Brazilian Journal of Poultry Science
Revista Brasileira de Ciência Avícola

e-ISSN: 1806-9061 2024 / v.26 / n.2 / 001-010

http://dx.doi.org/10.1590/1806-9061-2024-1902

Original Article

The Impact of Laying Hen Age, Egg-Laying Time, Cage Tier, and Cage Direction on Egg Quality Traits in Hens in an Enriched Cage System

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■Keywords

Laying hens, enriched cage system, egg quality traits, egg-laying time, cage positioning.

Submitted: 11/January/2024
Approved: 16/April/2024

ABSTRACT

This study determined the effect of layer age, egg-laying time, cage tier, and cage direction (outward-inward) on egg quality parameters of commercial layer chickens reared in enriched cages. A total of 944 layers (approximately 100 eggs every four weeks, from the 26 to the 58th week) obtained from Lohmann LSL-Classic layers reared at the Niğde Ömer Halisdemir University Ayhan Şahenk Agricultural Application and Research Centre were analyzed. Laying hen age significantly affected all the egg quality variables (p<0.01). Egg weight, shell-breaking strength, egg surface area, Haugh unit, and yolk color score differed across egg-laying time (p<0.05; p<0.01). There was variability in all the external egg quality traits among the cage tiers (p<0.05), but not for internal egg quality (p>0.05). Furthermore, it was found that cage direction had a significant effect on egg weight, shell thickness, egg surface area, and all the internal egg quality traits, except for yolk color score (p<0.05; p<0.01). In conclusion, our results highlight significant changes in egg quality traits due to layer age, egg-laying time, cage tier, and cage direction.

INTRODUCTION

In some countries (e.g., the European Union and the United Kingdom), the ban on conventional cage systems for laying hens (Directive EU, 2012) resulted in a significant shift to alternative systems, including enriched cage systems. With consumer attitudes under consideration, the enriched cage systems are developed with enhanced features such as increased floor space for birds, nesting area, and perches. Additionally, it has been shown that, to some extent, this system can meet the behavioral and welfare requirements of hens (Appleby *et al.*, 2002; FAW Council, 2007; Lay *et al.,* 2011; Tainika & Şekeroğlu, 2020).

Regardless of the production system, it is well-known that egg quality traits vary over the egg-laying period. For instance, Silversides & Scott (2001) respectively identified an increase and decrease in egg weight and albumen height with advancing ages in layer genotypes. Roberts *et al.* (2013) found that albumen height and Haugh unit reduced with an increase in layer ages. A number of studies have confirmed the significant effect of layer age on egg quality traits (Yılmaz Dikmen *et al.,* 2017; Samiullah *et al.,* 2017; Kowalska *et al.,* 2021; Nowaczewski *et al.,* 2021). Contrarily, Chung & Lee (2014) did not identify any effect of age on egg weight, Haugh unit, and shell thickness in Hy-Line Brown layers.

Previous studies have also reported the impact of egg-laying or collection time on some egg quality traits. For example, Tůmová *et al.* (2009) reported that the heaviest eggs were obtained at 06:00 as compared to 10:00 and 14:00, and shell-breaking strength differed between egg-laying times. Krawczyk *et al.* (2023) observed decreased egg weight, and increased albumen pH in eggs collected from 07:00 compared to 13:00. However, the latter authors did not identify differences in Haugh unit, shell thickness, shell breaking strength, and yolk color between eggs collected from 07:00 and 13:00.

Furthermore, there are reports on the effect of enriched cage system characteristics (e.g., cage tier and orientation) on egg quality traits. However, these reports are too scarce to provide clear evidence about the role of enriched cage properties in modifying egg quality traits.

Some previous studies have revealed no significant effect of cage tiers on egg quality traits (Sekeroglu *et al.,* 2014; Tunaydin & Yilmaz Dikmen, 2019). On the other hand, Eleroğlu (2019) reported a significant effect of cage tier on egg weight, shell-breaking strength, and albumen height. Akkus & Yildirim (2018) observed the highest egg weight, shape index, and shell thickness in eggs collected from the first cage tier, and the highest shell-breaking strength in eggs from the 3rd cage tier.

