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Abstract

This study investigated how storage temperature and egg washing influence the internal and external quality of eggs during a 42-day storage period. The study aimed to determine whether modest temperature differences within the 15–20 ℃ range, typical of distribution environments, accelerate freshness loss and whether sodium hypochlorite washing alters egg quality decline. A total of 1,560 freshly laid eggs from 62-week-old Hy-Line Brown hens were assigned to a 2 × 3 factorial arrangement comprising two washing treatments (washed vs. unwashed) and three storage temperatures (15, 18, and 20 ℃). Eggs were stored for 42 days, and internal and external quality traits were evaluated. Data were analyzed using two-way ANOVA to determine main and interaction effects. Orthogonal polynomial contrasts were used to evaluate the significance of linear or quadratic effects of the increasing storage temperature. Eggs held at 20 ℃ consistently showed greater weight loss, lower specific gravity, enlarged air cells, elevated albumen pH, and marked reductions in albumen height and Haugh unit compared with those stored at 15 ℃ ($p < 0.05$). Yolk index values declined more rapidly at higher temperatures. Washed eggs stored at 20 ℃ showed lower albumen height and Haugh unit on day 3, and higher albumen pH early in storage ($p < 0.05$). However, washing did not influence long-term deterioration. Across treatments, albumen pH increased progressively, paralleled by thinning of the thick albumen and a reduction in Haugh unit, whereas yolk index declined as storage progressed. Within the 15–20 ℃ interval, storage temperature was more consistently associated with the rate and extent of egg quality deterioration than sodium hypochlorite washing, whereas washing related effects were mainly observed during the early storage period. Maintaining storage temperature at or below 18 ℃ may help preserve albumen structure, yolk integrity, and freshness during extended storage.

Keywords: Egg quality, Storage temperature, Unwashed egg, Washed egg, Yolk index

Introduction

Eggs are a cost-effective and nutrient-dense food that contributes significantly to the dietary quality of populations at risk of inadequate nutrient intake, including children, pregnant women, and older individuals [1,2]. They provide an excellent source of high-quality protein and substantial amounts of unsaturated fatty acids, vitamins, and minerals that are essential for growth, tissue maintenance, and overall health [3,4]. However, due to their high moisture and protein content, eggs are highly perishable, and their physical and chemical properties deteriorate rapidly during storage [5,6], leading to considerable losses in quality and economic value [7]. Therefore, maintaining optimal storage conditions is crucial to preserving the nutritional and sensory quality of eggs.

Egg freshness, which determines consumer acceptability, is evaluated using various physical and chemical indicators such as shell color, weight, thickness, albumen height, yolk color, albumen pH, and viscosity. These parameters are influenced by multiple genetic and environmental factors, including strain, hen age, storage temperature, humidity, and storage duration [8]. Among these factors, storage temperature is considered the most critical determinant of egg quality deterioration [9,10]. Higher temperatures accelerate albumen thinning, reduce Haugh unit (HU), elevate albumen and yolk pH, and increase weight loss [9,11]. In particular, elevated storage temperatures accelerate the loss of freshness and exacerbate quality changes during subsequent washing and handling processes. Therefore, understanding how external factors such as washing interact with temperature to influence egg quality is essential for effective postharvest management.

The eggshell cuticle acts as a natural barrier that limits microbial penetration [12]. However, contamination may occur during collection and handling, which has led to the widespread use of washing and sanitizing treatments in commercial practice. Although washing with sodium hypochlorite is commonly applied to improve shell cleanliness [13], it may partially remove or alter the cuticle, a natural antimicrobial and physical barrier, thereby increasing shell permeability and potentially accelerating internal quality deterioration during storage [14,15]. Thus, understanding how washing affects the quality of eggs stored under practical distribution temperatures is essential for improving

postharvest handling and freshness preservation.

Prolonged storage increases albumen pH, decreases HU and yolk index (YI), and intensifies weight loss [16,17], with these changes being strongly affected by storage temperature. However, the interactive effects of washing and storage temperature on egg quality remain poorly understood. Therefore, this study aimed to investigate the effects of storage temperature and egg washing on egg quality using multiple internal and external quality parameters over the entire storage period. It was hypothesized that higher temperatures and washing treatments would accelerate the loss of internal freshness and shell integrity, particularly under warm storage conditions. The findings provide a scientific basis for improving temperature management and handling practices to ensure both food safety and quality preservation within the egg supply chain.

Materials and Methods

Egg preparation and experimental design

A total of 1,560 freshly laid eggs were obtained from 62-week-old Hy-Line Brown laying hens and used for quality evaluation. The eggs were divided into two groups according to washing treatment: washed and unwashed eggs. For the washing treatment, eggs were washed using water maintained at 40–50 ℃, and the sodium hypochlorite solution was adjusted to a sanitizer concentration of 10–50 ppm. For the unwashed treatment, physical brushing was applied without exposure to wash water or sodium hypochlorite solution. All eggs were stored under three temperature conditions (15, 18, 20 ℃) in a convection oven (VS-1202D4N, Vision; Bionex, Bucheon, Korea). The relative humidity was maintained at 40–50%. The experiment was conducted following a 2 × 3 factorial arrangement, with two washing treatments (washed vs. unwashed) and three storage temperatures, for a total duration of 42 days.

Percentage of egg weight loss

Egg weight loss was determined using 15 randomly selected eggs per treatment group. Weight

loss was calculated as the percentage difference between initial egg weight and the weight recorded at each storage interval (1, 3, 6, 9, 12, 15, 18, 21, 28, and 42 days) relative to the initial weight.

