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PHYSICAL EVALUATION OF THE EXTERNAL AND INTERNAL EGG QUALITY OF WHITE LEGHORN HENS (GALLUS GALLUS DOMESTICUS) FED WITH A LIQUID ADDITIVE OF APPLE YEAST

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SUMMARY
The use of probiotics in animal feed offers the potential to maintain the growth of animals fed antibiotic-free diets or under conditions of high stress. This study evaluated the physicochemical quality (internal and external indicators) of eggs from hens (Gallus gallus domesticus) of the White Leghorn breed fed with a liquid additive of yeasts (*S. cerevisiae* and *Kluyveromyces lactis*) as probiotics. In relation to the external indicators, a difference was observed in the pH of the white, being 8.98±0.22 for the egg of hens fed with the probiotic and 8.69±0.83 for the egg of the control treatment. Regarding the external indicators, eggs from hens fed with probiotics had a higher weight compared to control eggs. Regarding color, eggs from hens fed with probiotics had higher luminosity and lower values of a* ($P<0.05$). On the other hand, the statistical correlations between egg quality indicators suggest the possibility of implementing programs for the use of the liquid additive of apple yeast in poultry feed, to improve some egg quality indicators, such as egg weight.

INTRODUCTION

Modern poultry farming consists of the rational and methodical exploitation of poultry birds, to obtain from them two main products: eggs and meat. Eggs are one of the most complete foods of animal origin that exist. However, establishing the specifications and quality control of this for human consumption is not an easy problem to solve, since it involves numerous factors (Juárez-Caratachea *et al.*, 2010). A major problem in laying hens, as the age of production advances, is the reduction in shell quality, which depends, among other factors, on the level of calcium in the diet (Hernández *et al.*, 2006). Egg quality can be objectively defined as the sum of all sensory (taste, texture, color, etc.), nutritional (nutrient content and chemical status), safety (pathogens), and those related to egg processing (Schwägele, 2011). It is generally accepted that all characteristics of egg quality have a genetic basis. The most important quality criterion is the weight of the egg. The optimal weight required is between 53 and 73 g for medium and large eggs, respectively. The factors that influence the weight of the resulting egg are genetics, health conditions, and the diet of the hens (Schwägele, 2011; Ortiz & Mallo, 2019).
The main method of evaluating the quality of chicken eggs is the Haugh units, this is an empirical method that determines a relationship between the weight and height of the coarse albumin. When eggs age, coarse albumin breaks down into thin albumin. This decomposition results in a decrease in the height of the gross albumin, which is the main factor in the HU equation. A higher HU value (i.e., coarse albumin height) indicates a higher quality egg. An AA-quality egg has a HU greater than 72; an A-quality egg has a HU of 60 to 72; and a B-quality egg has a HU of 31 to 60. The most expensive item, in poultry production, is that corresponding to food. Diets must be nutritionally adequate, with an optimal cost and reduce the elimination of nitrogen from the

environment (Fuente-Martínez *et al.*, 2012). The primary objective of production is then the delivery of safe food for human consumption, taking into account animal welfare and respect for the environment (Gaggia *et al.*, 2010).

Given the growing awareness of consumers about the use of chemical substances that are likely to leave residues in the animal body, the possibility that some ingredients of natural origin such as probiotics will be directed in the future to the priority choice between the different feed efficiency enhancing additives is increasing (Ochoa, 2017). Various studies have emphasized the ability of probiotics to stimulate appetite, improve nutrient absorption, strengthen the host's immune system, improve its metabolism, health, and production (Martínez *et al.*, 2012; Díaz-Plascencia *et al.*, 2017; Díaz-Plascencia *et al.*, 2022). The application of probiotics in poultry feed is strictly associated with the concept of competitive exclusion (EC). In addition, they increase egg production and quality. (Gaggia *et al.*, 2010). Probiotic modes of action include: regulation of gut microbial homeostasis, stabilization of gastrointestinal barrier function, bacteriocin expression, enzyme activity inducing absorption and nutrition, immunomodulatory effects, inhibition of procarcinogenic enzymes, and interference with pathogens' ability to colonize and infect mucosa (Gaggia *et al.*, 2010). Probiotics include yeasts that induce positive effects in terms of productive performance in monogastric species, but cannot colonize the digestive tract (Gallegos, 2015; Díaz-Plascencia *et al.*, 2022). Therefore, probiotics offer the potential to maintain the growth of animals fed antibiotic-free diets or under high-stress conditions (Ruíz, 2016; Díaz-Plascencia *et al.*, 2017; Ruíz-Holguín *et al.*, 2021). The use of *S. cerevisiae* and *Kluyveromyces lactis* as probiotics, it helps to increase the biological value of nitrogenous compounds in the digestive tract and reduces stressors in animals (Özsoy *et al.*, 2018). Due to the aforementioned background in which the nutritional and functional importance of eggs is highlighted, the objective of this study was to evaluate the effect of the addition of a liquid yeast additive in the diet of laying hens on the internal and external physicochemical characteristics of the egg.