Studies on the effect of cage direction on internal and external egg quality traits are scarce and contradictory. For example, Yildiz *et al.* (2006) identified that cage direction affected egg weight, shell strength, albumen index, and Haugh unit; but Sahin (2012) showed that cage direction does not lead to significant changes in internal and external egg quality traits.

Since the introduction of enriched cages in the 1980s (Lay *et al.,* 2011), research has mainly focused on the influence of the manipulation of furniture items on the performance of hens. This has left a knowledge gap on whether other characteristics including cage tier and direction can impact egg quality traits. Engel *et al.* (2019) argued that the disruption of the biological function in hens can impact their physiological responses and impair their performance. Thus, it is speculated that there can be dissimilarity in elements such as light intensity level, and dust in cages with different tiers and positioning. This might adversely influence the biological activities of hens and consequently affect the physiological parameters related to egg quality traits.

Therefore, this study investigated the effect of laying hen age, egg-laying time, cage direction, and cage tier on the egg quality traits of Lohmann LSL-Classic laying hens reared in an enriched cage system.

MATERIAL AND METHODS

The eggs used in the present study were obtained from the "Lohmann LSL-Classic" commercial layers reared at Niğde Ömer Halisdemir University Ayhan Şahenk Agricultural Application and Research Centre – Laying hens unit.

These birds had been purchased from a private commercial enterprise at 16 weeks of age and produced in a poultry house, which consisted of 3 parallel cage lines of 3 tiers each. There was a total of 120 cage units (40 cage units per cage tier) on each cage line. Each cage unit measured 240 cm × 63.5 cm × 60 cm: length (L), width (W), and height (H), respectively. Additionally, each cage unit had a stainless-steel nipple drinking system, two parallel perches with nail shortener (each 180 cm long), a nesting area covered with dark blue curtains (40 cm × 33.5 cm × 30 cm; L × W × H), and scratchpad. The cage system was completed with a wire mesh floor, an egg conveyor belt, an automatic feeder system, and a polypropylene manure conveyor belt.

From 16 to 20 weeks of age, the hens were reared under a photoperiod of 12 hours of light (L) and 12 hours of darkness (D) (12L:12D). From 20 weeks, the photoperiod per week was increased by 30 min until 16L:8D, which was maintained up to week 54 (end of the study). The lighting program of the Lohmann management guide was followed (Lohmann, 2021). The light source was provided by 24-W light-emitting diode bulbs (20 lux - 3.2w / m^2).

From 16 to 18 weeks of age, birds received layer developer feed: 15% CP, 1% Ca, 0.37% P, and 11.514 MJ/Kg metabolic energy; from 19 weeks to 2% egg production, pre-laying feed: 17% CP, 2.00% Ca, 0.55% P, and 11.723 MJ/Kg metabolic energy; and from 2% egg production to 52 weeks of age, layer feed: 16.26% CP, 3.58% Ca, 0.44% P, and 11.723 MJ/Kg metabolic energy. Feeding and water were offered *ad-libitum.*

Throughout the study, birds were vaccinated as detailed in the management guide issued by the breeder company and as required by the region. All biosecurity procedures were also followed.

During the study, a single cage line, located near the wall, was utilized as the experimental area. The cage units in each tier were designed facing in opposite directions (20 facing outward (the wall) and 20 facing inward (the corridor) inside of the poultry house). Therefore, cage direction was either outward and inward of the poultry house. Cage tiers were marked as top, middle, or bottom: 3rd, 2nd, and 1st, respectively.

Moreover, a total of 18 cage units (9 inward or 9 outwards) positioned as the 5, 10, and 15th cage units were designated to represent each cage tier. The experimental plan of the study is shown in Figure.

