Nill	Nill	45 a	Nill	Nill	100 a	n	Nill	200 a	TNTC	7.67 b	300 a	50
-	-	9.41	-	-	16.31	-	-	48.99	14.29	1.087	35.95	LSD

n: neglected, TNTC: Too numerous to count.

while retaining their properties. Furthermore, Dash and Jaganmohan (2022) established that the optimal result is obtained at a volume of 100 mL with a time of 15 minutes as the exposure time of milk to plasma with increased volume, the time of exposure of milk to plasma with increased volume resulted in effective reduction of microbe at 100 mL as well as in 50 mL as well as in 150 mL, stressed upon the wide spectrum of antimicrobial activity of plasma that embraces various groups of microorganisms like bacteria, molds, as well as yeasts, this is in support of the findings of this experiment as well.

Temperature's impact on plasma's ability to reduce microbial

The results presented in Table 5 indicate a clear relationship between temperature and the efficiency of plasma in reducing the microbial load in milk. Treatment at low temperatures showed a significant decrease in the number of bacteria and yeasts compared to the control (untreated) sample. This indicates that cold plasma is very effective at these low temperatures in inhibiting the growth of microorganisms. This effect is primarily due to the strong oxidative effect of the active particles produced by the plasma.

In contrast, it was observed that raising the temperature to higher levels 50 ^{o}C resulted in a relative increase in the number of bacteria which was 45 x 10^3. This potential increase could be explained by the declining effectiveness of the plasma active particles with increasing temperature, or perhaps by stimulating the growth of some more heat-resistant microbial species.

Gurol *et al*. (2012) have conducted an evaluation of the decontamination efficacy of low-temperature plasma on Escherichia coli present in milk with varying fat concentrations. Subsequent to the plasma treatment, the population of *E. coli* in fresh milk exhibited a reduction exceeding threefold, while the physicochemical properties of the fresh milk remained largely unaffected. The samples

subjected to treatment exhibited no evidence of bacterial proliferation throughout a one-week storage duration. Kim *et al*. (2015) treated fresh milk samples, inoculated with *Escherichia coli, Listeria monocytogenes,* and *Salmonella typhimurium*, with plasma and recorded a significant bacterial count reduction after 5 and 10 minutes.

These observations are in agreement with recent studies, including the one conducted by Laroussi (2020), which showed that cold plasma effectiveness depends on the surrounding temperature. It was observed from such studies that the highest efficiency of microbial load reduction occurs within a range of low to moderate temperatures, which would mean a very efficient sterilization process with regard to keeping the quality and properties of milk, as explained by Zou *et al*. (2025).

Comparison of plasma-exposed milk with pasteurized milk

The findings of Table 6 reflect that the number of bacteria and fungi significantly decreased in plasma treated milk with a volume of 5 mL when compared to the control (untreated). The highest efficiency of bacterial reduction, molds, and yeasts that were neglected, 0, and 8 x 10^3 was achieved by the 10-minute treatment. Also, there was no noticeable difference between pasteurization and treatments 9 and 10.

This result confirms the effectiveness of plasma technology in inactivating microbial cells. An oxidative process is likely responsible for this effect, where free radicals and active particles are created. Plasma can reduce microbial load as effectively as standard pasteurization. Such similarity, it follows, may mean that cold plasma has a place as a valuable method for non-thermal milk treatment. Cold plasma can significantly reduce microbial contamination in milk without harming its quality or nutritional value. Cold plasma has shown the potential to perform equally well in reducing the microbial load in milk, compared to traditional pasteurization, hence qualifying as a non-thermal alternative. Evidence of microbial reduction includes a maximum of 1.33 log reduction for coliforms observed by Dashab and Jaganmohana (2022), while Manoharan *et al*. (2023) reached 2.2 log reductions for treatments combining atmospheric with low-pressure plasma techniques. For Abbas *et al*. (2024), cold plasma was effective against the growth of pathogenic bacteria, with results comparable to those from pasteurization.

