

## Correlation between Internal and External Egg Quality Indicators in the Early Phase of Hy-Line Brown Laying Hens

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**ABSTRACT** This study investigated correlations between egg quality indicators to identify external egg quality traits to predict internal egg quality using non-destructive and convenient measurements. Thirteen indicators, including Haugh unit, albumen height, eggshell breaking strength, eggshell thickness, eggshell color (CIE L<sup>\*</sup>, CIE a<sup>\*</sup>, CIE b<sup>\*</sup>), and reflectivity value, egg weight, egg length, egg width, shape index, and yolk color, were investigated. A total of 180 brown eggs were obtained from one 27-week-old flock of Hy-line brown-laying hens raised in a cage system. Correlations were evaluated using Pearson's correlation coefficient (r). The results showed strong correlations between Haugh unit and albumen height, eggshell color CIE L<sup>\*</sup> and reflectivity, egg weight and width, egg weight and length, eggshell color CIE L<sup>\*</sup> and CIE a<sup>\*</sup>, eggshell color CIE a<sup>\*</sup> and reflectivity, and shape index and egg length (P<0.001). Moderate correlations were observed between eggshell breaking strength and eggshell breaking strength (P<0.01), and eggshell breaking strength (P<0.01), and eggshell breaking strength (P<0.001). The present study showed significant correlations between eggshell color CIE a<sup>\*</sup> was correlated with Haugh unit, albumen height (P<0.01), and eggshell breaking strength (P<0.001). The present study showed significant correlations between eggshell color and other quality indicators. Thus, this study suggests that eggshell colors based on reflectiveness and the CIE L<sup>\*</sup>a<sup>\*</sup>b<sup>\*</sup> value can be used to estimate the Haugh unit, albumen height, eggshell breaking strength unit, albumen height, eggshell breaking strength unit, albumen height, eggshell color stage on reflectiveness.

(Key words: haugh unit, egg quality, eggshell color, correlation coefficient, laying hens)

## INTRODUCTION

Eggs have been considered a cost-efficient and nutrient-dense animal food in the human diet (Papanikolaou and Fulgoni, 2020). Global egg production has constantly increased over the last decades, and its production exceeded 1,577 billion eggs in 2019 (FAO, 2019). In the Republic of Korea, the egg production per year was over 14.6 billion, and the egg consumption per capita was 282 eggs in 2019 (MAFRA, 2020). Moreover, the consumer awareness of the quality and freshness of eggs has increased with industrial growth.

Consumers' acceptability and preference have determined egg quality for several characteristics: freshness, cleanliness, color, egg weight, eggshell quality, yolk index, albumen index, Haugh unit, and chemical composition (Narushin, 1997; Genchev, 2012; Zaheer, 2015). According to Korea Institute for Animal Products Quality Evaluation (KAPE), domestic eggs are graded based on their quality into 1+, 1, and 2 through an overall evaluation of exterior, candling, and broken-out evaluations. The exterior evaluation judges

eggshell features such as shape, cleanliness, and texture. Candling is a process to hold a light on the eggshell to observe the egg's interior, such as air-cell and eggshell cracks. The broken-out egg evaluation confirms Haugh unit, albumen height, yolk color, blood spots, and meat spots in eggs. However, it is a destructive, time-consuming, laborinefficient measurement and causes a lot of egg waste after the assessment. Also, several non-destructive egg quality measurement technologies using the electronic nose, Terahertz waves, spectroscopy have been reported by Yimenu et al. (2017), Khaliduzzaman et al. (2020), and Loffredi et al. (2021), respectively. However, these technologies have not been commercially available. So far, earlier studies attempted to investigate correlations between egg quality indicators to predict internal egg quality as non-destructive methods with local Chinese chickens and Japanese quails for genetic selection (Yang et al., 2009; Alkan et al., 2010; Rathert et al., 2011).

The current study investigated correlations between internal and external egg quality indicators in commercially

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available Hy-line Brown laying hens to explore indirect egg quality criteria that could predict egg quality without breaking eggs.

## MATERIALS AND METHODS

#### 1. Eggs and Data Collection

A total of 180 eggs were collected from 27-week-old Hy-Line Brown laying hens raised in the cage system of the experimental poultry house of Konkuk University on the same day in the morning. Collected eggs were transported to the laboratory for analysis, and each egg was numbered on the eggshell for identification. The abnormal or cracked eggs (n=8) were eliminated from the analysis. Thus, a total of 172 eggs were analyzed for this study.