MATERIALS AND METHODS

Localization

The experiment was carried out in the period from January to July at the bird metabolic unit of the Faculty of Zootechnics and Ecology (FZyE) of the Autonomous University of Chihuahua (UACH), located near the city of Chihuahua, Mexico. The unit is located between parallels 28° 38' LN and 106° 04' LO, at an altitude of 1,440 meters above sea level. The average annual temperature is 18.6 °C with an average annual rainfall between 200 and 600 mm (INEGI 2017).

Ethical aspects

The study was carried out in accordance with the internal code of bioethics and taking into account the animal welfare regulations of the FZyE of the UACH as well as the regulations of the Official Mexican Standard on animal care published by the Ministry of Agriculture and Rural Development (SAGARPA 1999).

Obtaining the Liquid Yeast Additive (ALL)

The ALL contained a mixture of the yeasts *Kluyveromyces lactis, Issatchenkia orientalis* and *Saccharomyces cerevisiae* that was obtained from the fermentation of apple bagasse with a concentration of 1.8 x109 CFU mL−1 of the Lebas® brand

Experiment design

The experiment was conducted with 1,254 hens divided into 627 animals for each treatment. The birds were 22 weeks old at the beginning of the experiment and the duration of the experiment was 24 weeks. One treatment consisted of adding to the diet a liquid additive of *Kluyveromyces lactis*, *Issatchenkia orientalis* and *Saccharomyces cerevisae* yeasts obtained from the solid state fermentation of apple bagasse (Díaz-Plascencia, *et. al.*, 2017) and the other treatment was represented as the control treatment (feed without additive). The feed used was balanced poultry feed prepared by Harinas de Chihuahua, S.A. de C.V., which contained the following ingredients:

ground grains, oilseed pastes and/or pork meal, grain by-products, vegetable oil, salt, sodium bentonite, alfalfa; vitamins A, D3, E, K3, B1, B2, niacin, pantothenic acid, B6, folic acid, biotin, B12, and choline; minerals such as: copper, iron, manganese, iodine, zinc, selenium, calcium, calcium orthophosphate; amino acids such as: L-lysine, DL-methionine, antioxidant (B.H.T), at the preventive level mycotoxin sequestering and antifungal. The feed had 17% minimum protein, 2% minimum fat, 5% maximum fiber, 8% maximum ash, 12% maximum moisture and 56% E.L.N (p. diff.). The liquid yeast additive was added at the rate of 1 L / 70 kg of feed which was served directly at the feeders every day at 07:00 am.

10 eggs were collected from hens fed the additive (treatment eggs) and 10 without it (control eggs) randomly, for each treatment, every 4 weeks during the 24 weeks of the experiment. The collected eggs were transported in egg carriers with a capacity of 12 eggs each, which were transported to the Laboratory of Technologies of Products of Animal Origin of the Faculty of Zootechnics and Ecology of the Autonomous University of Chihuahua to carry out the corresponding measurements.

External measurements

External measurements were: complete egg weight (g); weight (g) of egg without the shell and weight of the shell, for this purpose a digital scale with an accuracy of 0.1 g (Escali brand, model L600, China) was used. The thickness of the shell (mm) was measured with a handheld micrometer (Mitutoyo No. 7301, Japan). The polar or longitudinal diameter and the equatorial or short diameter (length and width of the egg) were measured with a Vernier (Scala brand, Mexico). The percentage of shell was estimated through the following equation:

% Husk = (shell weight / whole egg weight) x 100

The shape index was obtained by the formula:

IF= (equatorial diameter or smaller / polar diameter or larger) x 100.