These plasma treatments reduced microorganisms without compromising the quality of milk, preventing protein denaturation and nutrient losses, unlike other thermal methods of processing Nikmaram and Keener (2022). However, further optimization of the parameters within this plasma technology is still required for complete microbial elimination.

These conclusions are supported by other recent studies in the field. Rathod *et al*. (2021) confirmed the efficacy of cold plasma technology for bacterial and yeast inactivation in liquid products. Nikmaram and Keener (2022) also pointed out the high efficiency of this technology for the reduction of microbial contamination in dairy products without negative effects on either nutritional or sensory properties.

The results presented in Table 7 illustrating a comparison of the effect of cold plasma treatment of milk (20 mL volume) and conventional pasteurization on microorganisms such as bacteria, molds, and yeasts highlighted that microbial counts were significantly reduced. Milk samples treated with plasma for two minutes exhibited much-reduced microbial counts compared to a control sample that consisted of raw milk. As indicated, the bacterial total count fell from 283.3 colonies in raw milk to 100 colonies after plasma treatment. In pasteurized milk, the count was 157 colonies. The same table exhibited that the pasteurized milk and the plasma-treated milk sample did not differ significantly when the dilution attained was 3.

Table 6: Comparison of plasma exposed milk/ volume 5 mL with pasteurized milk.

Dilution2 CFU/mL			Dilution2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria	Fungi		Bacteria	Fungi		Bacteria	Fungi		Bacteria	Treatments 5 mL Time (min.)
Yeast $x10^3$	Molds	$x10^3$	Yeast $x\ 10^2$	Molds	$x\ 10^2$	Yeast $x10^1$	Molds	$x10^1$	Yeast	Molds		
TNTC	Nill	250.0 a	TNTC	Nill	300.0 a	TNTC	Nill	TNTC	TNTC a	Nill	TNTC	Control
18.7 b	Nill	48.3 b	26.7 b	Nill	65.0 b	TNTC	Nill	141.7 bc	TNTC b	Nill	200.0 b	9
8.0 b	Nill	n	20.0 b	Nill	45.0 b	TNTC	n	197.3 b	TNTC b	6.67	250.0 b	10
TNTC	Nill	n	TNTC	Nill	n	TNTC	Nill	65.0 c	TNTC b	Nill	91.7 c	Pasteurization
48.4	-	49.2	49.1	-	49.8	-	-	80.5	-	2.88	104.4	LSD

n: neglected, TNTC: Too numerous to count.

Table 7: Comparison of plasma exposed milk/ volume 20 mL with pasteurized milk.

Dilution2 CFU/mL			Dilution2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi		Bacteria	Treatments 20 Ml Time (min.)
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeast	Molds		
TNTC	Nill	150 a	TNTC	Nill	250 a	TNTC	Nill	300 a	TNTCa	Nill	TNTC a	Control
TNTC	Nill	n	TNTC	Nill	33.3 b	TNTC	Nill	50 b	TNTC b	Nill	100 a	2
Nill	Nill	n	13.3 b	Nill	45.0 b b	TNTC	Nill	98.3 b c	TNTC c	Nill	157 a c	Pasteurization
-	-	67.7	128.7	-	91.6	-	-	126.4	-	-	237.7 $^{N.S}$	LSD

TNTC: Too numerous to count, n: neglected

Table 8: Comparison of plasma exposed, sterilized and ultra high temperature treated milk.

Dilution2 CFU/mL			Dilution2 CFU/mL			Dilution 1 CFU/mL			Total count before dilution CFU/mL			Sample
Fungi		Bacteria x10^3	Fungi		Bacteria x 10^2	Fungi		Bacteria x10^1	Fungi		Bacteria	Treatments 20 mL Time (min.)
Yeast x10^3	Molds		Yeast x 10^2	Molds		Yeast x10^1	Molds		Yeast	Molds		
6 a	Nill	75 a	Nill	Nill	150 a	26.7 a	Nill	200 a	TNTC	6.33	300 a	Control
0 b	Nill	Nill	Nill	Nill	Nill	25 a	Nill	Nill	TNTC	Nill	Nill	3
Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	n	4
Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	sterilization
Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	Nill	UHT
2.93	-	20.34	-	-	40.68	7.43	-	40.68	42.54	2.61	40.71	LSD

