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Effects of Different Viol Types on Egg Shell Microbiology in Table Eggs at Different Storage Temperatures

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HIGHLIGHTS

- Plastic box viols were found to have lower levels of TAMP and mold-yeast than cardboard viols.
- Regarding coliform, there was no significant difference between cardboard viols, cardboard box viols and plastic box viols.
- Higher total bacteria and mold-yeast loads were detected in eggs stored at 4°C than those stored at 25°C.
- Regarding coliform, there was no significant difference between 4°C and 25°C.

Abstract

The purpose of this research was to examine the impact of storing table hen eggs in egg cardboard box viol, plastic box viol, and cardboard viols at various temperatures on the microbiology of the egg shell. A total of 150 table chicken eggs were used in the study. The eggs were randomly distributed into three groups: standard cardboard viol, plastic box viol, and cardboard box viol. The eggs were stored at room temperature and in the refrigerator for 28 days. On the 0, 7, 14, 21, and 28 days of the experiment, five eggs from each group were analyzed for total bacteria (TAMB), mold-yeast, and coliform. The Viol type x storage temperature interaction effect was significant ($P<0.05$) only on egg shell mold-yeast, but its effect on the TAMB and coliforms was insignificant. The effect of viol types on shell coliforms was insignificant. Egg TAMB and mold-yeast counts were found to be lower in plastic box viols than in other groups ($P <0.05$). TAMB and mold-yeast count were found to be higher in eggs that were stored in refrigerator conditions than in room conditions ($P <0.05$). From a microbial perspective, it can be said that storing eggs in plastic box viols is more suitable in terms of hygiene during storage.

Keywords: Viol; Eggshell, Total Aerobic Mesophilic Bacteria, Mold-Yeast, Coliform, Storage

1. Introduction

Although eggs are an animal protein source, their importance in human nutrition is high due to their containing unsaturated fatty acids, vitamins, and minerals (Şenköylü 2001; Sarıca and Erensayın 2014; Puglisi and Fernandez 2022; Tian et al. 2022). An egg obtained from a healthy animal has the highest quality value at the moment it is laid, but its quality may decrease depending on storage conditions, leading to marketing problems (Aygun 2017; Aygün and Narinç 2017; Brasil et al. 2019; Yenilmez and Bulancak 2020; Sariyel et al. 2022). There are between 300 and 500 bacteria on the shell when it is laid (North and Bell 1990). Within an hour

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of the egg being laid, this number may quickly reach 20,000 or 30,000 bacteria (North and Bell 1990). In the market, cardboard viols with a capacity of 30 eggs are used for storing or transporting eggs. However, in recent years, cardboard box viol and plastic box viol with a capacity of 6, 10, and 15 eggs have also started to be used. The use of cardboard box viol with a capacity of 6, 10, and 15 eggs during the sale of eggs makes it difficult for consumers to choose eggs based on their outer appearance, as the dirt on the surface of the eggshell cannot be seen. On the other hand, plastic box viols provide convenience for consumers in terms of choice. However, foreign materials on the surface of eggs in plastic box viol can be seen more easily, making it easier to evaluate the eggs based on their appearance. Additionally, during storage or sales, there is a possibility of condensation on the egg surface or inside the viols, which can lead to the eggs becoming wet and causing the preference for viols to become important. The number of bacteria present on an eggshell can vary depending on a number of factors, such as the hygiene practices used during egg handling, storage conditions, and the presence of any pathogens in the environment.

Uysal et al. (2002) did not find a significant difference in bacterial and fungal load between plastic box and cardboard egg viols for hatching eggs. Jones et al. (2004) studied the concentrations of bacteria, yeast, and mold on eggshells during a 10-week storage period at 4 °C. They found that the concentration of aerobic bacteria on the eggshell increased from 4.0 log CFU/ml to 5.3 log CFU/ml after 8 weeks of storage. As the storage period continued, the concentration of yeast and mold increased, reaching the highest levels of 2.9 and 2.6 log CFU/ml at 8 and 10 weeks, respectively. According to Huneau-Salaün et al. (2010), contamination increased with hen age, airborne dust concentration, manual packing of the eggs, and plastic packing as opposed to recycled-pulp egg-flats increased.

