

## Effect of microwave pasteurization on bacterial quality of table eggs during refrigerator storage

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### ABSTRACT

Experiments were carried out at the College of Veterinary Medicine, University of Baghdad, during the period from October 26<sup>th</sup> 2023 to December 20<sup>th</sup> 2023, to study the effect of pasteurizing treatments of shell table egg using traditional Microwave oven on its quality characteristics during Zero, 1, 2, 4 and 8 weeks of refrigerator storage. A total of 120 fresh table eggs (White shell eggs) were collected from 20000 Luhman layer hens flock at Al-Amir project commercial farm, Al-Musaib city. These eggs were divided into 4 treatment of microwave pasteurization treatments which were Zero, 10, 20, and 30 sec. Results revealed that the numbers of total bacteria and total coliform on the surface of table egg shells is affected by pasteurization with a microwave immediately after treatment (Zero time), the lowest number was for treatment with pasteurization for 30 seconds, and the number of bacteria was in pasteurization treatments were significantly lower compared to the control treatment. As the storage period progressed, the number of total bacteria on the surface of the shell continued to decrease significantly from the control treatment. This was also the case after storage for 2, 4, and 8 weeks, the two pasteurization treatments for 20 seconds and 30 seconds recorded the lowest values. In conclusion, microwave pasteurization of table egg is very imported to extending shelf life of table eggs during refrigerator storage.

**Keywords:** Table eggs, Pasteurization, Microwave, Bacterial count, Storage.

### Introduction

Eggs are considered highly perishable food, just like milk and meat. Therefore, they require storage and preservation conditions to protect them from deterioration and spoilage, especially microbial spoilage. Contamination of eggs with microorganisms begins as soon as they leave the hen's body (oviposition), and microorganisms quickly begin to increase on the surface of the egg shell as the storage period increases, especially in cases of high temperatures and high humidity, in addition to the deterioration of the internal qualitative characteristics of the egg as a result of the release of CO<sub>2</sub> gas from the inside of the egg (Al-Obaidi and Al-Shadeedi, 2022). Fresh eggs contain three layers of protection, which are the outer cuticle layer, or called

the outer waxy layer, then the calcareous shell, and then comes the inside membrane of the shell, which is an outer and inner membrane. Each of these layers has an important and effective role in protecting the egg and limiting or hindering the entry of microorganisms (Morsy et al., 2015; Al-Shadeedi, 2023; Salman et al., 2023).

There is no ideal method for storing eggs that prevents them from being damaged or spoiled, but eggs are stored to reduce the deterioration in their quality and prolong their storage period. Eggs are transported from production fields to refrigerated wholesale warehouses before they reach the consumer using cardboard trays with a capacity of 30 eggs, placed inside cardboard boxes of one capacity. A total of 12 layers of cardboard and in this form, it is

marketed to single-stores. Eggs prepared for storage must be clean and free of external defects, as these eggs are more resistant to microbial contamination and storage conditions (Dev, et al., 2008; Shenga et al., 2010; Al-Shadeedi, 2018; Oliviera et al., 2022). Numerous studies have been conducted over a long period of time to determine the ideal conditions for storing eggs in order to preserve their internal quality and increase their shelf life for consumption (Sliversides and Scott, 2001; Al-Shadeedi, 2009; Al-Shadeedi, 2010).

Microwave technology has been widely studied as an alternative to food pasteurization and sterilization processes. This technology is based on heating the volume produced by microwave radiation using the insulating properties of foods. The dielectric properties of eggs, such as dielectric constant and loss factor, are determined to explain how they interact with microwave energy (Dev et al., 2008).

Microwave techniques are used to inactivate microorganisms in food products by generating heat internally. Research shows that these techniques allow faster access to cold spots in the product, reducing processing time and causing less damage to product quality. The range of microwave technologies includes microwaves, radiofrequency and ohmic heating (Bermudez-Aguirre and Niemira, 2022).

These techniques show effectiveness in inactivating microorganisms, but a careful understanding of the dielectric properties and heating effects of eggs is required. Research indicates that the effects of

microwave radiation range from changes in gene expression to effects on protein and cell proliferation. Microwave irradiation is considered a promising option to improve egg pasteurization processes efficiently and at a lower cost, especially considering the depth of microwave penetration and its impact on microbial integrity in sensitive products (Dev et al., 2008; Bermudez-Aguirre and Niemira, 2022).