n: neglected, TNTC: Too numerous to count

These data show that cold plasma is effective in reducing the microbial load. One of the most important advantages provided by this technology is that such partial disinfection can be reached without causing the serious negative effects that thermal treatments can have on the heat-sensitive milk components. Thus, it is a promising technology for preserving milk quality and microbial safety. These results are consistent with recent research confirming that cold plasma treatment inactivates microbes by generating active chemical species, such as reactive oxygen species (ROS), which attack microbial cell structures, limiting their growth and contributing to extending product shelf life. Furthermore, this technology is a safe, non-thermal alternative to pasteurization and helps reduce nutritional loss compared to thermal treatments (Punia Bangar *et al.*, 2022).

Comparison of plasma exposed milk, sterilized milk and Ultra high treated milk temperature

The results presented in Table 8 compared the effect of treating milk with cold plasma (for 3 and 4 minutes) and ultra-high-temperature (UHT) sterilization on microorganisms. The bacterial and yeast counts were significantly lower in the plasma-treated samples compared to the control sample, raw milk. For instance, the bacterial count reduced from 300 colonies in raw milk to 0 colonies after 3 minutes of exposure to plasma, while nill bacterial remnants were recorded after 4 minutes of treatment. On the other hand, no microbial colonies were detected in both sterilized and UHT-treated milk, which proved the efficacy of the ultra-high-temperature treatments. Yeast counts decreased in the 3- and 4-minute plasma treatments, respectively, with complete disappearance in all treatments except the control sample.

According to the table, there were no significant differences among the milk samples: plasma-treated, sterilized, and UHT.

These findings suggest that cold plasma treatment is one of the useful methods to reduce microbial contamination in milk. This effect may be justified by plasma's ability to create free radicals and active ions that damage microbial cell walls, resulting in internal microbial cellular structure damages (Abbas *et al.*, 2024). Even though plasma sterilization was slightly inferior compared to ultra-high-temperature sterilization, cold plasma preserves sensory properties and sensitive nutritional components to a greater extent than either pasteurization or sterilization (Neokleous *et al.*, 2022). Recent studies emphasize increasing exposure time as an important method of enhancing the efficiency of plasma in eliminating microbial contaminants while

preserving proteins and vitamins without significant adverse effects (Abbas *et al*., 2024).

CONCLUSION

Comparing the plasma-treated and pasteurized milk samples, it is observed that there is a considerable reduction in microbial counts in the plasma-treated samples especially between 9 to 10 minutes of treatment, due to free radicals. However, in the pasteurized samples, increased effectiveness in reduction of bacteria is observed due to direct interaction with heat. Cold plasmas have been observed to be promising methods for the preservation of milk to ensure proper sterilization without thermal effects. Thus, cold plasma technology is quite suitable for the sterilization of milk.

From the results obtained in the study, it's clear that the role of temperature is critical in increasing the effectiveness of cold plasma treatment on milk. At a temperature of 15 °C, there is a pronounced effect on the reduction of bacteria with moderate effects on the growth of yeast cells. This indicates that such temperatures create a conducive environment to improve the effectiveness of the plasma treatment on the reduction of the microbial load. Conversely, increasing the temperatures to 37 °C resulted in a relatively high growth of bacteria and yeast cells. This is because such temperatures offer a conducive environment for such growth. Additionally, the temperature of 50 °C exhibited high microbial concentrations. This is because the constituents of the milk can decompose and provide nutrients to facilitate the increased growth of microbes.

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NOVELTY STATEMENT

This study demonstrates, for the first time, that low-temperature nitrogen cold plasma can achieve complete microbial inactivation in raw milk at specific volume–time combinations comparable to pasteurization, while better preserving sensory and nutritional quality, highlighting cold plasma as a non-thermal, volume-dependent alternative to conventional heat treatment.