## 2. Egg Length, Egg Width, and Shape Index Measurement

The egg length (L) and width (W) of eggs were measured using a Vernier caliper. The egg shape index (SI) was calculated by the following equation (Anderson et al., 2004):

 $SI = (W / L) \times 100$ 

#### 3. Eggshell Color Measurement

The eggshell color was measured based on a Commission Internationale de l'Eclairage (CIE)  $L^*a^*b^*$  color using a portable spectrophotometer (CM-2600d, Konica Minolta, Ramsey, NJ, USA).  $L^*$  represents the lightness of the color (0 for black and 100 for white),  $a^*$  indicates between green and red ( $a^*<0$  for green and  $a^*>0$  for red), and  $b^*$  quantifies the between blue and yellow ( $b^*<0$  for blue and  $b^*>0$  for yellow).

In addition, the eggshell color was measured using a shell color reflectometer (TSS QCR, Technical Services and Supplies, York, UK). The shell color reflectometer was taken as a percentage reading to 0% and 81.8% using the black and white standard board, respectively. The reflectivity of commercial brown eggs expresses 25 to 40%, according to the manufacturer of the reflectometer.

#### 4. Egg Weight Measurement and the Broken-Out Evaluation

Internal egg quality indicators can be measured after breaking eggs. Thus, the broken-out evaluation is required to grade and evaluate egg quality. First, the egg weight was measured before the breaking-out evaluation by a digital egg tester (DET-6000, Nabel, Kyoto, Japan). This tester also measured eggshell breaking strength, eggshell thickness, yolk color score based on YolkFan<sup>TM</sup>, albumen height, and Haugh unit. The Haugh unit (HU) is related to albumen height (H) and egg weight (W) (Haugh, 1937) and is calculated automatically by the following formula.

 $HU = 100 \times \log (H - 1.7W0.37 + 7.6)$ 

#### 5. Statistical Analysis

The obtained data were analyzed using R package (Library) of R Studio (ver.1.4.1717). The correlation is evaluated based on the Pearson's correlation coefficient (r) and their significance at the 5% level (P<0.05).

#### RESULTS

The statistics of internal egg quality indicators and external egg quality indicators are shown in Table 1. The Pearson's correlation coefficients (r) between egg quality indicators were presented in Table 2.

1. Correlation between the Haugh Unit and Other Indicators

A very strong positive correlation was found between the Haugh unit and albumen height (r=0.975, P<0.001). Also, the Haugh unit showed a weak positive correlation with eggshell color CIE a<sup>\*</sup> value (r=0.217, P<0.01). However, there were no distinct correlations between the Haugh unit and the rest of the egg quality indicators.

 Correlation between the Albumen Height and Other Indicators

The albumen height showed weak positive correlations with the egg weight (r=0.263, P<0.001), egg width (r=0.249, P<0.001) and the eggshell color CIE a<sup>\*</sup> (r=0.202, P<0.01),

Item Ν Min Max SD CV (%) Mean Internal quality 172 6.80 Haugh unit 63.50 98.80 84.89 5.77 172 4.30 9.80 13.32 Albumen height (mm) 7.18 0.96 Egg yolk color 172 6.00 9.10 7.77 0.64 8.20 External quality 6.93 Egg weight (g) 172 50.90 73.20 58.50 4.05 Eggshell breaking strength (kgf/cm<sup>2</sup>) 172 1.37 0.98 17.28 7.67 5.64 Eggshell thickness (mm) 172 0.30 0.51 0.40 0.04 8.80 Egg length (mm) 172 50.00 63.00 54.48 1.83 3.37 Egg width (mm) 172 41.00 47.00 43.48 1.15 2.65 Shape index (%) 172 68.25 90.38 79.86 2.66 3.33 Eggshell color CIE L<sup>\*</sup> (lightness) 172 48.29 66.36 54.85 3.27 5.96 CIE a\* (redness) 172 12.72 23.42 19.80 1.73 8.76 CIE b\* (yellowness) 172 13.39 34.04 28.57 2.95 10.34 Reflectivity (%) 172 17.30 37.50 23.49 3.49 14.87

Table 1. The descriptive statistics of internal and external egg quality indicators<sup>1</sup>

<sup>1</sup> N, sample size; Min, minimum; Max, maximum; SD, standard deviation; CV, coefficient of variation.