Egg quality measurements

Egg quality measurements were performed using 10 randomly selected eggs per treatment group at each storage interval (0, 1, 3, 6, 9, 12, 15, 18, 21, 28, and 42 days). The specific gravity of eggs was determined using the saline flotation method [18]. For this purpose, analytical grade sodium chloride (purity 99.5%) was dissolved in 3 L of distilled water to prepare solutions with specific gravities of 1.007, 1.015, 1.029, 1.035, 1.043, 1.060, 1.073, and 1.080 by dissolving 30, 60, 120, 150, 180, 240, 300, and 330 g of NaCl, respectively. The specific gravities were verified using a hydrometer and adjusted precisely by adding distilled water or additional NaCl. Egg weight, HU, albumen height, and yolk color were measured using an automatic egg quality analyzer (QCM+; Technical Services & Supplies Ltd., Dunnington, UK). After separating the yolk and albumen, albumen pH was determined using a pH meter (ST3100; Ohaus Corporation, Parsippany, NJ, USA). Yolk diameter and height were measured with a digital caliper (CD-20APX; Mitutoyo Corp., Kanagawa, Japan), and YI was calculated using the following equation:

$$YI = \frac{Yolk\ Height}{Yolk\ Width}, \quad (1)$$

Air cell diameter was measured by determining horizontal and vertical widths with a digital caliper (CD-20APX; Mitutoyo Corp.), and the mean value was used. Air cell depth was assessed using a digital height gauge (ACE-G3501; AICEYI, Guangdong, China) by recording the minimum and maximum depths of the air cell, with the average value expressed as the final result.

Statistical analyses

All data, except for egg weight loss, were analyzed using 10 eggs per treatment at each storage time, whereas egg weight loss was analyzed using 15 eggs per treatment. The individual egg was considered the experimental unit for measured egg quality traits. Statistical analyses were performed using the general linear model (GLM) procedure for two-way ANOVA in SPSS software (version 28.0;

SPSS Inc., Chicago, IL, USA). The main effects considered were washing treatment (washed vs. unwashed) and storage temperature. Results are presented as means ± standard errors. Post hoc comparisons among means were performed using Tukey's multiple range test at a 95% confidence level. Statistical significance was considered at $p<0.05$.

Results

Egg weight loss

The effects of egg washing and storage temperature on egg weight loss are presented in Table 1. Eggs stored at 20 ℃ consistently exhibited greater ($p < 0.01$) weight loss compared with those stored at 15 ℃. A significant interaction between egg washing and storage temperature was detected on days 1 and 3 ($p < 0.05$).

Egg weight

Table 2 summarizes the effects of egg washing and storage temperature on egg weight. At 18, 21, and 42 days of storage, eggs stored at 20 ℃ showed lower ($p < 0.05$) weights than those stored at 15 ℃.

Specific gravity

The effects of egg washing and storage temperature on specific gravity are presented in Table 3. On day 21 of storage, unwashed eggs showed a higher ($p < 0.05$) specific gravity than that of washed eggs. Eggs stored at 20 ℃ exhibited lower ($p < 0.05$) specific gravity values than those stored at 15 ℃ at all storage periods except for days 0 and 1. A significant interaction between egg washing and storage temperature was observed on day 6 ($p < 0.05$).

Albumen height

The effects of egg washing and storage temperature on albumen height are summarized in

Table 4. Eggs stored at 15 ℃ maintained greater ($p < 0.05$) albumen height than those stored at 20 ℃, except on days 0, 1, 6, and 42. A significant interaction between egg washing and storage temperature was detected on day 3 ($p < 0.01$), as washed eggs stored at 20 ℃ exhibited markedly lower albumen height than all other treatments.

Haugh unit

Table 5 summarizes the effects of egg washing and storage temperature on HU. Eggs stored at 15 ℃ exhibited significantly higher ($p < 0.01$) HU than those stored at 20 ℃ on all storage days except days 0, 1, 6, and 42. A significant interaction between egg washing and storage temperature was detected on day 3 ($p < 0.01$), as washed eggs stored at 20 ℃ showed markedly lower HU compared with all other treatments.

Albumen pH

Table 6 presents the effects of egg washing and storage temperature on albumen pH. On day 3 of storage, washed eggs showed higher ($p < 0.01$) albumen pH values than those of unwashed eggs. Eggs stored at 20 ℃ exhibited higher ($p < 0.001$) albumen pH than those stored at 15 ℃ at all storage periods except day 0. A significant interaction between egg washing and storage temperature was observed on day 9 ($p < 0.05$).

Yolk color

Table 7 summarizes the effects of egg washing and storage temperature on yolk color. Eggs stored at 20 ℃ showed significantly higher ($p < 0.05$) yolk color values than those stored at 15 ℃ on day 42 of storage. A significant interaction between egg washing and storage temperature was also observed on day 42 ($p < 0.01$), with washed eggs stored at 20 ℃ exhibiting higher yolk color values than those for washed eggs stored at 15 ℃.

Yolk index

The effects of egg washing and storage temperature on YI are summarized in Table 8. On day 21 of storage, washed eggs exhibited a lower ($p < 0.05$) YI than that of unwashed eggs. Eggs stored at 20 ℃ showed significantly lower ($p < 0.001$) YI values than those for eggs stored at 15 ℃ on days 12, 15, 21, 28, and 42.

Air cell dimension

Table 9 displays the effects of egg washing and storage temperature on air cell dimensions. On days 15 and 28 of storage, unwashed eggs had larger ($p < 0.05$) air cell dimensions than those for washed eggs. Eggs stored at 20 ℃ had significantly larger ($p < 0.001$) air cell dimensions than those stored at 15 ℃ at all storage periods except days 0 and 3. A significant interaction between egg washing and storage temperature was detected on day 3 ($p < 0.05$), as unwashed eggs stored at 20 ℃ and washed eggs stored at 15 and 20 ℃ exhibited larger air cell dimensions than those for unwashed eggs stored at 15 ℃.

Air cell depth

The effects of egg washing and storage temperature on air cell depth are presented in Table 10. On day 6 of storage, unwashed eggs exhibited a deeper ($p < 0.05$) air cell than that in washed eggs, whereas the opposite trend was observed on day 12, with washed eggs showing a deeper ($p < 0.05$) air cell than that in unwashed eggs. Eggs stored at 20 ℃ developed a significantly deeper ($p < 0.01$) air cell than those stored at 15 ℃ on all storage days except days 0, 1, and 6. Significant interactions between egg washing and storage temperature were detected on days 28 and 42 ($p < 0.05$).