Internal Measurements

Internal measurements included: yolk pigmentation, which was determined with the Roche fan (Ovocolor BASF D-6700, Germany). The diameter of the yolk, which was measured at four points in a cross with millimeter paper under flat transparent glass. Height of the white and yolk that were measured with the Vernier depth bar. Yolk index, which was determined with the following equation:

IY= yolk height / average yolk diameter x 100

Likewise, the yolk weight was evaluated using a digital scale with an accuracy of 0.1 g (Escali, model L600, China). The pH of the egg white and yolk was obtained with a potentiometer (compact pH METER B-213, USA). The Haugh units were determined through the equation:

$$UH = 100 \times \log10 (H - 1.7 W^{0.37} + 7.6)$$

Where: H represents the height of the white in mm and W represents the weight of the egg (Monira, et. al., 2003).

Likewise, the color of the yolk was measured, obtaining the luminosity values (L*), a* and b*. Likewise, the color difference (ΔE) was determined through the following formula:

$$\Delta E = \sqrt{(Lt - Lc)2 + (at - ac)2 + (bt - bc)2}$$

Where:
Lc, ac and bc = values of the control treatment
Lt, at and bt = probiotic treatment values

The color change (ΔE) is a calculated parameter (Equation 2) with the results of the parameters L*, a* and b*. Values below 2.7 are not perceptible to the human eye (0 – 0.7 not at all; 0.7 – 2.5 slightly; 2.5 – 3.0 remarkable; 3.0 – 6.0 appreciable; 6.0 – 12.0 considerable and 12.0 is larger) (Ramírez-Navas & de Stouvenel, 2012).

RESULTS AND DISCUSSION

External measurements

In relation to the external measurements, only significant differences (P < 0.05) were found in the weight of the egg with the shell and the weight and percentage of the shell.

Weight of the shelled egg: Schwägele (2011) mentions that egg weight is the most important quality criterion. The optimal weight required for medium and large eggs is between 53 and 73 g, respectively. The highest mean weight of the shelled egg was observed in the treatment eggs, with a weight of 57.04 ± 4.25 g, while the average of the control eggs was 56.68 ± 5.46 g (P < 0.05). The weight of the eggs belonged to the size of medium eggs. Schwägele, (2011) indicates that thefactors that influence the weight of the resulting egg are the genetics, age, health and diet of the hens as well as the environment and the time of year.

Polar diameter: in relation to this parameter, no differences (P > 0.05) were observed in the average polar diameter between control eggs (5.64 ± 0.19 cm) and treatment eggs (5.62 ± 0.23 cm). Juárez-Caratachea *et al.* (2011) point out that the polar diameter is 25% larger than the transverse diameter at most, which does not coincide with what was obtained in this study. It also indicates that, in hens, eggs with a larger longitudinal diameter (polar) are more prone to breakage as they adapt less to handling and do not slide as smoothly as round eggs, which produces cracks that detract from the final quality of the egg. Larger egg size may also be a factor influencing shell quality traits (Monira *et al.*, 2003; Ojeda *et al.*, 2022).

Transverse diameter: The largest average cross-section diameter was observed in control eggs with a measurement of 4.25 ± 0.11 cm, while treatment eggs were 4.24 ± 0.12 cm. Juárez-Caratachea *et al.* (2011) mention that the degree of variation in longitudinal (polar) diameter compared to transverse diameter suggests that egg weight may be more influenced by egg length than egg width.

Shell thickness: In this parameter, no significant differences were found (P > 0.05) between treatments, being 0.358 ± 0.037 mm for the treatment egg; and 0.358 ± 0.034 mm for the control egg. Díaz-Plascencia *et al.* (2022), mention that with the use of probiotics, a thickening in the eggshell is quickly verified, due to the increase in calcification of the best nourished animal. In this study, no differences were detected in this parameter, which could be due to the length of time the study lasted.

Weight and percentage of the shell: in relation to these two parameters, significant differences were detected (P < 0.05), with the treatment egg obtaining the highest weights of 0.35 ± 0.03 for the weight of the shell and 12.40 ± 1.01 for the percentage of the same.

Internal parameters

In relation to the internal parameters, only significant differences (P < 0.05) were found for the pH of the egg white and the mean diameter of the white one.

Egg white weight: The highest average white weight, with no significant difference (P > 0.05), was found in treatment eggs with a weight of 34.95 ± 3.24 g, compared to control eggs that was 34.41 ± 3.49 g. According to Juárez-Caratachea *et al.* (2011) The weight of chicken egg white represents between 55 and 60 % of the total egg weight, a percentage that is also represented in the data obtained in this study.