In our literature review, it was observed that there are a limited number of studies on the effect of storing table chicken eggs in cardboard box viols or plastic box viols (with a capacity of 6, 10, and 15 eggs) on egg quality. Therefore, the main question of the research is which tray type can preserve egg quality for a longer period of time during storage under different storage conditions. Therefore, the aim of this study is to investigate the changes in egg quality by storing table chicken eggs in cartons, cardboard box viol, and plastic box viols under room and refrigerator conditions.

2. Materials and Methods

Table eggs that were daily purchased from the Konya commercial egg producer were used in the study. A total of 150 table eggs were used. The research was carried out in the Egg Quality Laboratory of the Department of Animal Science, Faculty of Agriculture, Selcuk University.

Eggs were randomly distributed into 3 groups: cardboard viol, plastic box viol, and cardboard box viol. Eggs were stored in viols at room temperature (25±2°C) and in the refrigerator (4±2°C) for 28 days. On the 0, 7, 14, 21, and 28 days of the experiment, total bacteria, mold-yeast, and coliform analyses were performed in the egg shells of 5 eggs from each group.

For microbiological analysis, each egg sample was transferred into sterile pouches containing 50 ml of peptone water (0.1%) (Merck, Germany), and they were kept in these solutions for 2-3 minutes. After that, serial decimal dilutions were prepared, and 1 mL of appropriate dilutions were spread on agar plates in duplicate. The count of total aerobic mesophilic bacteria (TAMB) was determined on Plate Count Agar (PCA; Merck) incubated at 37°C for 48 h while mold-yeast counts on Potato Dextrose Agar acidified by sterile tartaric acid (10 %) (Merck) were incubated at 25°C for 5 days. Coliform bacteria were cultured on Violet Red Bile Agar (VRBA; Merck) anaerobic incubated at 37°C for 24 h (Aygun and Sert, 2013). The results are given as log CFU/g.

The experiment was carried out in 2x3 randomized plots according to a factorial design in order to compare the shell microbial load of eggs packed in plastic box viol, cardboard box viol, and cardboard viols to be stored at room temperature and in the refrigerator (Duzgunes et al. 1987). The statistical analyses were conducted

using the One-Way Analysis of Variance (ANOVA). The MINITAB 16 package program was used in the analysis, and the Tukey multiple comparison test was used to determine the different groups.

3. Results

3.1. Total mesophilic aerobic microorganisms

Table 1 summarizes the effects of storage temperature, viol types, and the storage temperature x viol type interaction on total mesophilic aerobic microorganisms. In all periods, the effect of storage temperature x viol type interaction on total mesophilic aerobic bacteria was not significant. The effect of viol types on eggshell TAMB was significant in all weeks except the second week ($P<0.05$). On the 28th day of storage, group C had the highest TAMB value (4.47 log cfu/g egg) ($P<0.05$), and the difference in TAMB value between the CB (3.98 log cfu/egg) and Pl (3.99 log cfu/g egg) groups was statistically insignificant. The effect of storage temperature on eggshell TAMB was significant in all weeks except the first week ($P<0.05$). The shell TAMB levels of eggs stored at 4 °C (4.32 log cfu/g egg) were found to be higher than those stored at 25 °C (3.97 log cfu/g egg) after 28 days of storage.