Discussion

This study examined how storage temperature (15–20 ℃) and sodium hypochlorite washing shape internal and external egg quality over time. Temperature emerged as a dominant driver of egg quality deterioration within the 15–20 ℃ storage range, exerting substantially greater influence than

did sodium hypochlorite washing treatment. The current study demonstrated that higher storage temperatures were consistently associated with greater weight loss, lower specific gravity, enlarged air cells, elevated albumen pH, and coordinated declines in albumen height and HU. YI values were also lower when eggs were stored at elevated temperatures. These observations are consistent with previous studies identifying temperature as the primary determinant of freshness loss during storage and confirm that significant quality degradation occurs at temperatures well below the conventional room temperature threshold of 21–25 ℃. Previous studies comparing refrigeration (0–5 ℃) with markedly higher temperatures (21–29 ℃) have consistently demonstrated steep declines in albumen height, HU, and specific gravity at elevated temperatures [17,19,20]. The present experiment extends this understanding by demonstrating that deterioration initiates even before eggs reach classic room-temperature thresholds, with quality loss becoming substantially accelerated within the narrow 15–20 ℃ band typical of distribution and retail environments.

Storage temperature increases the diffusion rate of water vapor and carbon dioxide through eggshell pores in a temperature-dependent manner [21]. In the present study, eggs stored at 20 ℃ consistently exhibited significantly greater weight loss compared with those stored at 15 ℃. Because water vapor and carbon dioxide loss were not directly measured in the present study, the observed increase in egg weight loss should be interpreted as an indirect indicator of greater evaporative loss under higher storage temperature, as previously described in [17, 22]. The resulting decline in specific gravity directly reflects sustained evaporative loss with minimal compensatory volume change [23]. Specifically, the current results demonstrated that specific gravity decreased significantly with increasing storage temperature, with eggs stored at 20 ℃ showing lower values than those stored at 15 ℃ at all periods except days 0 and 1. Similarly, air-cell expansion, quantified by increased diameter and depth, is consistent with progressive enlargement of the internal gas space during storage, which has been associated in previous studies with moisture loss and carbon dioxide diffusion through the egg shell [22]. Remarkably, eggs stored at 20 ℃ developed significantly larger air cell dimensions than those stored at 15 ℃ throughout most of the storage period (days 6–42), with linear and quadratic increases observed as the temperature increased. These findings are consistent with kinetic models of

egg storage showing that temperature substantially impacts diffusion-limited processes controlling freshness indicators [21]. At 5, 19.5, and 30 ℃, Gogo et al [24] reported that storage temperature exerted significant effects on egg physical components, with effects becoming increasingly adverse as temperature rose. Moreover, storage at 19.5 ℃ led to an extensive reduction in HU, YI, and egg white height, while simultaneously increasing weight loss and albumen diameter compared with those at 5 ℃ storage, consistent with the present results showing deterioration within the 15–20 ℃ range.

Albumen pH elevation may provide an important indicator linking storage temperature with subsequent changes in albumen quality [25]. Samli et al. [17] reported that eggs from old laying hens stored at 32 ℃ exhibited albumen pH increases from approximately 7.47 at day 0 to 8.0–9.0 by day 7 of storage, whereas eggs maintained at 4 ℃ showed only marginal pH increases throughout the same period. In the present study conducted with 62-week-old hens, eggs stored at 20 ℃ showed significantly elevated albumen pH compared with those stored at 15 ℃ at all periods except day 0, indicating that even modest temperature differences within the 15–20 ℃ range can influence albumen alkalinization. Similar findings at intermediate temperatures have been reported by Cedro et al. [26], who reported that eggs stored at 25 ℃ presented lower HU and YI, and higher albumen and yolk pH compared with those kept at lower temperatures. Based on previous studies, the increase in albumen pH during storage is generally attributed to carbon dioxide diffusion from the egg and consequent changes in the carbonic acid-bicarbonate buffer system [25,27]. Although carbon dioxide loss, buffering capacity, and albumen protein chemistry were not directly measured in the present suty, the observed increase in albumen pH is consistent with this previously proposed mechanism. Therefore, the higher albumen pH observed at 20 ℃ may partly explain the concurrent reductions in albumen height and HU, but this interpretation should be considered inferential rather than directly demonstrated.

The rise in albumen pH may be associated with changes in the ovomucin–lysozyme protein complex, as proposed by [28]. Under acidic conditions (pH ~7.5), electrostatic repulsion within the complex is maximized, maintaining high viscosity and gel strength. As albumen becomes more alkaline during storage, alterations in electrostatic interactions within this complex have been suggested to reduce albumen viscosity and weaken gel integrity [28]. Although the present study did not directly

measure ovomucin, lysozyme, β-ovomucin stability, or protein interactions, the observed reductions in albumen height and HU are consistent with his proposed explanation. In the present study, eggs stored at 15 ℃ maintained significantly greater albumen height compared with those stored at 20 ℃ on most days (except days 0, 1, 6, and 42), with significant decreases in albumen height observed as temperature increased, particularly on days 9, 15, 18, 21, and 28. Correspondingly, HU was significantly higher in eggs stored at 15 ℃ compared with those under 20 ℃ storage (except days 0, 1, 6, and 42). These results extend earlier observations by Samli et al. [17], who noted the relationship between storage temperature and albumen structural integrity by providing quantitative evidence that even 5 ℃ differences within the distribution-relevant 15–20 ℃ range produce measurable declines in HU and albumen height. The more alkaline milieu reduces β-ovomucin stability and weakens electrostatic interactions within the ovomucin–lysozyme complex, lowering viscosity and gel strength [28]. This biochemical relaxation explains the paired declines in albumen height and HU observed in the present study and previous research on storage temperature effects.

Water progressively migrates from the albumen to the yolk during storage, driven by albumen protein hydration changes and diffusional water loss [29, 30]. Exposure to an increasingly alkaline albumen environment has been associated with reductions in vitelline membrane strength, likely through physicochemical changes that occur during storage [31]. The weakened membrane gradually loses structural rigidity, allowing the yolk to flatten and lose its characteristic spherical shape. These changes lead to lower YI values, providing a mechanistic explanation for the temperature-dependent alteration of yolk morphology. In the present study, YI was significantly lower in eggs stored at 20 ℃ compared with those under 15 ℃ storage on days 12, 15, 21, 28, and 42, with significant linear and quadratic decreases occurring in both unwashed and washed eggs as temperature increased. Notably, washed eggs showed a significantly lower YI than did unwashed eggs on day 21, suggesting that washing may increase susceptibility of the membrane to temperature-induced weakening. In addition to changes in yolk structure, prolonged storage also influences yolk pigmentation. Water moving from the albumen into the yolk dilutes yolk pigments, resulting in reduced yolk color intensity [32,33]. In the present study, however, the higher yolk color values observed at 20 ℃ on day 42 may reflect the

combined effects of pigment concentration due to dehydration and morphological changes occurring during storage. Storage time and temperature jointly affected YI, with higher temperatures associated with increased water transfer and greater susceptibility of the vitelline membrane to weakening, consistent with the progressive decline in YI observed at 20 ℃.