The aim of the current research is to study the effect of treating fresh table eggs for different periods of time in a microwave oven (0, 10, 20 and 30 sec.) as a means of pasteurization and its effect on their bacterial count characteristics after storing in the refrigerator for (0, 1, 2, 4, and 8) weeks.

### Materials and Methods

**Period of the study:** Experiments were carried out at the College of Veterinary Medicine, University of Baghdad, during the period from October 26<sup>th</sup> 2023 to December 20<sup>th</sup> 2023, to compare between three period of pasteurizing treatments of shell table egg which were 10, 20, and 30 second using traditional Microwave oven on its quality characteristics, egg components percentage and microbial counts during Zero, 1, 2, 4 and 8 weeks of refrigerator storage.

**Egg collection:** A total of 120 fresh table eggs (White shell eggs) were collected from 20000 Luhman layer hens flock at Al-Amir project commercial farm, Al-Musaib city, the layers were fed a production ration as explain in Table (1).

**Table (1): Components and ingredients quantity of ration.**

Components	Ingredients quantity in 2 tons
Corn	650
Soya Meal	500
Wheat	200
Wheat bran	100
Rice bit	200
Chickpea	100
Premix	50
Vegetable oil	25
Limestone	125
Total	2000 Kg

**Treatments:** the experiment was to compare between three period of pasteurizing treatments of shell table egg which were microwave exposed for 10, 20, and 30 second using traditional Microwave oven on its quality characteristics, egg components percentage and microbial counts during Zero, 1, 2, 4 and 8 weeks of refrigerator storage. After egg

collection 120 eggs were immediately distributed into four groups of treatments:

T1: 30 eggs were untreated eggs as control group.

T2: 30 eggs were microwave exposed for 10 sec.

T3: 30 eggs were microwave exposed for 20 sec.

T4: 30 eggs were microwave exposed for 30 sec.

All eggs were refrigerator storage immediately after treatment.

**Storage periods:** All washed eggs were refrigerator storage for 1, 2, 4 and 8 weeks. At each period 5 eggs from each treatment were randomly collected and individually weight and interior quality parameter were studied.

**Microwave oven:** A commercial microwave oven was used, one of the types of ovens used for home purposes, type (SAMSUNG), Chinese origin.

**Bacterial counts:** All bacterial count tests were conducted in the laboratory of the Milk Hygiene at the College of Veterinary Medicine, University of Baghdad, and were done on three replicates of eggs for each treatment and after each storage period. Two eggs were placed in nylon bags containing 100 ml of sterile peptone water (Accumix company) at sterile conditions and rinsed with accurate for 2 min. then 1ml of the solution was transferred to screw capped bottles containing 9 ml of sterile peptone water to make decimal dilution of 10 ml each. The culture media used in bacteriological examinations were sterilized at 121 °C for 15 minutes and under a pressure of 15 pounds / square inch. As for the different glassware, they were sterilized in an electric oven at 180°C for three hours.

**Total bacterial count:** Total bacteria were counted using the pour-plate method mentioned by Harrigan and McCance (1976), by transferring (1 ml) of each decimal diluent with a sterile pipette into two empty and sterilized Petri dishes (Duplicate) and directly added to each dish. (15 ml) of the sterile culture medium (Nutrient Agar, Accumix company) kept in a water bath at (46 °C), then the culture medium is mixed with the dilution of the bacterial suspension well by rotating the dishes towards the right and towards the left with stirring it each time and after the culture medium hardened, the dishes were preserved inverted in the incubator at (37°C) for 24 hours, after which the growing colonies were counted in dishes containing (30-300) colonies, and the number of bacteria was estimated by multiplying the number of colonies by the reciprocal of dilution to extract the number in one milliliter of one egg.

**Coliform bacterial count:** coliform bacteria were counted using the pour-plate method mentioned by Harrigan and McCance (1976) to estimate the numbers of coliform bacteria by transferring (1 ml) of each decimal diluent with a sterile pipette into two empty and sterilized Petri dishes (Duplicate) and directly (15 ml) of sterile culture medium (MacConkey Agar, Accumix company) kept in a water bath at (46°C) is added to each plate, then the culture medium is mixed with the dilution of the bacterial suspension well by rotating the plates towards the

right and towards the left with stirring each time and after the medium hardens. The culture dishes were kept upside down in the incubator at (37°C) for 24 hours after which the growing colonies were counted in the dishes containing (30-300) colonies, and the numbers of bacteria were estimated by multiplying the number of colonies by the reciprocal of dilution to extract the number in one milliliter of one egg.