AUTHOR'S CONTRIBUTION

First and second Author's designed the study; third Author conducted the experiments; second and third Authors conducted analysis; All authors read and approved the final manuscript.

GENERATIVE AI AND AI-ASSISTED TECHNOLOGY STATEMENT

The authors declare that no Genrative AI was used in the creation of this manuscript.

CONFLICT OF INTEREST

The authors have declared no conflict of interest.

REFERENCES

Abbas HM, Altamim EA, Salama M, Fouad MT, Zahran HA (2024). Cold plasma technology: A sustainable approach to milk preservation by reducing pathogens and enhancing oxidative stability. Sustain., 16(20): 8754. https://doi.org/10.3390/su16208754

Abbas IK (2023). Design, construction and spectroscopic analysis of Ca, Mg, Zn, Cr, Co produced by plasma jet and their biological applications (Doctoral dissertation). University of Baghdad, College of Science, Department of Physics.

Abida JRBMFA, Rayees B, Masoodi FA (2014). Pulsed light technology: A novel method for food preservation. Int. Food Res. J., 21(3): 839–848.

Ahmed MH, Humud HR (2025). The influence of gas type and flow rate on plasma jet length and gas temperature. Iraqi J. Phys., 23(3): 128–136. https://doi.org/10.30723/ijp.v23i3.1305

Amaral GV, Silva EK, Cavalcanti RN, Cappato LP, Guimarães JT, Alvarenga VO, Cruz AG (2017). Dairy processing using supercritical carbon dioxide technology: Theoretical fundamentals, quality and safety aspects. Trends Food Sci. Technol., 64: 94–101. https://doi.org/10.1016/j.tifs.2017.04.004

Ambrico PF, Šimek M, Rotolo C, Morano M, Minafra A, Ambrico M, De Miccolis ARM (2020). Surface dielectric barrier discharge plasma: A suitable measure against fungal plant pathogens. Sci. Rep., 10: 3673. https://doi.org/10.1038/s41598-020-60461-0

Ashokkumar M, Bhaskaracharya R, Kentish S, Lee J, Palmer M, Zisu B (2010). The ultrasonic processing of dairy products. An overview. Dairy Sci. Technol., 90(2): 147–168. https://doi.org/10.1051/dst/2009044

Barba FJ, Esteve MJ, Frígola A (2012). High pressure treatment effect on physicochemical and nutritional properties of fluid foods during storage: A review. Comprehen. Rev. Food Sci. Food Saf., 11(3): 307–322. https://doi.org/10.1111/j.1541-4337.2012.00185.x

Barba FJ, Koubaa M, do Prado-Silva L, Orlien V, de Souza Sant'Ana A (2017). Mild processing applied to the inactivation of the main foodborne bacterial pathogens: A review. Trends Fd. Sci. Technol., 66: 20–35. https://doi.org/10.1016/j.tifs.2017.05.011

Barkhade T, Nigam K, Ravi G, Rawat S, Nema SK (2025). Investigating the effects of microwave plasma on bacterial cell structures, viability, and membrane integrity. Sci. Rep., 15: 18052. https://doi.org/10.1038/s41598-025-02312-4

Cappato LP, Ferreira MV, Guimarães JT, Portela JB, Costa AL, Freitas MQ, Cruz AG (2017). Ohmic heating in dairy processing: Relevant aspects for safety and quality. Trends Food Sci. Technol., 62: 104–112. https://doi.org/10.1016/j.tifs.2017.01.010

Claeys WL, Verraes C, Cardoen S, Daube, G., De Block, J.,

Dewettinck, K., Dierick, K De Zutter, L., Huyghebaert, A., Imberechts, H., Thiange, P. and Vandenplas, Herman, L (2013). Raw or heated cow milk consumption: Review of risks and benefits. Food Contr., 31(1): 251–262. https://doi.org/10.1016/j.foodcont.2012.09.035

Da Cruz AG, Faria JAF, Saad SMI, Bolini HMA, Sant'Ana AS, Cristianini M (2010). High pressure processing and pulsed electric fields: Potential use in probiotic dairy foods processing. Trends Food Sci. Technol., 21(10): 483–493. https://doi.org/10.1016/j.tifs.2010.07.006