The coherence among weight loss, specific gravity decline, air-cell expansion, pH elevation, and albumen thinning suggests that these changes arise from interconnected processes related to shell permeability, temperature-driven moisture diffusion, and pH-dependent alterations in albumen structure [17]. Although these quality parameters are measured independently, their concurrent deterioration aligns with well-documented effects of temperature on gaseous and moisture exchange through the shell and on albumen protein stability. This linkage helps explain the proportional decline observed across all quality indices within the 15–20 ℃ range and clarifies why storage temperature exerts a stronger influence on long-term egg quality than washing procedures, which primarily modify only the initial shell surface barrier. In the present study, washing treatment did not produce significant differences in weight loss, specific gravity (except on day 21), albumen height, or HU for most of the storage period, whereas storage temperature (15 vs. 20 ℃) consistently generated marked differences across all measured variables. These findings support the view that storage temperature determines the longer-term trajectory of egg quality degradation regardless of washing status. The effects of storage time on weight loss, specific gravity, HU, albumen height, air-cell development, YI, and albumen and yolk pH have been extensively documented in eggs from hens of diverse physiological and genetic backgrounds [34]. Across these studies, storage temperature has consistently been shown to accelerate internal quality loss, while refrigeration substantially slows these changes compared with unrefrigerated conditions. This pattern is consistent with the present results, which demonstrate measurable deterioration even within the relatively narrow 15–20 ℃ range typical of distribution environments.

Sodium hypochlorite washing was associated with early changes in albumin quality, particularly under higher temperature storage conditions. In the present study, washed eggs stored at 20 ℃ on day 3 exhibited significantly lower albumen height and HU than those of unwashed eggs, along with higher albumen pH values. These early changes are consistent with previous reports showing

that egg washing can modify eggshell cuticle integrity and surface properties [13], which may temporarily increase shell permeability and facilitate water vapor and carbon dioxide loss during the initial period after washing. The resulting elevation in albumen pH could contribute to early reductions in gel stability. However, washing-related responses were not consistently observed across later storage times. Under the present experimental conditions, storage temperature showed a more consistent association with long-term changes in egg quality than washing treatment, as reflected by progressive weight loss, reduced specific gravity, air cell enlargement, increased albumen pH, and decreases in albumen height, HU, and YI. Because washing-related responses may be influenced by cuticle integrity, shell permeability, sanitizer concentration, wash-water temperature, drying conditions, and storage-unit variability, further studies incorporating direct measurements of cuticle coverage, shell conductance, and microbial status, together with larger independent storage-unit replication, are needed to clarify the sustained effects of washing on egg quality during extended storage. Throughout the 42-day storage period, temperature-dependent differences persisted across all measured traits, whereas washing effects remained limited and short-lived. Variability in washing outcomes observed across studies is also influenced by substantial differences among commercial washing protocols, including sanitizer concentration, contact time, wash-water temperature, brushing intensity, rinsing, and drying procedures, as well as inherent variation in eggshell cuticle structure among hen strains [35-37]. These factors collectively contribute to the wide range of washing-related quality effects reported in literature.

Conclusion

Within the 15–20 ℃ range, storage temperature was the primary determinant of internal and external egg quality over 42 days. Warmer storage consistently led to greater weight loss, lower specific gravity, enlarged air cells, higher albumen pH, reduced albumen height and HU, and a lower YI. Sodium hypochlorite washing produced only a transient early effect at warm temperatures and did not alter the longer-term deterioration trajectory governed by temperature. These findings identify temperature control as the primary control point for quality preservation along the supply chain. Maintaining storage

at or below 18 ℃, and closer to 15 ℃ when shelf life beyond 2 weeks is required, is a practical target. For routine monitoring, the early weight-loss slope, together with albumen pH, provides useful leading indicators of subsequent declines in HU and albumen structure. Standardizing washing parameters such as sanitizer level, contact time, wash-water temperature, and drying method can further minimize short-term variability while the dominant temperature effect is controlled.

Conflict of Interests

No potential conflict of interest relevant to this article was reported.

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445 **Table 1.** Effects of egg washing and storage temperature on egg weight loss (%).

Treatment		Storage period (Day)									
Egg washing	Storage temperature (℃)	1	3	6	9	12	15	18	21	28	42
	15	0.22^{ab}	0.58^{ab}	1.01	1.60	2.00	2.43	2.86	3.46	4.49	6.44
Unwashed eggs	18	0.18^{ab}	0.58^{ab}	1.08	1.74	2.23	2.76	3.30	3.94	5.10	7.46
	20	0.27^{b}	0.81^{cd}	1.54	2.52	3.22	3.93	4.65	5.59	7.29	10.69
	15	0.13^{a}	0.49^{a}	0.97	1.54	1.97	2.38	2.87	3.39	4.44	6.38
Washed eggs	18	0.23^{ab}	0.67^{bc}	1.24	1.90	2.38	2.94	3.43	4.10	5.22	7.55
	20	0.26^{b}	0.90^{d}	1.71	2.81	3.62	4.45	5.15	6.36	8.16	11.85
SEM[1)]		0.011	0.016	0.026	0.035	0.048	0.057	0.07	0.082	0.105	0.152
Main Effect											
Egg washing											
Unwashed eggs		0.23	0.66	1.21	1.95	2.48	3.04	3.60	4.33	5.63	8.20
Washed eggs		0.21	0.69	1.31	2.08	2.66	3.26	3.82	4.62	5.94	8.60
SEM[1)]		0.012	0.022	0.038	0.061	0.081	0.099	0.115	0.143	0.182	0.268
Storage temperature (℃)											
15		0.17^{a}	0.53^{a}	0.99^{a}	1.57^{a}	1.99^{a}	2.40^{a}	2.86^{a}	3.43^{a}	4.46^{a}	6.41^{a}
18		0.21^{ab}	0.63^{a}	1.16^{b}	1.82^{b}	2.30^{b}	2.85^{b}	3.37^{b}	4.02^{b}	5.16^{b}	7.51^{b}
20		0.27^{b}	0.86^{b}	1.63^{c}	2.67^{c}	3.42^{c}	4.19^{c}	4.90^{c}	5.98^{c}	7.72^{c}	11.27^{c}
SEM[1)]		0.011	0.016	0.026	0.036	0.049	0.059	0.070	0.085	0.107	0.153
p-value											