Yolk weight: No significant differences were detected in this parameter between treatment and control eggs (P > 0.05). Treatment eggs had an average of 14.29 ± 1.06 g and control eggs had an average of 14.05 ± 0.94 g. Juárez-Caratachea *et al.*, (2011) point out that the yolk of the chicken egg represents 30% of the weight of the complete egg, a fact that does not coincide with what was found in the present study since both the control and treatment eggs the yolk represented 25% of the weight of the complete egg.

Dense white height: Control and treatment eggs did not present significant differences (P > 0.05), being 9.57 ± 1.77 mm for control eggs and 9.25 ± 1.08 mm for treatment eggs. Juárez-Caratachea *et al.* (2011) said that the height of the white is one of the most used variables to determine the internal quality of the egg, expressed as Haugh Units. The same source mentions that the freshness of the egg is measured through the height of the white, that is, whites with higher height suggest fresher eggs, as are found in Control eggs.

Haugh units (UH): In this parameter no significant differences were found between treatments (P > 0.05). Being 99.84 ± 6.84 for control eggs and 98.27 ± 4.64 for treatment eggs. Schwägele, (2011) indicates that relatively high Haugh Units (> 75), is indicative that the egg is fresh. The same source mentions that a higher UH value (i.e., coarse albumin height) is related to a higher quality egg. An AA quality egg has a UH value greater than 72; an A quality egg has UH values between 60 and 72; and a quality B egg has UH values between 31 and 60. In addition to quality, the functionality of the egg is important. So, in both cases, the eggs were found in category AA.

Egg white index and yolk index: in these two parameters, no significant differences were detected (P > 0.05) between control and treatment eggs. Juárez-Caratachea *et al.*, (2011) mention that the white index is considered one of the most important indicators related to egg quality, since it affects the yolk index.

Percentage of white: no significant differences were detected (P > 0.05) for this parameter, with the percentage being 61.23 ± 2.30 % for treatment eggs and 61.10 ± 7.53 % for control eggs.

Clear pH: in relation to this parameter, treatment eggs had a higher pH (P < 0.05) of the white (8.98 ± 0.22) compared to treatment eggs with a result of (8.98 ± 0.22), while control eggs (8.69 ± 0.83).

Mean diameter of the white: The highest mean (P < 0.05) of mean white diameter corresponded to the control eggs with 12.76 ± 1.26 cm compared to the treatment eggs obtained 11.89 ± 1.00.

Color: Color parameters are shown in Table 5.The treatment eggs had a higher luminosity (L*) (P < 0.05) than the control eggs, this means that the treatment eggs had a brighter color than the control eggs. On the other hand, control eggs showed a greater (P < 0.05) tendency to red (+a*) than treatment eggs and in terms of the value of b*, no significant differences were detected between treatments (P > 0.05). Schwägele, (2011) mentions that differences in yolk color can be due to the ingredients used in the diet (various types of carotenoids).

Correlations between quality indicators: The correlation coefficients between the physical characteristics of the quality of the White Leghorn hen (Gallus gallus domesticus) were determined (Table 6). Between the height of the dense span and the Haugh units, the correlation was highly significant (r = 0.941; P < 0.01). Likewise, the height of the yolk and the yolk index presented a positive and highly significant correlation (r = 0.927; P < 0.01).

Shell weight and shelled egg weight showed a negative correlation (r = -0.321; P < 0.01), this indicates that, as the weight of the shell increases, the weight of the unshelled egg will be less than the weight of a whole egg; the weight of the shell and the thickness of the shell (r = 0.259; P < 0.01), the percentage of the shell (r = 0.620; P < 0.01), the transverse diameter (r = 0.309; P < 0.01), the polar diameter (r = 0.352; P < 0.01) and the Haugh units (r = 0.277; P < 0.01) showed a positive correlation, indicating that they are positively associated. The weight of the shelled egg and the percentage of the shell (r = -0.211; P < 0.05) and Haugh units (r = -0.197; P < 0.01) showed a negative correlation; the weight of the shelled egg and the weight of the yolk (r = 0.318; P < 0.01), the transverse diameter (r = 0.370; P < 0.01) and the polar diameter (r = 0.409; P < 0.01), presented a high and positive correlation. The weight of the yolk and the percentage of the shell (r = -0.262; P < 0.01) presented a negative correlation, this means that as the weight of the yolk increases, the percentage of the shell will be lower; the weight of the bud and the transverse diameter (r= 0.448; P < 0.01) and the polar diameter (r = 0.566; P <0 .01) showed a high and positive correlation, indicating that the weight of the bud determines the polar and transverse diameter.