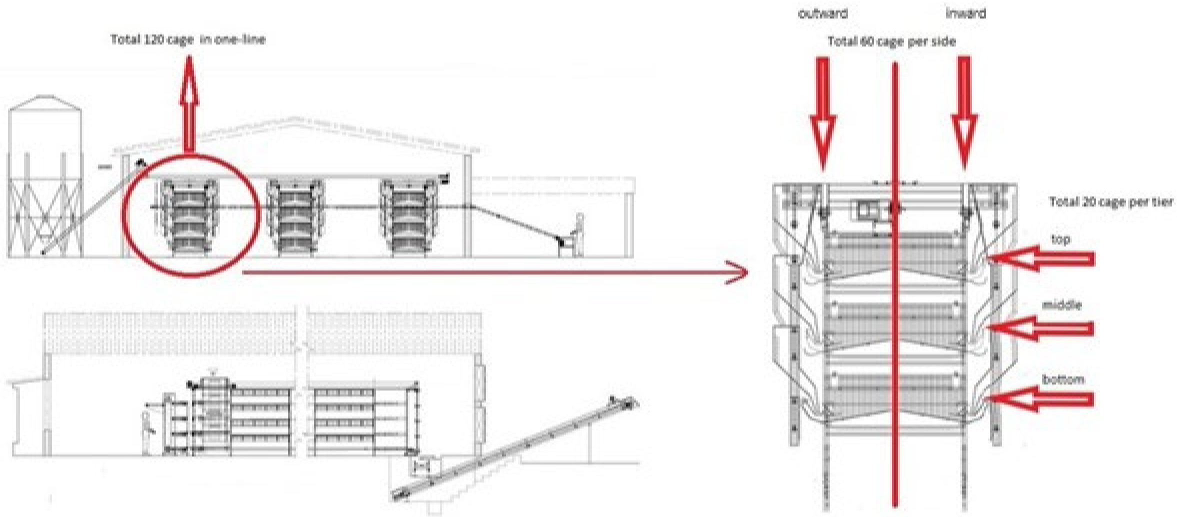

Figure – The enriched cage system showing the study design.

Data collection

Approximately 100 eggs were randomly collected once a day every 4 weeks (26, 30, 34, 38, 42, 46, 50, 54, and 58 weeks of age). This considered the egg-laying time, cage tier, and cage direction. Considering the laying time, 315 eggs in the morning (8:30 a.m.), 316 eggs at noon (12:30 p.m.), and 314 eggs in the evening (4:30 p.m.) were analyzed. In terms of cage direction, 468 eggs were analyzed from the cages facing outside the poultry house and 474 eggs from the cages facing the inside of the poultry house. For the cage tier, 316 eggs from the 1st tier, 310 from the 2nd tier, and 316 eggs from the 3rd tier were analyzed. In total, 944 eggs were analyzed.

The eggs were taken to the laboratory and stored for 24 hours at room temperature before internal and external egg quality trait analyses were conducted.

Egg quality traits were analyzed following the procedures reported by Stadelman and Cotterill (1995) and Altan (2015). Egg weight (g) was measured with a weighing scale of 0.01-gram precision. Egg width (EW) and egg length (EL) were determined with a digital caliper to calculate shape index; Shape index (SI, %) = (EW / EL) × 100. Eggshell breaking strength (Kg. f) was determined with an egg force reader (Orka food tech. FGV-10XY (5.000 kg) EFO493/2013). ESA (cm^2) was calculated as 3.9782 × egg weight in grams $^{0.70}$, according to Carter (1975).

Subsequently, eggs were broken on a special glass table to assess internal egg quality traits. After a pause of 10 min, the yolk height was determined with a 3-foot 0.01 mm sensitivity manual micrometer. Albumen length, albumen width, and yolk diameter were measured with a digital caliper (0 - 150 mm), and the albumen and yolk index were calculated using the formulas below:

Albumen index (AI, %) = ((egg albumen height (mm)) / ((egg albumen length (mm) + egg albumen width (mm))/2)) x 100.