Egg washing	0.399	0.298	0.065	0.070	0.075	0.060	0.124	0.084	0.137	0.197
Storage temperature	0.003	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Egg washing × Storage temperature	0.033	0.033	0.165	0.148	0.208	0.132	0.330	0.103	0.170	0.206

446 [1)] Pooled standard error of the mean.

447 [a–d] Values in a row with different superscripts differ significantly (p<0.05).

448 **Table 2.** Effects of egg washing and storage temperature on egg weight (g)

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
	15	70.77	63.34	64.00	63.17	63.40	62.90	62.35	62.64	61.59	60.50	59.23
Unwashed eggs	18	70.29	63.81	63.65	63.53	62.59	62.41	62.33	62.82	61.45	60.37	59.14
	20	71.50	64.61	63.92	62.00	62.66	63.06	60.93	60.65	58.97	60.22	56.72
	15	70.65	63.07	63.47	62.52	63.06	63.16	62.15	61.96	62.42	59.63	60.04
Washed eggs	18	69.55	63.71	62.77	62.34	63.62	63.14	61.61	62.01	61.13	61.46	59.41
	20	71.31	63.86	62.99	62.48	61.17	61.89	61.96	59.34	60.57	59.33	58.35
SEM[1)]		0.392	0.261	0.267	0.266	0.294	0.275	0.295	0.286	0.291	0.278	0.288
Main Effect												
Egg washing												
Unwashed eggs		70.86	63.92	63.86	62.90	62.88	62.79	61.87	62.04	60.67	60.37	58.36
Washed eggs		70.50	63.55	63.08	62.45	62.62	62.73	61.90	61.10	61.38	60.14	59.27
SEM[1)]		0.387	0.259	0.26	0.264	0.301	0.272	0.292	0.313	0.310	0.282	0.305
Storage temperature (℃)												
15		70.71	63.21	63.74	62.85	63.23	63.03	62.25	62.30[b]	62.00b	60.06	59.64[b]
18		69.92	63.76	63.21	62.94	63.11	62.77	61.97	62.42[b]	61.29ab	60.92	59.27[b]
20		71.41	64.24	63.45	62.24	61.92	62.48	61.44	59.99[a]	59.77a	59.78	57.53[a]
SEM[1)]		0.383	0.256	0.266	0.264	0.294	0.273	0.291	0.286	0.292	0.278	0.289

p-value											
Egg washing	0.654	0.473	0.150	0.395	0.649	0.911	0.952	0.108	0.231	0.686	0.123
Storage temperature	0.310	0.280	0.725	0.515	0.141	0.709	0.531	0.001	0.009	0.231	0.010
Egg washing × Storage temperature	0.940	0.872	0.945	0.430	0.225	0.343	0.467	0.896	0.407	0.258	0.625

449 [1)] Pooled standard error of the mean.

450 [a,b] Values in a row with different superscripts differ significantly ($p<0.05$).

Table 3. Effects of egg washing and storage temperature on egg specific gravity

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
Unwashed eggs	15	1.078	1.080	1.080	1.077^{ab}	1.069	1.064	1.058	1.055	1.060	1.048	1.016
	18	1.080	1.080	1.080	1.070^{ab}	1.072	1.058	1.057	1.048	1.048	1.036	1.015
	20	1.080	1.079	1.078	1.073^{ab}	1.057	1.055	1.043	1.030	1.031	1.020	0.998
Washed eggs	15	1.079	1.080	1.079	1.077^{ab}	1.075	1.064	1.058	1.055	1.049	1.042	1.018
	18	1.080	1.080	1.080	1.077^{b}	1.067	1.065	1.055	1.051	1.045	1.035	1.015
	20	1.080	1.080	1.076	1.069^{a}	1.061	1.047	1.041	1.040	1.028	1.015	0.998
SEM[1)]		0.0000	0.0000	0.0000	0.0010	0.0010	0.0010	0.0010	0.0010	0.0010	0.0010	0.0010
Main Effect												
Egg washing												
Unwashed eggs		1.079	1.080	1.079	1.073	1.066	1.059	1.053	1.044	1.047	1.035	1.010
Washed eggs		1.080	1.080	1.078	1.074	1.068	1.059	1.051	1.048	1.040	1.030	1.010
SEM[1)]		0.0000	0.0000	0.0000	0.0010	0.0010	0.0010	0.0010	0.0020	0.0020	0.0020	0.0020
Storage temperature (℃)												
15		1.079	1.080	1.080^{b}	1.077^{b}	1.072^{b}	1.064^{b}	1.058^{b}	1.055^{b}	1.055^{c}	1.045^{c}	1.017^{b}
18		1.080	1.080	1.080^{b}	1.074^{ab}	1.070^{b}	1.061^{b}	1.056^{b}	1.049^{b}	1.046^{b}	1.035^{b}	1.015^{b}
20		1.080	1.080	1.077^{a}	1.071^{a}	1.059^{a}	1.051^{a}	1.042^{a}	1.035^{a}	1.030^{a}	1.017^{a}	0.998^{a}
SEM[1)]		0.0000	0.0000	0.0000	0.0010	0.0010	0.0010	0.0010	0.0010	0.0010	0.0010	0.0010

p-value											
Egg washing	0.542	0.322	0.269	0.380	0.427	0.937	0.498	0.080	0.019	0.064	0.776
Storage temperature	0.207	0.375	0.005	0.016	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Egg washing × Storage temperature	0.688	0.375	0.592	0.020	0.148	0.074	0.889	0.252	0.398	0.671	0.921

452 [1)] Pooled standard error of the mean.