**Statistical analyses:** Data were analyzed by using the General Linear Model Procedure of SAS (2001). Means were compared by the Duncan's Multiple Range test at 5% probability (Steel and Torrie, 1980).

## Results and Discussion

Table (2) Shows that the numbers of total bacteria on the surface of table egg shells is affected by pasteurization with a microwave immediately after treatment (Zero time), and it ranged between ( $9 \times 10^2$  to  $68 \times 10^2$ ), where the lowest number was for treatment with pasteurization for 30 seconds, and the number of bacteria was in Pasteurization treatments were significantly lower compared to the control treatment. As the storage period progressed, the number of total bacteria on the surface of the shell continued to decrease significantly from the control treatment. This was also the case after storage for two and four weeks, and when storage reached eight weeks, the two pasteurization treatments for 20 seconds and 30 seconds recorded the lowest values, reaching ( $124 \times 10^3$  and  $62 \times 10^3$ ) Respectively, with a significant difference from the 10 second pasteurization treatment, which recorded total bacteria numbers of ( $89 \times 10^4$ ). This, in turn, was significantly lower than the control treatment, which recorded the highest total bacterial  $161 \times 10^5$  with a significant difference from the rest of the treatments. Table (3) Shows that the number of total coliform bacteria on the surface of table egg shells is affected by microwave pasteurization immediately after treatment (Zero time) Between ( $5 \times 10^2$  to  $26 \times 10^2$ ). The lowest number of treatments was in pasteurization for 30 seconds, and the number of bacteria in the pasteurization treatments was significantly lower compared to the control treatment. As the storage period progressed, the numbers of coliform bacteria on the surface of the peel continued to decrease significantly from the control treatment. This was also the case after storage for two and four weeks. When storage reached eight weeks, the two pasteurization treatments for 20 and 30 seconds recorded the lowest values, reaching ( $240 \times 10^2$  and  $201 \times 10^2$ ) Respectively, with a significant difference from the 10 second pasteurization treatment, which recorded the

number of coliform bacteria at  $87 \times 10^3$ . This, in turn, was significantly lower than the control treatment, which recorded the number of coliform bacteria and

their value  $162 \times 10^4$ . With a significant difference from the rest of the treatments.

**Table (2): Effect of microwave treatments on table egg total bacterial count after refrigerator storage (cfu/egg).**

Microwave Treatments	Zero time	1 week	2 weeks	4 weeks	8 weeks
Control	$68 \times 10^2$ $\pm 21.24$ a	$108 \times 10^3$ $\pm 44.07$ a	$235 \times 10^3$ $\pm 36.15$ a	$177 \times 10^4$ $\pm 41.65$ a	$161 \times 10^5$ $\pm 33.87$ a
10 Sec.	$27 \times 10^2$ $\pm 22.16$ b	$133 \times 10^2$ $\pm 38.19$ b	$98 \times 10^3$ $\pm 38.74$ b	$186 \times 10^3$ $\pm 43.28$ b	$89 \times 10^4$ $\pm 36.16$ b
20 Sec.	$23 \times 10^2$ $\pm 19.20$ b	$51 \times 10^2$ $\pm 43.61$ b	$117 \times 10^2$ $\pm 42.57$ c	$205 \times 10^2$ $\pm 37.29$ c	$124 \times 10^3$ $\pm 34.95$ c
30 Sec.	$9 \times 10^2$ $\pm 18.35$ b	$32 \times 10^2$ $\pm 39.75$ b	$97 \times 10^2$ $\pm 37.29$ c	$180 \times 10^2$ $\pm 38.15$ c	$62 \times 10^3$ $\pm 40.43$ c
Significant	*	*	*	*	*