Yolk index (YI, %) = (Egg yolk height (mm) / egg yolk diameter (mm)) x 100.

Haugh unit was calculated using the following formula by Haugh (1934): 100 log (H+7.57 – 1.7 $W^{0.37}$)), H; albumen height (mm) W; egg weight (g).

Shell thickness was determined as the average of shell thickness measurements for samples taken from the blunt, center, and pointed regions of eggs without eggshell membranes, with a manual metric micrometer (0.01 mm – 0-10 mm). Finally, a DSM yolk color fan was used to determine the yolk color.

Statistical analyses

In the study, the normality assumption was examined with the Shapiro-Wilk test, and the variance homogeneity test with the Levene test. It was determined that the data met the assumption of normal distribution (p>0.05) and their variances were homogeneous (p>0.05). For this reason, analysis of variance was used to analyze the data. The following model was used in the analysis of the variables:

$$Y_{ijklm} = \mu + \alpha_i + \beta_j + \gamma_k + \delta_l + \alpha\beta_{ij} + \alpha\gamma_{ik} + \alpha\delta_{il} + \beta\gamma_{jk} + \beta\delta_{jl} + \gamma\delta_{kl} + \alpha\beta\gamma_{ijk} + \alpha\beta\delta_{ijl} + \alpha\gamma\delta_{ikl} + \beta\gamma\delta_{jkl} + \alpha\beta\gamma\delta_{ijkl} + \varepsilon_{ijklm}$$

Where Y_{ijklm}: observation value, μ: population mean α_i: i. age effect (week),β_j: j. egg-laying time, γ_k: k. cage tier effect, δ_l: l. cage direction effect, $\alpha\beta_{ij}$; $\alpha\gamma_{ik}$; $\alpha\delta_{il}$; $\beta\gamma_{jk}$; $\beta\delta_{jl}$; $\gamma\delta_{kl}$; $\alpha\beta\gamma_{ijk}$; $\alpha\beta\delta_{ijl}$ + $\alpha\gamma\delta_{ikl}$; $\beta\gamma\delta_{jkl}$; $\alpha\beta\gamma\delta_{ijkl}$:interaction effects and ε_{ijklm}: random error.

As descriptive statistics values, the mean of the observations and the standard error of the mean are given. Duncan's multiple comparison test was used to determine differences between groups. The IBM SPSS 21 package program was used for all statistical procedures (IBM Corp., 2012).

RESULTS AND DISCUSSION

The data for the impact of layer age, egg-laying time, cage tier, and cage direction on internal and external egg quality traits is shown in Tables 1 and 2, respectively.

Table 1 – Effect of layer age (weeks), egg-laying time, cage tier, and cage direction on external egg quality characteristics (mean ± SEM).