453 [a–c] Values in a row with different superscripts differ significantly ($p<0.05$).

454 **Table 4.** Effects of egg washing and storage temperature on albumen height (mm)

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
	15	7.91	7.76	7.21^{b}	6.38	6.54	6.28	5.95	5.69	4.84	4.89	3.93
Unwashed eggs	18	7.79	7.60	7.10^{b}	6.33	5.85	5.64	4.97	5.05	4.21	4.31	3.88
	20	8.14	7.58	7.24^{b}	5.87	5.30	5.32	4.57	4.24	3.51	4.06	3.56
	15	7.81	7.52	7.94^{b}	6.18	6.53	6.14	6.16	5.91	5.19	4.99	4.21
Washed eggs	18	8.06	7.05	7.08^{b}	6.31	5.96	5.60	5.64	4.96	4.01	4.72	3.98
	20	8.06	7.41	5.90^{a}	5.88	5.88	4.85	4.43	4.24	3.91	3.85	3.87
SEM[1)]		0.111	0.129	0.108	0.077	0.095	0.100	0.094	0.090	0.099	0.074	0.061
Main Effect												
Egg washing												
Unwashed eggs		7.95	7.65	7.18	6.19	5.90	5.75	5.17	4.99	4.18	4.42	3.79
Washed eggs		7.98	7.33	6.98	6.12	6.12	5.53	5.41	5.04	4.37	4.52	4.02
SEM[1)]		0.109	0.127	0.13	0.079	0.107	0.115	0.125	0.121	0.121	0.09	0.062
Storage temperature (℃)												
15		7.86	7.64	7.58^{b}	6.28^{ab}	6.53^{b}	6.21^{b}	6.06^{c}	5.80^{c}	5.02^{b}	4.94^{b}	4.07
18		7.93	7.33	7.09^{ab}	6.32^{b}	5.91^{a}	5.62^{ab}	5.31^{b}	5.00^{b}	4.11^{a}	4.51^{b}	3.93
20		8.10	7.50	6.57^{a}	5.87^{a}	5.59^{a}	5.08^{a}	4.50^{a}	4.24^{a}	3.71^{a}	3.95^{a}	3.71
SEM[1)]		0.109	0.128	0.121	0.075	0.095	0.099	0.095	0.088	0.099	0.074	0.062

p-value											
Egg washing	0.903	0.218	0.342	0.665	0.243	0.284	0.199	0.801	0.355	0.522	0.065
Storage temperature	0.670	0.610	0.002	0.040	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.064
Egg washing × Storage temperature	0.753	0.810	0.001	0.829	0.420	0.657	0.222	0.777	0.411	0.242	0.743

455 [1)] Pooled standard error of the mean.

456 [a–c] Values in a row with different superscripts differ significantly ($p<0.05$).

Table 5. Effects of egg washing and storage temperature on Haugh unit

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
Unwashed eggs	15	86.07	87.13	83.52^{b}	78.30	79.45	77.43	75.25	72.82	65.80	66.79	57.93
	18	85.20	86.02	82.95^{b}	77.73	74.39	72.91	67.26	67.46	59.19	61.68	57.61
	20	87.07	85.53	83.68^{b}	74.95	69.99	69.50	63.92	60.71	52.59	59.09	55.05
Washed eggs	15	85.43	85.89	88.27^{b}	77.07	79.55	76.65	77.20	75.21	68.96	68.48	60.79
	18	87.21	81.78	83.05^{b}	78.20	74.60	72.25	73.12	67.28	57.57	65.28	58.71
	20	86.98	84.51	74.22^{a}	74.96	75.07	66.03	61.26	61.49	56.97	57.04	57.81
SEM[1)]		0.697	0.904	0.767	0.556	0.738	0.814	0.850	0.780	1.092	0.719	0.690
Main Effect												
Egg washing												
Unwashed eggs		86.12	86.23	83.38	76.99	74.61	73.28	68.81	67.00	59.19	62.52	56.86
Washed eggs		86.54	84.06	81.84	76.74	76.40	71.64	70.53	67.99	61.16	63.60	59.10
SEM[1)]		0.680	0.888	0.916	0.567	0.823	0.934	1.123	1.025	1.274	0.877	0.687
Storage temperature (℃)												
15		85.75	86.51	85.89^{b}	77.68	79.50^{b}	77.04^{b}	76.22^{c}	74.02^{c}	67.38^{b}	67.63^{b}	59.36
18		86.21	83.90	83.00^{ab}	77.96	74.50^{a}	72.58^{b}	70.19^{b}	67.37^{b}	58.38^{a}	63.48^{b}	58.16
20		87.03	85.02	78.95^{a}	74.96	72.53^{a}	67.77^{a}	62.59^{a}	61.10^{a}	54.78^{a}	58.06^{a}	56.43
SEM[1)]		0.683	0.896	0.849	0.543	0.744	0.805	0.866	0.765	1.084	0.720	0.690

p-value											
Egg washing	0.760	0.236	0.321	0.824	0.229	0.319	0.317	0.527	0.371	0.456	0.110
Storage temperature	0.751	0.502	0.002	0.059	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.228
Egg washing × Storage temperature	0.717	0.720	0.001	0.814	0.299	0.730	0.132	0.795	0.499	0.273	0.845

458 [1)] Pooled standard error of the mean.

459 [a–c] Values in a row with different superscripts differ significantly ($p<0.05$).