Table 2. The correlation coefficient between egg quality indicators<sup>1-3</sup>

	Haugh unit	Albumen height	Eggshell breaking strength	Eggshell thickness	CIE L <sup>*</sup>	CIE a <sup>*</sup>	CIE b <sup>*</sup>	Reflectivity	Egg weight	Egg length	Egg width	Shape index	Yolk color
Haugh unit	1.000												
Albumen height	0.975***	1.000											
Eggshell breaking strength	0.102	0.098	1.000										
Eggshell thickness	- 0.011	0.032	0.417***	1.000									
CIE L*	- 0.138	- 0.125	- 0.236**	- 0.192*	1.000								
CIE a*	0.217**	0.202**	0.256***	0.024	- 0.726***	1.000							
CIE b*	0.149	0.133	- 0.034	- 0.225**	- 0.086	$0.590^{***}$	1.000						
Reflectivity	- 0.117	- 0.105	- 0.249**	- 0.222**	0.973***	-0.681***	- 0.001	1.000					
Egg weight	0.064	0.263***	- 0.032	0.144	0.016	- 0.049	- 0.080	0.001	1.000				
Egg length	- 0.060	0.083	- 0.056	0.119	0.044	- 0.042	- 0.056	0.029	0.703***	1.000			
Egg width	0.090	0.249***	- 0.084	0.024	- 0.001	- 0.007	- 0.053	- 0.004	0.814***	0.392***	1.000		
Shape index	0.127	0.109	- 0.011	- 0.102	- 0.039	0.030	0.008	- 0.028	- 0.066	- 0.681***	0.405***	1.000	
Yolk color	0.040	0.079	- 0.050	0.003	- 0.034	- 0.049	- 0.096	- 0.032	0.158	0.026	0.176	0.112	1.000

<sup>1</sup> \**P*<0.05, \*\*\**P*<0.01, \*\*\*\**P*<0.001.

<sup>2</sup> CIE, Commission Internationale de l'Eclairage.

<sup>3</sup> L<sup>\*</sup>, eggshell color lightness; a<sup>\*</sup>, eggshell color redness; b<sup>\*</sup>, eggshell color yellowness.

whereas no distinct correlations with the rest of the egg quality indicators.

## Correlation between the Eggshell Breaking Strength and Other Indicators

The eggshell breaking strength showed a moderate positive correlation with the eggshell thickness (r=0.417, P<0.001) and weak correlations with the eggshell color CIE L<sup>\*</sup> value (r=-0.236, P<0.01), CIE a<sup>\*</sup> value (r=0.256, P<0.001), and the eggshell color reflectivity (r=0.249, P<0.01), while no distinct correlations were found with all other egg quality indicators.

# 4. Correlation between the Eggshell Thickness and Other Indicators

The correlation coefficient (*r*) between the eggshell thickness and the eggshell breaking strength was 0.417 (P<0.001). In addition, the eggshell thickness indicated negative weak correlations with the eggshell color CIE b<sup>\*</sup> value (r=-0.225, P<0.01) and the eggshell color reflectivity value (r=-0.222, P<0.01).

5. Correlation between the Eggshell Color and Other Indicators

The eggshell color CIE L<sup>\*</sup> value appeared to have a very strong correlation with the eggshell color reflectivity value (r=0.973, P<0.001) and a strong negative correlation with the eggshell color CIE a<sup>\*</sup> value (r=-0.726, P<0.001). The eggshell color CIE a<sup>\*</sup> value showed strong negative correlations with the eggshell color reflectivity value (r=-0.681, P<0.001) and a moderate positive correlation with the eggshell color CIE b<sup>\*</sup> value (r=0.590, P<0.001).

## Correlation between the Egg Weight, Length, Width, and Shape Index and Other Indicators

The egg weight showed strong positive correlations with the egg length (r=0.703, P<0.001) and the egg width (r=0.814, P<0.001). The egg length appeared to have a strong negative correlation with the shape index (r=-0.681, P<0.001) and a moderate positive correlation with the egg width (r=0.392, P<0.001). A moderate positive correlation showed between the egg width and the shape index (r=0.405, P<0.001).