	Egg weight (g)	Shape index (%)	Shell thickness (mm)	Shell breaking strength (kg. f)	Egg surface area (cm²)
Age (A)					
26	61.96^{a}	75.92^{c}	0.385^{a}	4.71^{cd}	71.44^{a}
30	63.53^{b}	75.46^{bc}	0.402^{d}	5.07^{e}	72.69^{b}
34	64.92^{bcd}	75.37^{bc}	0.414^{e}	4.77^{d}	73.82^{ce}
38	65.27^{cd}	74.57^{ab}	0.399^{cd}	4.56^{cd}	74.10^{de}
42	65.19^{cd}	74.23^{a}	0.399^{cd}	4.54^{cd}	74.01^{de}
46	63.60^{b}	74.59^{ab}	0.393^{bc}	4.29^{ab}	72.76^{bcd}
50	64.69^{bc}	74.61^{ab}	0.389^{ab}	4.48^{bc}	73.62^{bcd}
54	66.26^{de}	76.02^{c}	0.384^{a}	4.25^{a}	74.87^{ef}
58	67.08^{e}	73.81^{a}	0.389^{ab}	4.13^{a}	75.54^{f}
Laying Time (LT)					
8.30 a.m.	66.14^{c}	75.11	0.396	4.62^{b}	74.79^{c}
12.30 p.m.	64.68^{b}	74.95	0.394	4.54^{ab}	73.60^{b}
4.30 p.m.	63.32^{a}	74.80	0.395	4.44^{a}	72.53^{a}
Cage Tier (CT)					
1	65.13^{b}	75.32^{b}	0.399^{b}	4.62^{b}	73.96^{b}
2	64.09^{a}	74.47^{a}	0.394^{a}	4.54^{ab}	73.16^{a}
3	64.90^{b}	75.06^{ab}	0.393^{a}	4.43^{a}	73.79^{b}
Cage Direction (CD)					
Outward	65.04	75.14	0.392	4.49	73.91
Inward	64.39	74.77	0.398	4.58	73.37
SEM	0.16	0.12	0.00	0.03	0.13
p values					
A	**	**	**	**	**
LT	**	NS	NS	*	**
CT	*	*	*	*	*
CD	*	NS	**	NS	*

Abbreviations: SEM; standard error of mean, NS; non significant. Means within columns with different superscript letter differ significantly (* $p<0.05$; ** $p<0.01$).

Effect of flock age on egg quality traits

There was an increase in egg weight with advancing hen age. Statistically, the lowest (61.96 g) and highest (67.08 g) egg weights were at week 26 and week 58, respectively ($p<0.01$). These results are consistent with some studies that identified a significant increase in egg weight with an increase in the flock age (Sekeroğlu *et al.,* 2014; Samiullah *et al.,* 2017; Yilmaz Dikmen *et al.,* 2017; Kraus *et al.,* 2021; Hammershøj *et al.,* 2021).

It was found that there was a significant effect of hens' age on shape index, which decreased with an increase in hen age ($p<0.01$). A similar result was also observed in other studies (Yilmaz Dikmen *et al.,* 2017; Kraus *et al.,* 2021).

It was determined that shell breaking strength reduced with advancing hen age, and the effect was significant ($p<0.01$). The findings in the present study are in line with the observations of previous authors (Roberts *et al.,* 2013; Şekeroğlu *et al.,* 2014; Yilmaz Dikmen *et al.,* 2017). The present study showed that the highest (0.414 mm) and lowest (0.385 mm) shell thicknesses were at weeks 34 and 26, respectively; and that hen age statistically affected shell thickness ($p<0.01$). This is in agreement with many other studies (Roberts *et al.,* 2013; Şekeroğlu *et al.,* 2014; Yilmaz Dikmen *et al.,* 2017; Samiullah *et al.* 2017; Hammershøj *et al.,* 2021) that reported age related changes in shell thickness. On the contrary, Chung & Lee (2014) found no significant effect of hen age on eggshell thickness in Hy-line laying hens.

Table 2 – Influence of layer age (week), egg-laying time, cage tier, and cage direction on internal egg quality characteristics (mean ± SEM).