Table 1. The effect of storage temperature, viol types and storage x viol type interaction on Total aerobic mesophilic bacteria (Log cfu/g egg)

Treatment		Fresh eggshell TAMB	Total aerobic mesophilic bacteria			
			7 days	14 days	21days	28 days
Storage Temperature (°C)	25	4.19	3.80	3.78	3.94	3.97
	4	4.19	3.97	4.17	4.25	4.32
	SEM	0.032	0.074	0.095	0.069	0.083
	P-value	0.977	>0.05	<0.05	<0.05	<0.05
Viol type[1]	C	4.23	3.51^{b}	3.86	4.29^{a}	4.47^{a}
	CB	4.19	4.16^{a}	4.04	4.05^{ab}	3.98^{b}
	PL	4.16	3.99^{a}	4.02	3.95^{b}	3.99^{b}
	SEM	0.039	0.090	0.116	0.084	0.103
	P-value	0.541	<0.05	>0.05	<0.05	<0.05
Storage Temperature (°C) x Viol Type	25 x C	4.20	3.52	3.59	4.15	4.23
	25 x CB	4.19	4.09	3.85	3.87	3.82
	25 x PL	4.19	3.79	3.89	3.81	3.87
	4 x C	4.25	3.49	4.12	4.43	4.71
	4 x CB	4.19	4.23	4.22	4.24	4.13
	4 x PL	4.13	4.19	4.15	4.09	4.11
	SEM	0.055	0.127	0.164	0.119	0.144
	P-value	0.624	>0.05	>0.05	>0.05	>0.05

$^{a-b}$Significant differences exist between the means of a column using different superscripts ($P<0.05$); [1]C: Cardboard viol; CB: Crbboard box viol; PL: Plastic box viol; SEM: Standard Error Mean

3.2. Mold-yeast

At the beginning of this study and after 7 days of storage, no mold-yeast was found in the eggshell (Table 2). Except for 14 days of storage, the effect of the storage temperature x viol type interaction on mold-yeast was significant at 21 and 28 days of storage ($P<0.05$). On the 28th day of storage, the highest mold-yeast value was found in eggs stored at 4 °C in cardboard viols, and the lowest mold-yeast value was found in eggs stored at 25 °C in plastic box viols ($P<0.05$). On the other hand, when the viol types were investigated, the highest amount of mold was found in the C group and the lowest amount of mold was found in the PL group at the

end of storage (P<0.05). On the 28th day of storage, eggs stored in the refrigerator had a higher mold value than those stored at room temperature (P<0.05).

Table 2. The effect of storage temperature, viol types and storage x viol type interaction on egg mold-yeast (Log cfu/g egg)

Treatment		Fresh eggshell mold-yeast	Mold-yeast			
			7 days	14 days	21days	28 days
Storage Temperature (°C)	25	0.000	0.000	0.501	1.821	2.117
	4	0.000	0.000	0.747	3.001	3.473
	SEM			0.308	0.089	0.086
	P-value			>0.05	<0.05	<0.05
Viol type[1]	C	0.000	0.000	0.245	3.280[a]	3.882[a]
	CB	0.000	0.000	0.784	2.434[b]	2.850[b]
	PL	0.000	0.000	0.844	1.519[c]	1.653[c]
	SEM			0.377	0.110	0.106
	P-value			>0.05	<0.05	<0.05
Storage Temperature (°C) x Viol Type	25 x C	0.000	0.000	0.490	3.113[ab]	3.790[ab]
	25 x CB	0.000	0.000	0.494	2.350[b]	2.560[c]
	25 x PL	0.000	0.000	0.520	0.000[c]	0.000[d]
	4 x C	0.000	0.000	0.000	3.448[a]	3.973[a]
	4 x CB	0.000	0.000	1.074	2.518[b]	3.140[bc]
	4 x PL	0.000	0.000	1.168	3.038[ab]	3.307[ab]
	SEM			0.533	0.154	0.149
	P-vaalue			>0.05	<0.05	<0.05

[a-d]Significant differences exist between the means of a column using different superscripts (P<0.05); [1]C: Carrdboard viol; CB: Cardboard box viol; PL: Plastic box viol; SEM: Standard Error Mean

3.3. Coliform

Table 3 summarizes the effects of storage temperature, viol types, and the storage temperature x viol type interaction on coliform load. In all periods, the effect of storage temperature, viol types and storage temperature x viol type interaction on coliform load was not significant.