The pH of the egg white and the pH of the yolk (r = -0.386; P < 0.01) and the percentage of the shell (r = -0.240; P < 0.01) showed a negative correlation. The percentage of shell and cross-sectional diameter (r = – 206; P < 0.05) and the polar diameter (r = -0.220; P < 0.05) indicated a negative correlation; the percentage of shell and Haugh units showed a positive correlation (r = 0.301; P < 0.01). The transverse diameter y, the polar diameter (r = 0.471; P < 0.01) and the shape index (r = 0.709; P < 0.01) showed a negative correlation, indicating that the transverse diameter determines the polar diameter and the shape index. The polar diameter and the shape index show a negative correlation (r = -0.287; P < 0.01). The Haugh units and the yolk index also showed a negative correlation (r = 0.201; P < 0.05).

CONCLUSIONS

The addition of a yeast additive in the diet of White Leghorn (Gallus gallus domesticus) increased egg weight and yolk luminosity (L*), which also showed a decrease in the a* value. On the other hand, the statistical correlations between egg quality indicators suggest the possibility of implementing programs for the use of the liquid additive of apple yeast in poultry feed, to improve some egg quality indicators, such as egg weight.

Table 1. Descriptive statistics for the external quality variables of White hen eggs Leghorn.

	Average	D.E	Minimal	Maximum
Treatment				
Shell weight (g)	7.06	0.66	5.90	9.13
Shell thickness (mm)	0.36	0.04	0.29	0.44
Percentage of shell (%)	12.41	1.01	10.31	14.81
Form Index (IF)	132.38	5.36	118.60	146.51
Egg-shelled weight (g)	57.04	4.26	49.43	66.24
Cross Diameter (cm)	4.25	0.12	4.00	4.50
Polar diameter (cm)	5.62	0.23	5.10	6.30
Control				
Shell weight (g)	7.50	0.79	5.96	9.57
Shell thickness (mm)	0.36	0.03	0.28	0.46
Percentage of shell (%)	13.33	1.88	10.36	19.97
Form Index (IF)	132.88	3.74	124.39	140.91
Egg-shelled weight (g)	56.68	5.47	36.31	68.13
Cross Diameter (cm)	4.25	0.11	4.00	4.50
Polar diameter (cm)	5.65	0.19	5.10	6.20

Table 2. Descriptive statistics for the internal quality variables of the egg of White Leghorn hen.

Variable	Average	D.E	Minimal	Maximum
Treatment				
Weight of shelled egg (g)	49.25	3.82	41.82	58.84
Dense Clear Height (mm)	9.25	1.08	7.00	13.00
Height of the yolk (cm)	1.85	0.16	0.90	2.10
Bud diameter (cm)	4.03	0.13	3.70	4.40
Yolk weight (g)	14.29	1.06	11.74	17.26
Clear pH	8.98	0.22	8.55	9.35
pH bud	6.06	0.09	5.85	6.24
Yolk Index (IY)	46.02	3.99	21.95	52.50
Haugh Units (UH)	98.28	4.65	88.51	112.10
Clear Mean Diameter (cm)	11.89	1.01	9.60	13.75
Clear Weight (g)	34.96	3.25	30.08	43.55
Clear percentage (%)	61.24	2.31	56.42	67.82
Percentage of yolk (%)	25.11	1.56	22.03	28.98
Clear Index (CI)	7.85	1.23	5.76	10.89
Control				
Weight of shelled egg (g)	47.71	6.90	38.51	59.49
Dense Clear Height (mm)	9.57	1.78	6.00	20.00
Height of the yolk (cm)	1.81	0.17	1.10	2.10
Bud diameter (cm)	4.07	0.17	3.70	4.50
Yolk weight (g)	14.06	0.95	11.69	16.46
Clear pH	8.69	0.83	3.98	9.50
pH bud	6.16	0.45	5.77	9.15
Yolk Index (IY)	44.57	4.66	26.19	52.50
Haugh Units (UH)	99.84	6.84	83.82	131.19
Clear Mean Diameter (cm)	12.76	1.27	9.75	15.20
Clear Weight (g)	34.42	3.49	26.82	43.03
Clear percentage (%)	61.11	7.53	46.03	95.04
Percentage of yolk (%)	24.99	2.71	20.66	38.36
Clear Index (CI)	7.61	1.83	4.29	16.33

Table 3.
Averages of internal quality indicators of white hen eggs Lghorn, fed with liquid additive from apple yeast.