	Albumen index (%)	Haugh unit	Yolk index (%)	Yolk color score (DSM)
Age (A, week)				
26	9.11^{e}	81.99^{f}	46.25^{f}	10.34^{a}
30	7.08^{cd}	70.29^{d}	43.01^{de}	11.03^{c}
34	6.30^{b}	66.15^{bc}	42.28^{cd}	10.69^{b}
38	6.18^{b}	63.90^{ab}	43.33^{e}	10.85^{b}
42	6.97^{cd}	68.32^{cd}	43.32^{e}	11.03^{c}
46	5.77^{a}	62.88^{a}	41.18^{ab}	10.37^{a}
50	7.32^{d}	72.90^{e}	41.80^{bc}	10.77^{b}
54	6.73^{c}	70.41^{d}	40.50^{a}	11.28^{d}
58	7.10^{cd}	73.23^{e}	42.66^{de}	11.51^{e}
Laying Time (LT)				
8.30 a.m.	6.80	68.55^{a}	42.60	10.95^{b}
12.30 p.m.	7.01	70.66^{b}	42.75	10.86^{ab}
4.30 p.m.	7.08	71.07^{b}	42.81	10.81^{a}
Cage Tier (CT)				
1	7.07	70.83	43.01	10.94^{b}
2	6.93	69.89	42.59	10.87^{ab}
3	6.89	69.56	42.55	10.81^{a}
Cage Direction (CD)				
Outward	7.30	71.38	43.18	10.88
Inward	6.63	68.82	42.26	10.87
SEM	0.06	0.33	0.10	0.03
p values				
A	**	**	**	**
LT	NS	**	NS	*
CT	NS	NS	NS	NS
CD	**	**	**	NS

Abbreviations: SEM; standard error of mean. NS; non signigificant. Means within columns with different superscript letter differ significantly (* $p<0.05$; ** $p<0.01$).

Significant differences ($p<0.01$) were shown in albumen quality traits, albumen index and Haugh unit. As the age of hens increased, so did the albumen index, which is in agreement with several other studies (Silversides & Scott, 2001; Zita *et al.,* 2009; Roberts *et al.,* 2013; Sekeroğlu *et al.,* 2014; Yilmaz Dikmen *et al.,* 2017). However, Haugh unit decreased with the increase of hen age, which is also in line with previous studies (Roberts *et al.,* 2013; Sekeroglu *et al.,* 2014; Yilmaz Dikmen *et al.,* 2017; Eleroglu, 2019). Contrary to our study, the Haugh unit values increased with an increase in the age of hens in a study by Zita *et al.,* (2009).

The yolk quality can be determined by the yolk index and color score. The age of hens significantly affected yolk index ($p<0.01$) and yolk color score ($p<0.05$). The highest (46.25%) and lowest (40.50%) yolk indexes were at week 26 and 54, respectively. The highest (11.51) and lowest (10.34) yolk color scores were at week 58 and 26, respectively. (Sekeroğlu *et al.* 2014; Yilmaz Dikmen *et al.* 2017; Hammershøj *et al.,* 2021) observed a significant difference in yolk index and color score due to age of hens, which is consistent with the present study.

Besides layer age, various factors including production system, genotype, and housing system conditions can also influence egg quality traits (Petek *et al.,* 2009; Vlčková *et al.,* 2018, 2019; Popova *et al.,* 2020; Yurtseven *et al.,* 2021; Dalle Zotte *et al.,* 2021; da Silva Pires *et al.,* 2021; Tabib *et al.,* 2021).

Effect of egg-laying time on egg quality traits

In this study, there was a decreasing trend in egg weight, shell breaking strength, egg surface area, and yolk color score from morning to afternoon ($p<0.01$; $p<0.05$). In contrast, an increasing trend was observed in Haugh unit from morning to afternoon. Egg-laying time was also found to influence some egg quality traits by other studies. For instance, Tůmová *et al.* (2009) reported that the heaviest eggs were obtained from 6:00 a.m. compared to 10:00 a.m., and 2:00 p.m., and shell-breaking strength differed among different egg-laying times.

Krawczyk *et al.* (2023) observed decreased egg weight in eggs collected at 7:00 a.m. compared to 1:00 p.m. Eleroğlu (2021) found heavier eggs at 3:00 p.m. than at 10:00 and 12:00 a.m. Furthermore, in a study that compared eggs laid at 10:00 a.m., 12:00 and 3:00 p.m., Eleroğlu & Taşdemir (2020) identified a higher shape index at 12:00 a.m., the lowest egg weight and Haugh unit at 3:00 p.m., and higher shell thickness at 3:00 p.m.

Variations among studies in the trends of egg-laying time effects on egg quality might usually be linked to the egg sample size, the season of the year, study area, feeds and feeding, housing environment and system, etc.