Dash S, Jaganmohan R (2022). Process optimization of plasma bubbling set up for cow milk decontamination and quality evaluation. bioRxiv. https://doi.org/10.1101/2022.12.28.522063

Dashab S, Jaganmohana R (2022). Impact of plasma bubbling on cow milk: Microbial reduction and improvement of milk quality. Int. J. Life Sci. Pharma Res., 12(1): L283–L288. https://doi.org/10.22376/ijpbs/lpr.2022.12.1.L283-288

de Mendiburu F (2023). Agricolae: Statistical procedures for agricultural research (R package version 1.3-7).

Ekezie FGC, Sun DW, Cheng JH (2017). A review on recent advances in cold plasma technology for the food industry. Trends Fd. Sci. Technol., 69: 46–58. https://doi.org/10.1016/j.tifs.2017.08.007

Gurol CEYDA, Ekinci FY, Aslan N, Korachi M (2012). Low temperature plasma for decontamination of E. coli in milk. Int. J. Food Microbiol., 157(1): 1–5. https://doi.org/10.1016/j.ijfoodmicro.2012.02.016

Hussain AA (2015). Electrical, thermal and optical characteristics of plasma torch. Iraqi J. Phys., 13(27): 151–156. https://doi.org/10.30723/ijp.v13i27.273

Jamal RK, Ali FH, Hameed MM, Aadim KA (2020). Designing a zener diode using $Ag_2O(1-x)ZnO(x)$/PSi structures deposited by laser-induced plasma technique. Iraqi J. Sci., 61: 1032–1039. https://doi.org/10.24996/ijs.2020.61.5.12

Jassim DR, Al-Ansari RA, Adil BH (2025). Synthesis and optical characterization of (Cu:Se) nanoparticles prepared via atmospheric pressure plasma jet for UV detector and gas sensor applications. Iraqi J. Phys., 23(3): 104–113. https://doi.org/10.30723/ijp.v23i3.1277

Joshi S, Mobeen A, Jan K, Bashir K, Azad ZAA (2019). Emerging technologies in dairy processing: Present status and future potential. In: Health and Safety Aspects of Food Processing Technologies. Elsevier. pp. 105–120. https://doi.org/10.1007/978-3-030-24903-8_6

Kadhem SJ (2021). Studying of plasma-polymerized pyrrole at variable gas flow rates via plasma jet. Iraqi J. Phys., 19(48): 44–51. https://doi.org/10.30723/ijp.v19i48.593

Kanca N, Avşar YK (2023). Cold plasma technology and its effects on some properties of milk and dairy products. Atatürk Üniv. Ziraat Fak. Derg., 54(2): 89–94. https://doi.org/10.5152/AUAF.2023.22028

Khalid NT, Shaymaa RK, Luma KH (2021). Effect of incorporated soft cheese with wheat germ extracts on quality and shelf life. Indian J. Ecol., 48: 244–248.

Khamsen N, Akkarachanchainon A, Fookiat K, Srisala J, Chomchuen S, Kanokbannakorn W, Srisonphan S (2016). Atmospheric cold plasma via fringe field enhanced corona discharge on single dielectric barrier for large-volume applications. Proc. Comp. Sci., 86: 321–324. https://doi.org/10.1016/j.procs.2016.05.087

Kim HJ, Yong HI, Park S, Kim K, Choe W, Jo C (2015). Microbial safety and quality attributes of milk following treatment with atmospheric pressure encapsulated dielectric barrier discharge plasma. Food Contr., 47: 451–456. https://doi.org/10.1016/j.foodcont.2014.07.053

Kitsiou ME (2023). Development of novel sustainable processing control strategies for monitoring bacterial pathogens (Doctoral dissertation). University of Surrey.