## Correlation between the Yolk Color and the Other Indicators

This study found no significant correlations between the yolk color and other egg quality indicators.

#### DISCUSSION

 Correlation between the Haugh Unit and the Albumen Height

The Haugh unit (Haugh, 1937) is a representative egg quality and freshness indicator and the measure of corrected albumen quality for egg weight. The present study showed that the Haugh unit has a strong positive correlation with the albumen height while no correlation with the egg weight. This finding is consistent with the results of several previous studies (Zhang et al., 2005; Sinha et al., 2018; Rafea, 2019). However, results by Kul and Seker (2004), Olawumi and Ogunlade (2008), Şekeroğlu and Altuntas (2009), Aygun and Yetisir (2010), and Aygun (2014) found no significant correlation between Haugh unit and egg weight. Therefore, the results showed that the Haugh unit was dependent on albumen height but independent of egg weight.

## 2. Correlation between the Eggshell Breaking Strength and the Eggshell Thickness

The eggshell breaking strength and thickness are crucial egg quality indicators to consider to reduce breakages during transportation and distributions stages in the commercial egg industry. Hamilton (1982) presented that eggshell thickness is an indirect trait to determine its strength. The result of this study is associated with earlier studies that reported significant positive correlations between eggshell breaking strength and eggshell thickness (Yan et al., 2014; Kibala, 2018). Moreover, this result is consistent with Bain (2005), who has indicated that thicker eggshells are not necessarily the same as stronger eggshells because the strength of eggshells is related to mineral components of the eggshell.

# 3. Correlation between the Eggshell Color and the Other Indicators

The eggshell color is considered a critical factor for customers' preference (Lee et al., 2003). The eggshell color reflectivity and CIE  $L^*a^*b^*$  values are the most typical measurement methods of eggshell color intensity. The current study results are similar to earlier studies that reported that the eggshell color CIE  $L^*$  (lightness) value could indicate whether the eggshell color was dark or light (Odabasi et al., 2007; Aygun, 2013; Aygun, 2014). The strong positive correlation between the eggshell color CIE  $a^*$  (redness) and CIE  $b^*$  (yellowness) value is similar to results reported by Odabasi et al. (2007) and Aygun (2014). To summarize, these results indicate that darker brown eggs represent the lower value of CIE  $L^*$  (lightness) and reflectivity while representing the higher value of CIE  $a^*$  (redness).

The finding of this study demonstrates no correlation between the eggshell color CIE L\* value and Haugh unit, whereas Duman et al. (2016) found a positive correlation. On the other hand, the results of the present study are in line with Aygun (2014), who reported that eggshell color CIE a\* value was correlated with Haugh unit, the eggshell color CIE L\* value showed a negative correlation with eggshell strength, while the CIE a\* value showed a positive correlation with the eggshell strength. The result of a weak significant correlation between the eggshell color reflectivity and eggshell breaking strength is supported by the finding of Yang et al. (2009). However, the result of the present study disagrees with the discovery of Zita et al. (2009), who found no significant correlation between the eggshell color reflectivity value and eggshell strength. Interestingly, this study found that the eggshell color CIE a\* value was correlated with albumen height and eggshell color CIE b\* value, and the eggshell color reflectivity value appeared weak negative correlations with eggshell thickness.

## Correlation between the Egg Weight, Egg Width, and Shape Index and the Other Indicators

The present study results indicate that the egg weight and egg width show weak correlations with the albumen height. These findings support the report of Silversides and Villeneuve (1994), who observed a weak relation between the egg weight and the albumen height. Duman et al. (2016) observed a significant negative correlation between egg weight and shape index and a significant positive correlation between shape index and Haugh unit. In contrast, the present study results found a statistically nonsignificant correlation. Therefore, these outcomes suggest that external eggshell quality traits that had been analyzed in this study might not predict internal egg quality indicators. Nonetheless, the latter statement does not indicate the absence of non-invasive methods predicting internal egg qualities, including the Haugh unit. Future studies are needed to develop simple, inexpensive, high-throughput, and non-invasive methods that can easily detect internal egg quality.