4. Discussion

Eggs stored at four degrees have been found to have higher TAMP and mold-yeast levels in their shells compared to eggs stored at room temperature (P<0.05). This result is consistent with the studies of Board and Tranter (1995), which indicate that bacteria penetrate the shells of eggs stored at high temperatures (37 °C) less than those stored at lower temperatures (20 °C). It can be said that the number of microorganisms that pass from the shell to the inside of the egg is higher in eggs stored at high temperatures. The result that the storage temperature has no effect on the coliform load during storage contradicts the study of Aygun and Sert (2013), which found that eggs stored in refrigerator conditions contained less coliform than those stored at room temperature.

Eggs stored in plastic and covered cardboard cartons have been found to have a lower TAMP and mold-yeast load on their shells compared to eggs stored in standard cardboard cartons. This may be due to the easy contamination of the environment with microorganisms. It has been observed that storing eggs in closed packages limit the possibility of contamination by microorganisms. In other words, packaging and storing eggs limit the possibility of contamination by microorganisms. The result of our study, which found that eggs stored in plastic cartons have a higher TAMP load than eggs stored in standard cardboard cartons, contradicts the Huneau-Salaün et al. (2010) study, which found that the bacterial load in eggs stored in plastic cartons was

higher than that of eggs stored in cardboard cartons. The reason is that the cartons used in our study were a new brand. According to Huneau-Salaün et al. (2010), recycled pulp viols were destroyed after use, but plastic box egg viols could be reused by farms.

Table 3. The effect of storage temperature, viol types and storage x viol type interaction on coliform (Log cfu/g egg)

Treatment		Fresh eggshell coliform	Coliform			
			7 days	14 days	21days	28 days
Storage Temperature (°C)	25	0.000	0.298	0.133	0.000	0.000
	4	0.000	0.313	0.609	0.353	0.692
	SEM		0.205	0.226	0.176	0.270
	P-value		>0.05	>0.05	>0.05	>0.05
Viol type[1]	C	0.000	0.200	0.284	0.260	0.000
	CB	0.000	0.269	0.630	0.269	0.420
	PL	0.000	0.447	0.200	0.000	0.618
	SEM		0.251	0.277	0.216	0.331
	P-value		>0.05	>0.05	>0.05	>0.05
Storage Temperature (°C) x Viol Type	25 x C	0.000	0.000	0.000	0.000	0.000
	25 x CB	0.000	0.000	0.400	0.000	0.000
	25 x PL	0.000	0.894	0.000	0.000	0.000
	4 x C	0.000	0.400	0.568	0.520	0.000
	4 x CB	0.000	0.538	0.860	0.538	0.840
	4 x PL	0.000	0.000	0.400	0.000	1.236
	SEM		0.355	0.392	0.306	0.468
	P-value		>0.05	>0.05	>0.05	>0.05

[1]C: Cardboard viol; CB: Cardboard box viol; PL: Plastic box viol; SEM: Standard Error Mean

In our study, it was concluded that storage temperature and viol types did not have a significant effect on eggshell coliform load during storage. This is consistent with Uysal et al. (2002), who found that different viol types had a negligible effect on shell coliform load during storage of hatching eggs.

5. Conclusions

Our study revealed that different types of viols used in the storage of table eggs have a significant effect on eggshell total bacteria and mold-yeast load. It was observed that storing eggs in plastic box viols resulted in lower eggshell total bacteria and mold-yeast load compared to storing them in cardboard viols. Different types of viols did not have a significant effect on eggshell coliform load. It was determined that the total bacteria and mold-yeast load in eggs stored at 4 °C were higher than those stored at 25 °C. From a microbial perspective, it can be said that storing eggs in plastic box viols is more suitable in terms of hygiene during storage.

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