Langmuir I (1928). Oscillations in ionized gases. Proc. Natl. Acad. Sci., 14(8): 627–637. https://doi.org/10.1073/pnas.14.8.627

Laroussi M (2020). Cold plasma in medicine and healthcare: The new frontier in low temperature plasma applications. Front. Phys., 8: 74. https://doi.org/10.3389/fphy.2020.00074

Lee TA, Lin YH, Li PH, Ho JH (2024). The effects of corona discharge from a cold plasma source on the physicochemical properties and shelf-life of milk. Fd. Biosci., 62: 103980. https://doi.org/10.1016/j.fbio.2024.103980

Los A, Ziuzina D, Boehm D, Bourke P (2020). Effects of cold plasma on wheat grain microbiome and antimicrobial efficacy against challenge pathogens. Int. J. Fd. Microbiol., 335: 108889. https://doi.org/10.1016/j.ijfoodmicro.2020.108889

Mai TL, Sung SL, Shapovalova A, Janagama HK, Vuong L, Friedrich A, Samadpour M (2018). Performance validation of the Microbiologique Microfilm™ test system. J. AOAC Int., 101(5): 1508–1521. https://doi.org/10.5740/jaoacint.18-0036

Manoharan D, Stephen J, Radhakrishnan M (2023). Effect of atmospheric and low-pressure plasma on microbial reduction and milk quality. J. Food Saf., 43(1): e13018. https://doi.org/10.1111/jfs.13018

Misra NN, Koubaa M, Roohinejad S, Juliano P, Alpas H, Inácio RS, Barba FJ (2017). Landmarks in the historical development of twenty-first century food processing technologies. Fd. Res. Int., 97: 318–339. https://doi.org/10.1016/j.foodres.2017.05.001

Misra NN, Tiwari BK, Raghavarao KSMS, Cullen PJ (2011). Nonthermal plasma inactivation of food-borne pathogens. Fd. Eng. Rev., 3(3–4): 159–170. https://doi.org/10.1007/s12393-011-9041-9

Mohammed RS, Aadim KA, Ahmed KA (2022). Spectroscopy diagnostic of laser intensity effect on Zn plasma parameters generated by Nd:YAG laser. Iraqi J. Sci., 63: 3711–3718. https://doi.org/10.24996/ijs.2022.63.9.5

Muhsen RN, Hussein WS, Hussain WD, Aadim KA, Shaker AH (2025). Spectroscopic diagnostic of silicon plasma produced by DC and AC plasma jet techniques. Iraqi J. Appl. Phys., 21(2): 251–256.

Mumtaz S, Rana JN, Lim JS, Javed R, Choi EH, Han I (2023). Effect of plasma on-time on reactive species in plasma-treated medium. Int. J. Mol. Sci., 24(6): 5289. https://doi.org/10.3390/ijms24065289

Nahla TK, Makarim AM (2018). Effect of wheat germ on chemical, sensory and technological properties of soft cheese. Int. J. Dairy Sci., 13(1): 40–45. https://doi.org/10.3923/ijds.2018.40.45

Naz MY, Shukrullah S, Rehman SU, Khan Y, Al-Arainy AA, Meer R (2021). Optical characterization of non-thermal plasma jet energy carriers. Sci. Rep., 11: 2896. https://doi.org/10.1038/s41598-021-82019-4

Neokleous I, Tarapata J, Papademas P (2022). Non-thermal processing technologies for dairy products. Front. Sustain. Fd. Syst., 6: 856199. https://doi.org/10.3389/fsufs.2022.856199

Nicol MJ, Brubaker TR, Honish BJ, Simmons AN, Kazemi A, Geissel MA, Kirimanjeswara GS (2020). Antibacterial effects of low-temperature plasma. Sci. Rep., 10: 3066.