**Table (3): Effect of microwave treatments on table egg total coliform count after refrigerator storage (cfu/egg).**

Microwave Treatments	Zero time	1 week	2 weeks	4 weeks	8 weeks
Control	$26 \times 10^2$ $\pm 9.65$ a	$104 \times 10^2$ $\pm 9.53$ a	$55 \times 10^3$ $\pm 8.78$ a	$122 \times 10^3$ $\pm 10.38$ a	$162 \times 10^4$ $\pm 10.32$ a
10 Sec.	$14 \times 10^2$ $\pm 8.45$ b	$65 \times 10^2$ $\pm 9.72$ b	$134 \times 10^2$ $\pm 9.25$ b	$271 \times 10^2$ $\pm 9.66$ b	$87 \times 10^3$ $\pm 9.58$ b
20 Sec.	$11 \times 10^2$ $\pm 9.77$ b	$33 \times 10^2$ $\pm 8.99$ b	$79 \times 10^2$ $\pm 8.66$ b	$164 \times 10^2$ $\pm 10.17$ c	$240 \times 10^2$ $\pm 10.64$ c
30 Sec.	$05 \times 10^2$ $\pm 9.16$ b	$24 \times 10^2$ $\pm 9.36$ b	$51 \times 10^2$ $\pm 8.72$ b	$9 \times 10^2$ $\pm 9.51$ c	$201 \times 10^2$ $\pm 9.74$ c
Significant	*	*	*	*	*

Eggs are perishable food material that requires preservation and cold storage once they are produced from the fields in order to preserve the qualitative characteristics for the longest possible period during storage and to deliver them to the consumer with the best quality and least deterioration (Stadelman and Cotterill, 1995; Al-Shadeedi, 2010; Al-Obaidi and Al-Shadeedi, 2022). The problems of microorganisms on the surface of the egg shell are represented in the penetration of these microorganisms into the egg period and its membranes through the open pores in the egg shell and access to the internal contents of the egg, the first of which is the egg white, and thus the rotting and spoilage of the eggs (North, 1984; Al-Obaidi et al., 2010ab).

The results obtained from this experiment clearly indicated, as shown in Tables (13, 14), that the numbers of total bacteria and coliform bacteria were significantly reduced on the surface of the microwave-pasteurized egg shell, with a significant

difference from the control treatment, and this treatment recorded the highest numbers of total bacteria and coliform bacteria in the same time. At the same time, we find that the percentage of decrease in the number of total bacteria and coliform bacteria increases with the increase in the duration of exposure to microwave pasteurization. This indicates that microwave treatment has a positive and important role in reducing the number of bacteria on the surface of the peel, or what is called reducing the initial microbial load, and this in turn contributed to reducing these groups of bacteria on the surface of egg shells in microwave pasteurization treatments after storage in the refrigerator continue to have a significant role in the bacterial population in the control treatment over the microwave pasteurization treatments. These results are consistent with the results of previous research (Zhang et al., 2013; Stadelman et al., 1996; Stanley and Petersen, 2017; Keener, 2017; Skowron et al., 2022) because pasteurization in particular and thermal treatments in

general play a major role. It is effective in reducing the number of bacteria on the surface of eggs, as most of the total bacteria, coliform bacteria, and microorganisms contaminating eggs are mesophilic bacteria that are killed by the heat of pasteurization. At the same time, these bacteria do not grow well or reproduce at low temperatures. Thus, storing in the refrigerator contributed to reducing their numbers. The microwave is an effective way to pasteurize eggs and preserve them from deterioration during storage and prolong their suitability for consumption, as research indicates (Stadelman and Cotterill, 1995; Sivaramakrishnan, 2010; Al-Shadeedi, 2010; Al-Obaidi and Al-Shadeedi, 2022).

Studies indicate that table eggs are completely free of microorganisms the moment they leave the hen's body, but they soon begin to become contaminated through contact with the outer surface of the egg with the external components. The degree of contamination of the egg shell surface depends on several external factors related to field management or egg production halls, including the degree of cleanliness of the nests (North, 1984).

The effectiveness of microwave pasteurization depends on the initial count of microorganisms. When the initial loads are low, microwave pasteurization can be effective at the temperature of 65°C to 90°C for a few minutes (Stanley and Peterson, 2017). A commercial patent in South Africa was submitted to sell pasteurized eggs using microwave radiation. The process takes 30 to 40 min, reducing 2 to 5 log reductions of microorganisms with some changes in the albumen. However, microwave penetration depth ( $\approx 0.3$  cm) can compromise microbial safety in products like eggs, mainly the yolk (Geveke et al., 2017; Yang et al, 2020).

### Conclusions

We conclude from the current research that the using microwave pasteurization for 10, 20, and 30 seconds contributed to reducing the numbers of total bacteria and coliform bacteria on the surface of table egg shells. contributed to maintaining good quality of table eggs when stored for 8 weeks and the best process for pasteurization in the microwave is 30 sec., compared to 10 sec. Microwave pasteurization procedures gave the best internal quality characteristics of table eggs when stored in the refrigerator for a period of up to 8 weeks.

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