460 **Table 6.** Effects of egg washing and storage temperature on albumen pH

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
Unwashed eggs	15	8.37	8.98	9.26	9.63	9.64[a]	9.62	9.67	9.64	9.71	9.70	9.72
	18	8.40	9.03	9.32	9.70	9.71[abc]	9.67	9.73	9.73	9.74	9.72	9.74
	20	8.34	9.09	9.41	9.75	9.84[d]	9.78	9.77	9.72	9.77	9.77	9.76
Washed eggs	15	8.42	8.88	9.32	9.62	9.69[ab]	9.66	9.68	9.66	9.70	9.70	9.71
	18	8.47	9.05	9.37	9.71	9.75[bc]	9.73	9.68	9.73	9.74	9.72	9.74
	20	8.41	9.14	9.47	9.77	9.79[cd]	9.77	9.75	9.76	9.77	9.77	9.78
SEM[1)]		0.021	0.014	0.009	0.007	0.008	0.012	0.007	0.009	0.006	0.005	0.004
Main Effect												
Egg washing												
Unwashed eggs		8.37	9.03	9.33	9.69	9.73	9.69	9.72	9.70	9.74	9.73	9.74
Washed eggs		8.43	9.02	9.39	9.70	9.74	9.72	9.70	9.72	9.74	9.73	9.74
SEM[1)]		0.02	0.017	0.012	0.01	0.012	0.013	0.009	0.011	0.007	0.006	0.005
Storage temperature (℃)												
15		8.40	8.93[a]	9.29[a]	9.63[a]	9.66[a]	9.64[a]	9.67[a]	9.65[a]	9.71[a]	9.70[a]	9.72[a]
18		8.43	9.04[b]	9.34[b]	9.70[b]	9.73[b]	9.70[a]	9.71[a]	9.73[b]	9.74[ab]	9.72[a]	9.74[a]
20		8.38	9.11[b]	9.44[c]	9.76[c]	9.82[c]	9.78[b]	9.76[b]	9.74[b]	9.77[b]	9.77[b]	9.77[b]
SEM[1)]		0.021	0.014	0.01	0.007	0.008	0.012	0.008	0.009	0.006	0.005	0.004

p-value											
Egg washing	0.127	0.733	0.003	0.723	0.389	0.207	0.114	0.319	0.674	0.892	0.629
Storage temperature	0.503	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Egg washing × Storage temperature	0.982	0.069	0.978	0.657	0.011	0.441	0.221	0.764	0.951	0.974	0.289

461 [1)] Pooled standard error of the mean.

462 [a–d] Values in a row with different superscripts differ significantly ($p<0.05$).

463 **Table 7.** Effects of egg washing and storage temperature on yolk color

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
Unwashed eggs	15	9.20	8.40	8.80	8.40	8.00	7.50	7.90	8.60	8.10	9.10	9.60[ab]
	18	8.70	8.20	7.90	8.20	7.30	7.60	8.20	8.40	9.40	9.20	9.60[ab]
	20	8.80	8.50	7.80	7.90	8.00	8.30	8.30	7.90	9.50	8.90	9.50[ab]
Washed eggs	15	8.40	8.70	8.60	8.10	7.50	7.90	8.10	8.50	9.40	8.40	9.00[a]
	18	8.60	8.20	8.20	7.60	7.80	8.10	7.80	8.40	9.40	9.20	9.40[ab]
	20	8.60	8.50	8.50	8.10	8.20	8.80	8.10	8.60	10.00	9.00	10.00[b]
SEM[1)]		0.106	0.115	0.115	0.118	0.182	0.156	0.163	0.171	0.315	0.092	0.065
Main Effect												
Egg washing												
Unwashed eggs		8.90	8.37	8.17	8.17	7.77	7.80	8.13	8.30	9.00	9.07	9.57
Washed eggs		8.53	8.47	8.43	7.93	7.83	8.27	8.00	8.50	9.60	8.87	9.47
SEM[1)]		0.105	0.113	0.119	0.117	0.181	0.158	0.159	0.168	0.31	0.095	0.074
Storage temperature (℃)												
15		8.80	8.55	8.70	8.25	7.75	7.70	8.00	8.55	8.75	8.75	9.30[a]
18		8.65	8.20	8.05	7.90	7.55	7.85	8.00	8.40	9.40	9.20	9.50[ab]
20		8.70	8.50	8.15	8.00	8.10	8.55	8.20	8.25	9.75	8.95	9.75[b]
SEM[1)]		0.108	0.112	0.116	0.118	0.180	0.155	0.160	0.169	0.311	0.094	0.070

p-value											
Egg washing	0.090	0.665	0.250	0.327	0.856	0.139	0.684	0.562	0.344	0.282	0.445
Storage temperature	0.842	0.410	0.053	0.464	0.465	0.067	0.846	0.775	0.426	0.145	0.024
Egg washing × Storage temperature	0.353	0.828	0.284	0.383	0.521	0.989	0.747	0.586	0.698	0.164	0.004

464 [1)] Pooled standard error of the mean.

465 [a,b] Values in a row with different superscripts differ significantly ($p<0.05$).

Table 8. Effects of egg washing and storage temperature on yolk index

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
	15	0.39	0.39	0.39	0.38	0.37	0.37	0.36	0.37	0.32	0.31	0.30
Unwashed eggs	18	0.39	0.38	0.39	0.36	0.36	0.34	0.34	0.33	0.29	0.29	0.27
	20	0.39	0.39	0.37	0.35	0.35	0.32	0.31	0.30	0.27	0.25	0.22
	15	0.39	0.40	0.38	0.37	0.36	0.36	0.36	0.37	0.33	0.32	0.29
Washed eggs	18	0.41	0.39	0.38	0.37	0.35	0.33	0.34	0.39	0.30	0.29	0.28
	20	0.40	0.38	0.37	0.36	0.34	0.32	0.33	0.31	0.29	0.28	0.23
SEM[1)]		0.003	0.003	0.004	0.003	0.003	0.004	0.003	0.012	0.003	0.003	0.003
Main Effect												
Egg washing												
Unwashed eggs		0.39	0.39	0.38	0.36	0.36	0.34	0.34	0.33	0.29	0.28	0.26
Washed eggs		0.40	0.39	0.38	0.37	0.35	0.34	0.34	0.35	0.31	0.29	0.27
SEM[1)]		0.003	0.003	0.004	0.004	0.003	0.004	0.004	0.012	0.004	0.004	0.005
Storage temperature (℃)												
15		0.39	0.40	0.38	0.37	0.36	0.37[b]	0.36[b]	0.37	0.33[c]	0.31[c]	0.30[c]
18		0.40	0.38	0.39	0.36	0.36	0.34[a]	0.34[a]	0.36	0.30[b]	0.29[b]	0.28[b]
20		0.40	0.38	0.37	0.36	0.35	0.32[a]	0.32[a]	0.30	0.28[a]	0.26[a]	0.23[a]
SEM[1)]		0.003	0.003	0.004	0.003	0.003	0.004	0.003	0.012	0.003	0.003	0.003

p-value											
Egg washing	0.051	0.483	0.468	0.699	0.174	0.520	0.629	0.351	0.016	0.080	0.351
Storage temperature	0.638	0.099	0.450	0.134	0.173	<0.001	<0.001	0.057	<0.001	<0.001	<0.001
Egg washing × Storage temperature	0.583	0.570	0.781	0.241	0.972	0.928	0.332	0.560	0.441	0.143	0.127

467 [1)] Pooled standard error of the mean.