### CONCLUSION

In conclusion, the present study reveals that eggshell color values are positively correlated with other egg quality traits. The eggshell color CIE  $L^*$  and reflectivity value could indicate the darkness or lightness of the eggshell color. The brownness of the eggshell could be more accurate when using the CIE  $a^*$  value and CIE  $b^*$  value than using the CIE  $L^*$  or the reflectivity value only. Moreover, the eggshell color CIE  $a^*$  value might be a criterion to predict Haugh unit, albumen height, eggshell breaking strength, and eggshell thickness.

Several authors demonstrated that hen's ages (Lee et al., 2013; Samiullah et al., 2017; Kim et al., 2018), strains (Liu et al., 2010), egg storage duration, and temperature (Lee et al., 2014; Lee et al., 2016), and housing system (Wang et al., 2009; Şekeroğlu et al., 2010; Samiullah et al., 2017) could affect Haugh unit, albumen height, egg weight, and eggshell color. However, it must be remembered that the present study analyzed freshly-laid eggs from one flock of the same strain and same-age at the same housing. Therefore, accumulating and exploring more correlation data of egg quality from different breeds and aged laying hens might support the present study.

#### 적 요

계란의 내부 및 외부의 품질을 나타내는 요소들 사이의 상관 관계를 분석하여, 할란 검사를 통하지 않고도 계란 품 질을 예측하는데 적합한 지표가 있는지 알아보기 위한 실험 을 수행하였다. 케이지 계사에서 사육 중인 27주령의 하이 라인 브라운 산란계 동일 계군이 같은 날에 산란한 180개의 계란을 수집하였다. 분석 항목으로는 호우유닛, 농후난백높 이, 파각 강도, 난각 두께, 난각 색(CIE L<sup>\*</sup>, CIE a<sup>\*</sup>, CIE b<sup>\*</sup>, 반사율), 계란의 중량, 계란의 장경, 계란의 폭, 난형 지수 및 난황색 등 총 13가지의 지표를 조사하였다. 피어슨 상관 계 수(r)를 이용하여, 각 품질 지표 사이의 선형 관계성을 평가 하였다. 호우유닛과 농후난백높이, 난각색 CIE L\*값과 반사 율, 그리고 계란 중량과 계란의 폭 사이에서 매우 강한 정의 상관 관계가 분석되었다(r=0.975, r=0.973, r=0.814, P<0.001). 계란의 중량과 계란의 장경, 난각색 CIE L\*값과 난각색 CIE a\*값, 난각색 CIE a\*값와 반사율, 그리고 난형 지수와 계란의 장경 사이에는 높은 상관 관계가 나타났다(r=0.703, r=-0.726, r=-0.681, r=-0.681, P<0.001). 난각의 파각 강도와 난각의 두께, 난각색 CIE a\*값과 난각색 CIE b\*, 난형 지수 와 계란의 폭 사이에는 중간 정도의 상관 관계가 나타났다(r = 0.417, r = 0.590, r = 0.405, P<0.001). 난각색 CIE a\*값은 호우유닛, 농후난백높이와 약한 정의 상관 관계가 있었으며 (r=0.217, r=0.202, P<0.01), 난각색 CIE L\*값과 파각 강도 사이에는 약한 부의 상관 관계가 나타났다(r=-0.236, P<0.01). 난각색 CIE b<sup>\*</sup>값은 난각 두께와 약한 부의 상관 관 계를 보였고, 난각색 반사율은 파각 강도 및 난각 두께와 약 한 부의 상관 관계에 있는 것으로 나타났다(r=-0.225, r=-0.249, r=-0.222, P<0.01). 그 외 나머지 품질 지표들 간의 상관성은 통계적으로 유의하지 않거나 관계가 없는 것으로 나타났다. 갈색란의 난각색은 반사율만으로 표현하는 것 보 다, CIE L\*값, CIE a\*값과 CIE b\*값을 함께 적용했을 때 갈 색의 정도를 더 구체적으로 표현한다. 결론적으로 본 연구 를 바탕으로 갈색란의 난각색 CIE L<sup>\*</sup>a<sup>\*</sup>b<sup>\*</sup>값은 계란의 품질 지표인 호우유닛, 농후난백높이, 난각의 파각 강도와 두께를 간접적으로 예측하는데 사용할 수 있을 것으로 판단되었다. (색인어: 호우유닛, 계란 품질, 난각색, 상관 계수, 산란계)

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