Effect of cage tier on egg quality traits

There were significant differences in egg weight, shape index, shell-breaking strength, shell thickness, and egg surface area among cage tiers ($p<0.05$). In agreement with the present study, some authors found a significant effect of cage tier on egg weight (Onbasilar & Aksoy, 2005; Akkus & Yildirim, 2018); shell breaking strength (Yildiz *et al.,* 2006; Akkus & Yildirim, 2018; Eleroglu, 2019) and shell thickness (Onbasilar & Aksoy, 2005; Akkus & Yildirim, 2018), shape index (Tunaydin & Yilmaz Dikmen, 2019), and egg surface area (Akkus & Yildirim, 2018).

In contrast to the present study, several authors reported no significant effect of cage tier on egg weight (Eleroglu, 2019); shape index (Onbasilar & Aksoy, 2005; Yildiz *et al.,* 2006; Sekeroglu *et al.,* 2014; Akkus & Yildirim, 2018; Eleroglu, 2019) and shell breaking strength thickness (Onbasilar & Aksoy, 2005; Sekeroglu *et al.,* 2014; Tunaydin & Yilmaz Dikmen, 2019).

In the current study, cage tier did not have a significant effect on albumen index, Haugh unit, yolk index and yolk color score ($p>0.05$). This would be consistent with some studies that did not find a statistical difference in albumen and yolk quality traits among cage tiers (Onbasılar & Aksoy, 2005; Yıldız *et al.,* 2006; Sahin, 2012; Sekeroglu *et al.,* 2014; Tunaydin & Dikmen, 2019). On the other hand, Eleroglu (2019) identified an influence of cage tier on yolk color score.

Effect of cage direction on egg quality traits

The findings of the present study indicated significant differences between cage directions for egg weight, shell thickness, and egg surface area ($p<0.05$). However, shape index and shell-breaking strength were similar between cage directions ($p>0.05$). Previous studies reported dissimilar egg weights and similar shape index (Yildiz *et al.,* 2006), and similarity in shape index and shell-breaking strength (Sahin, 2012) between cage directions. Both studies would agree with the present findings. In contrast with the current findings, Sahin (2012) identified similar egg weight and shell thickness between cage directions. Also, Yildiz *et al.* (2006) reported that shell-breaking strength was affected by cage position.

In the present study, the difference in albumen index and Haugh unit between the cage directions was significant ($p<0.05$). The albumen index and Haugh Unit were higher in the eggs collected from cages that faced outward (7.30 % and 71.38, respectively) as compared to those that faced inward (6.89 % and 68.82) of the poultry house. Yıldız *et al.* (2006) observed a similar effect, but Sahin (2012) found that the albumen index and Haugh unit were not affected by cage direction, which is not in line with the present study.

The difference between outward and inward cage direction was significant ($p<0.05$) for yolk index and non-significant ($p>0.05$) for yolk color score. The yolk index and yolk color score in the outward and inward cage direction were 43.18% and 42.26%, and 10.88 and 10.87, respectively. Sahin (2012) found a significant effect of cage direction on yolk index, which is in line with our study, but contrary to Yıldız *et al.* (2006). Sahin (2012) and Yildiz *et al.* (2006) reported that cage direction had no effect on yolk color score, which is in accordance with our findings.

The data for the interaction effects between and among layer age, egg-laying time, cage tier, and cage direction on egg external egg quality traits is shown in Table 3.

There were significant interaction effects: age × egg-laying time for egg weight and shape index ($p<0.01$), age × cage tier for shell thickness ($p<0.05$), and age × cage direction × cage tier for shape index ($p<0.01$). It is argued that these effects might be change patterns of egg quality traits that occur due to the oviposition time and changes in the cage environment among cage tiers and between cage directions.

The data for the interaction effects between and among layer age, egg-laying time, cage tier, and cage direction on internal egg quality traits is shown in Table 4.