468 [a–c] Values in a row with different superscripts differ significantly ($p<0.05$).

469 **Table 9.** Effects of egg washing and storage temperature on air cell dimension (mm)

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
	15	17.35	17.86	18.30[a]	20.71	21.74	22.69	24.91	24.71	27.52	28.65	31.61
Unwashed eggs	18	16.17	17.11	19.04[ab]	20.76	22.31	23.72	25.32	26.32	27.61	30.10	31.16
	20	16.56	17.76	20.16[b]	22.47	24.77	25.33	29.53	26.98	30.74	32.68	33.85
	15	17.34	17.33	19.57[b]	20.69	22.12	22.71	22.92	24.66	28.15	27.54	31.61
Washed eggs	18	17.07	17.64	19.03[ab]	21.09	22.99	24.41	25.24	26.85	29.01	28.89	32.76
	20	16.69	17.77	20.15[b]	22.43	24.70	26.30	26.38	26.75	31.73	31.37	33.62
SEM[1)]		0.141	0.155	0.114	0.143	0.162	0.199	0.300	0.234	0.289	0.242	0.228
Main Effect												
Egg washing												
Unwashed eggs		16.69	17.58	19.17	21.32	22.94	23.91	26.59	26.01	28.62	30.48	32.21
Washed eggs		17.03	17.58	19.58	21.40	23.27	24.47	24.85	26.09	29.63	29.27	32.66
SEM[1)]		0.146	0.153	0.138	0.172	0.222	0.256	0.373	0.26	0.342	0.317	0.257
Storage temperature (℃)												
15		17.34	17.60	18.93[a]	20.70[a]	21.93[a]	22.70[a]	23.91[a]	24.69[a]	27.83[a]	28.10[a]	31.61[a]
18		16.62	17.37	19.03[a]	20.93[a]	22.65[a]	24.07[b]	25.28[a]	26.59[b]	28.31[a]	29.49[a]	31.96[a]
20		16.62	17.77	20.16[b]	22.45[b]	24.74[b]	25.81[c]	27.95[b]	26.87[b]	31.23[b]	32.03[b]	33.73[b]
SEM[1)]		0.142	0.153	0.121	0.139	0.160	0.199	0.325	0.229	0.29	0.249	0.230

p-value											
Egg washing	0.237	0.997	0.073	0.764	0.319	0.164	0.005	0.864	0.087	0.015	0.324
Storage temperature	0.063	0.587	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Egg washing × Storage temperature	0.372	0.385	0.037	0.841	0.642	0.606	0.118	0.787	0.861	0.985	0.211

[1)] Pooled standard error of the mean.

[a,b] Values in a row with different superscripts differ significantly ($p<0.05$).

Table 10. Effects of egg washing and storage temperature on air cell depth (mm)

Treatment		Storage period (Day)										
Egg washing	Storage temperature (℃)	0	1	3	6	9	12	15	18	21	28	42
Unwashed eggs	15	2.59	2.78	3.14	4.23	4.03	4.01	4.38	5.27	5.48	6.86^{abc}	9.38^{ab}
	18	2.80	3.00	3.45	4.63	4.49	4.53	5.23	5.87	7.26	8.3^{bcd}	8.47^{ab}
	20	2.89	2.77	3.64	4.57	5.25	5.59	6.67	6.51	7.67	8.78^{cd}	11.87^{c}
Washed eggs	15	2.89	2.80	2.95	3.99	4.13	4.81	5.04	5.63	6.63	6.51^{ab}	8.25^{a}
	18	2.67	2.67	3.44	3.96	4.52	4.90	5.03	6.56	7.67	6.04^{a}	10.10^{abc}
	20	2.68	2.94	3.62	4.49	5.36	5.46	5.94	6.47	8.10	9.01^{d}	10.31^{bc}
SEM[1)]		0.066	0.073	0.074	0.081	0.070	0.084	0.121	0.122	0.200	0.198	0.200
Main Effect												
Egg washing												
Unwashed eggs		2.76	2.85	3.41	4.48	4.59	4.71	5.43	5.88	6.80	7.98	9.91
Washed eggs		2.75	2.80	3.34	4.15	4.67	5.06	5.33	6.22	7.47	7.19	9.55
SEM[1)]		0.066	0.071	0.079	0.083	0.095	0.105	0.151	0.133	0.220	0.238	0.250
Storage temperature (℃)												
15		2.74	2.79	3.04^{a}	4.11	4.08^{a}	4.41^{a}	4.71^{a}	5.45^{a}	6.05^{a}	6.68^{a}	8.82^{a}
18		2.74	2.83	3.45^{ab}	4.30	4.51^{b}	4.72^{a}	5.13^{a}	6.22^{b}	7.47^{b}	7.17^{a}	9.28^{a}
20		2.79	2.86	3.63^{b}	4.53	5.31^{c}	5.53^{b}	6.31^{b}	6.49^{b}	7.88^{b}	8.90^{b}	11.09^{b}
SEM[1)]		0.066	0.072	0.073	0.083	0.068	0.088	0.123	0.122	0.201	0.211	0.217

p-value											
Egg washing	0.902	0.749	0.616	0.046	0.564	0.043	0.691	0.171	0.102	0.05	0.385
Storage temperature	0.942	0.938	0.007	0.113	<0.001	<0.001	<0.001	0.003	0.001	<0.001	<0.001
Egg washing × Storage temperature	0.261	0.368	0.853	0.326	0.966	0.086	0.069	0.475	0.688	0.033	0.004

473 [1)] Pooled standard error of the mean.

474 [a–c] Values in a row with different superscripts differ significantly ($p<0.05$).