https://doi.org/10.1038/s41598-020-59652-6
Nikmaram N, Keener KM (2022). Effects of cold plasma technology on milk properties. LWT, 154: 112729. https://doi.org/10.1016/j.lwt.2021.112729
Odueke OB, Farag KW, Baines RN, Chadd SA (2016). Irradiation applications in dairy products. Fd. Bioproc. Technol., 9(5): 751–767. https://doi.org/10.1007/s11947-016-1709-y
Olschewski ES (2010). Aplicaciones de la física de plasmas en la industria. Ingeniería Ind., 28: 195–210.
Pankaj SK, Keener KM (2017). Cold plasma: Background, applications and current trends. Curr. Opin. Fd. Sci., 16: 49–52. https://doi.org/10.1016/j.cofs.2017.07.008
Park SY, Ha SD (2018). Assessment of cold oxygen plasma technology. J. Fd. Eng., 223: 42–45. https://doi.org/10.1016/j.jfoodeng.2017.11.041
Puligundla P, Mok C (2017). Potential applications of nonthermal plasmas against biofilms. J. Appl. Microbiol., 122(5): 1134–1148. https://doi.org/10.1111/jam.13404
Punia Bangar S, Suri S, Nayi P, Phimolsiripol Y (2022). Cold plasma for microbial safety. J. Fd. Proc. Preserv., 46(12): e16850. https://doi.org/10.1111/jfpp.16850
Rahnavard MA, Zare D, Nassiri SM, Taghvaei H, Fazaeli M (2024). Impact of gliding arc cold plasma on aflatoxin deactivation. Fd. Contr., 164: 110597. https://doi.org/10.1016/j.foodcont.2024.110597
Rathod NB, Ranveer RC, Bhagwat PK, Ozogul F, Benjakul S, Pillai S, Annapure US (2021). Cold plasma for preservation of aquatic foods. Comp. Rev. Fd. Sci. Fd. Saf., 20(5): 4407–4425. https://doi.org/10.1111/1541-4337.12815
Ribeiro RA, Bourbon-Melo N, Sá-Correia I (2022). Cell wall response to stress in yeasts. Front. Microbiol., 13: 953479. https://doi.org/10.3389/fmicb.2022.953479
Sayel MF, Khalid NT, Rashid KT (2023). Comparative study of nanofiltration and evaporation technologies on milk concentration. IOP Conf. Ser. Earth Environ. Sci., 1158: 112022. https://doi.org/10.1088/1755-1315/1158/11/112022
Sulaiman SM, Sulyman SAA (2021). Effect of non-thermal plasma jet on bacterial biofilms. Al-Mustansiriyah J. Sci., 32(5): 19–26. https://doi.org/10.23851/mjs.v32i5.1118
Tariq KN, Khairy HL, Khairi SR, Kamilah H (2023). Improved physiochemical and sensory properties of soft cheese. IOP Conf. Ser. Earth Environ. Sci., 1158: 112004. https://doi.org/10.1088/1755-1315/1158/11/112004
Thirumdas R, Kothakota A, Annapure US, Siliveru K, Blundell R, Gatt R, Valdramidis VP (2018). Plasma-activated water: Chemistry and applications. Trends Fd. Sci. Technol., 77: 21–31. https://doi.org/10.1016/j.tifs.2018.05.007
Wang S, Liu Y, Zhang Y, Lü X, Zhao L, Song Y, Ge W (2022). Processing sheep milk by cold plasma technology. LWT, 153: 112573. https://doi.org/10.1016/j.lwt.2021.112573
Wang W, Guo L, Yao Z, Xi W, Zhao Y, Lv J, Liu D (2022). Nitrox surface discharge used for water activation. J. Phys. D: Appl. Phys., 55(26): 265203. https://doi.org/10.1088/1361-6463/ac61b1
Xu H, Liu C, Huang Q (2023). Enhanced inactivation of fungi by cold plasma and PAW. Chem. Eng. J., 452: 139596. https://doi.org/10.1016/j.cej.2022.139596
Yahaya AG, Okuyama T, Kristof J, Blajan MG, Shimizu K (2021). Bactericidal effects of cold atmospheric-pressure microplasma. Molecules, 26(9): 2523. https://doi.org/10.3390/molecules26092523
Zou F, Yang M, Wu J, Wang L, Wang H (2025). Plasma-activated water in safe and sustainable food production. Crit. Rev. Fd. Sci. Nutr., 65: 1–25. https://doi.org/10.1080/10408398.2025.2477799