In the present study, we observed a significant effect of the interactions age × cage direction and cage tier × cage direction on albumen index and Haugh Unit ($p<0.05$; $p<0.01$). Moreover, the interaction of egg-

Table 3 – Results for interaction effects on external egg quality parameters.

	Egg weight (g)	Shape index (%)	Shell thickness (mm)	Shell breaking strength (kg. f)	Egg surface area (cm²)
A × CD	NS	NS	**	NS	NS
CD × LT	NS	NS	NS	NS	NS
CD × CT	NS	NS	NS	NS	NS
A × LT	**	**	NS	NS	NS
A × CT	NS	NS	*	NS	NS
LT × CT	NS	NS	NS	NS	NS
A × CD × CT	NS	**	NS	NS	NS
CD × LT × CT	NS	NS	NS	NS	NS
A × LT × CT	NS	NS	NS	NS	NS
A × LT × CT × CD	NS	NS	NS	NS	NS

Abbreviations: A; age of hen, LT; egg-laying time, CT; cage tier, CD; cage direction, NS; non significant. Significant difference (* $p<0.05$; ** $p<0.01$).

laying time × cage tier was significant for albumen index ($p<0.05$). It is thought that the effect of the above interactions on albumen index and Haugh unit might be associated with variables including age of hens and change in light intensity levels, temperature, and bird's activity in the cage tier and direction. These factors might impact feed consumption, thus influencing albumen height, length, and width, which are the traits used in determining albumen index and Haugh unit.

There was significant effect of the interactions age × cage direction, age × cage tier, and egg-laying time × cage tier on the yolk index ($p<0.01$; $p<0.05$). It is proposed that the above effect of interactions might be associated with the effect of hens' age and the variations in light intensity and temperature in cage tiers. This would impact feed consumption, thus influencing the yolk height and diameter, the traits used in determining yolk index.

Table 4 – Results for interaction effects on internal egg quality traits.

	Albumen index, %	Haugh unit	Yolk index, %	Yolk color score (DSM)
A × CD	**	**	**	NS
CD × LT	NS	*	NS	NS
CD × CT	*	**	NS	NS
A × LT	NS	NS	NS	NS
A × CT	NS	NS	*	NS
LT × CT	*	NS	**	NS
A × CD × CT	NS	NS	NS	NS
CD × LT × CT	NS	NS	NS	NS
A × LT × CT	NS	NS	NS	NS
A × LT × CT × CD	NS	NS	NS	NS

Abbreviations: A; age of hen, LT; egg-laying time, CT; cage tier, CD; cage direction, NS; non significant. Significant difference (* $p<0.05$; ** $p<0.01$).

CONCLUSIONS

The present study determined age-related variations in egg quality traits. Egg weight, shell-breaking strength, egg surface area, and yolk color score decreased, but Haugh Unit increased as egg-laying time moved from morning to afternoon. Cage tier resulted in variability in the external egg quality traits, with no effect on internal egg quality traits. Nevertheless, eggs collected from cages facing outward of the poultry house were superior in terms of egg weight, egg surface area, albumen index, Haugh unit, and yolk index. Eggs collected from cages facing inward of the poultry house were superior for shell-breaking strength. Generally, further studies are needed to refine the role of cage tier and cage direction on biological functions or physiological responses that might be associated with impact on some egg quality traits.

ACKNOWLEDGMENTS

The authors acknowledge the Niğde Ömer Halisdemir University, Ayhan Şahenk Agricultural Application and Research Center for its support during the study.

FUNDING

No funding was received for conducting this study.

CONFLICT OF INTEREST

The authors have no competing interests that are relevant to the content of this article to declare.

AUTHORS CONTRIBUTION

AŞ: Conceptualization, methodology, resources, and data analysis; YEŞ: Investigation; BT: writing – original and final draft. MD: Investigation; AA: Investigation. All authors participated in reading the final draft and approved its publication.

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