Indicators	Treatment	Control
Dense Clear Height (mm)	9.25±1.08a	9.57±1.77a
Height of the yolk (cm)	1.85±0.15a	1.80±0.17a
Bud diameter (cm)	4.03±0.12a	4.06±0.16a
Weight of shelled egg (g)	49.25±3.81a	47.70±6.89a
Yolk weight (g)	14.29±1.06a	14.05±0.94a
Clear pH	8.98±0.22a	8.69±0.83b
pH bud	6.06±0.08a	6.15±0.44a
Yolk Index (IY)	46.01±3.98a	44.56±4.65a
Haugh Units (UH)	98.27±4.64a	99.84±6.84a
Clear Mean Diameter (cm)	11.89±1.00a	12.76±1.26b
Clear Weight (g)	34.95±3.24a	34.41±3.49a
Clear percentage (%)	61.23±2.30a	61.10±7.53a
Percentage of yolk (%)	25.10±1.55a	24.99±2.70a
Clear Index (CI)	7.84±1.22a	7.61±1.82a

[a,b] Different superscripts indicate difference ($P<0.05$) in the row.

Table 4. Averages of external indicators of egg quality of White Lghorn hens, fed with liquid additive of apple yeasts.

Indicators	Treatment	Control
Shell weight (g)	7.06±0.65a	7.49±0.78b
Shell thickness (mm)	0.35±0.03a	0.35±0.03a
Percentage of shell (%)	12.40±1.01a	13.33±1.87b
Form Index (IF)	132.37±5.36a	132.88±3.73a
Cross Diameter (cm)	4.24±0.12a	4.25±0.11a
Polar diameter (cm)	5.62±0.23a	5.64±0.19a
Egg-shelled weight (g)	57.04±4.25a	56.68±5.46b

[a,b] Different superscripts indicate statistical difference ($P<0.05$) in the row.

Table 5. Yolk color.

	L*	to*	b*	ΔE
Treatment	44.24±3.56a	2.32±0.94a	43.43±5.12a	————
Control	43.50±2.99b	2.88±1.25b	43.47±3.97a	0.895

[a,b] Different superscripts indicate statistical differences ($P<0.05$) within the column.
L*= luminosity, a*= green (-) and red (+), b*= blue (-) and yellow (+), ΔE= color difference.

Table 6. Correlation coefficients between physical indicators of the egg of White Leghorn hens.

Quality indicator	Acd	Alas	Dy	PC	Phc	Py	Phc	Phy	EC	Pcn	Dt	Dp	Uh	Oh	Ifo
Dense Clear Height (Acd)	1	0.202*	-0.101	0.273**	0.078	-0.077	-0.162	-0.065	0.001	0.243**	0.038	0.063	0.941**	0.222*	-0.012
Height of the yolk (Ay)		1	-0.005	0.161	0.184*	0.259**	0.004	0.017	0.007	0.040	0.173	0.189*	0.178	0.927**	0.034
Bud diameter (Dy)			1	0.072	0.023	0.458**	0.003	-0.016	0.188*	-0.231*	0.253**	0.300**	-0.104	-0.378**	0.034
Shell Weight (Pc)				1	0.072	0.154	-0.321**	0.020	0.259**	0.620**	0.309**	0.352**	0.277**	0.121	0.052
Unshelled Egg Weight (Phc)					1	0.318**	0.026	-0.039	-0.111	-0.211*	0.370**	0.409**	-0.197**	0.160	0.076
Yolk weight (Py)						1	0.152	-0.006	0.085	-0.262**	0.448**	0.566**	-0.132	0.067	0.032
Clear pH (pHc)							1	-0.386**	0.142	-0.240**	-0.051	0.013	-0.172	0.007	-0.065
pH bud (pHy)								1	-0.057	0.089	-0.005	-0.060	-0.058	0.021	0.039
Shell thickness (Ec)									1	0.130	0.004	0.124	0.017	-0.064	-0.096
Percentage of shell (Pcn)										1	-0.206*	-0.220*	0.301**	0.123	-0.051
Cross Diameter (Dt)											1	0.471**	-0.009	0.066	0.709**

Polar diameter (Dp)	1	-0.015	0.064	-0.287**
Haugh Units (Uh)		1	0.201*	-0.001
Yolk Index (ly)			1	0.018
Shape Index (Ifo)				1

*(P<0.05), **(P<0.01)

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