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# INFLUENCE OF FEATHER GENOTYPE, STORAGE DURATION AND TEMPERATURE ON THE EXTERNAL AND INTERNAL QUALITIES OF CHICKEN TABLE EGGS

Dawolor Nusue KANASUAH<sup>1</sup>, Kwaku ADOMAKO<sup>1</sup>, Bernard Ato HAGAN<sup>2</sup> ond Oscar Simon OLYMPIO<sup>1</sup>

<sup>1</sup>Department of Animal Science, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana <sup>2</sup>Department of Animal Production and Health, School of Agriculture and Technology, University of Energy and Natural Resources, Sunyani, Ghana

Email: bernard.hagan@uenr.edu.gh

Supporting Information

ABSTRACT: A study was carried out to determine the influence of the feather genotype, storage duration, temperature and method on the internal and external qualities of chicken table eggs. A total of 864 table eggs collected from naked neck (Nanaff), frizzle (nanaFf) and normal feathered (nanaff) birds were used in the study. A Completely Randomized Design of four factors namely, feather genotypes, storage temperatures (5°C and 26°C), storage duration (0, 7, 14, 21 and 28 days) and storage methods (with or without vegetable oil application) was used. The GLM procedure of GenStat (17th Edition) was used to determine the effects of the four factors and their interactions on external qualities (egg weight, length, and width, shell weight and thickness) and internal qualities (albumen height and weight, yolk height, weight, diameter and colour and Haugh unit) of table eggs. The effect of chicken genotype on proximate composition and nutritional values of table eggs were also determined. Feather genotype had significant (P<0.05) effect on yolk colour and weight whilst storage duration, temperature and method had significant (P<0.05) effects on all the internal qualities of eggs studied except effect of storage duration on yolk colour. The 2-way and 3-way interactions of the factors studied were important sources of variation for many of the internal qualities of eggs studied. With the exception of storage temperature, the other factors studied had significant (P<0.05) effects on many of the external qualities of eggs. The interactions of the factors were not significant (P>0.05) sources of variation for most of the external qualities of eggs. Mutant feather genes (Na and F) positively influence egg qualities which could be utilised to segment the commercial chicken egg market.



Keywords: Feather, Frizzle, Naked neck, Nutritional value, Yolk colour,.

# INTRODUCTION

Eggs contain nutrients which are essential for improving human health. Proper functioning of the body is impeded if essential amino acids, which are the main nutrients in eggs, are lacking. Chicken egg albumen and yolk are reported to contain essential amino acids (Ali et al., 2019; Attia et al., 2020). The United States Department of Agriculture (USDA) (2008) reported that eggs are rich foundations of minerals and vitamins.

A complete egg is composed of parts such as the shell (exterior and interior shell membranes), albumen, air cell, cuticle or bloom, chalazae, germinal disk, nucleus of pander, yolk and vitelline membrane (EDINFORMATICS, 2013). Egg quality is built around a number of traits including albumen height, albumen weight, yolk height, yolk diameter, yolk index, yolk weight, shell ratio, shell thickness, shell weight, egg length, egg weight, egg width and Haugh Unit (Murshed and Qaid, 2024). Several studies have reported significant and positive relationships among egg quality parameters of poultry (Zhang et al., 2005; Inca et al., 2020; Guni et al., 2021). Farooq et al. (2001a) reported positive and significant relationships among egg weight, egg width and egg length for eggs from Fayoumi birds. Similarly, largely positive correlations were reported among egg quality traits of two layer chicken breeds in South Africa (Tyasi et al., 2022). In Japanese qualis, Farooq et al. (2001b) reported that there were positive correlations among shell weight, egg weight and shell thickness of quail eggs. Several external factors such as cleanliness, freshness, egg weight and shell weight are important for consumers' acceptability of eggs (Hamilton, 1982; Sonaiya and Swan, 2004; Batkowska et al., 2023). Internal characteristics such as yolk index, Haugh Unit and chemical composition are also important in poultry breeding because of their influence on growth of chicks, breeding performance and egg quality for consumption (Yahaya et al., 2021). The external and internal quality traits of eggs of hens have influence on the hatchability of fertile eggs, and the weight and development of chicks (Sahan et al., 2014; Iqbal et al., 2016; Hegab and Hanafy, 2019).

The external and internal egg qualities are also influenced by storage duration and storage temperature. Eggs stored at low temperature maintain better egg quality (Samli et al., 2005). Egg weight, shell weight, albumen height, albumen viscosity, Haugh Unit and yolk colour decreased with increasing storage temperature of hens (Lee et al., 2016; Martínez et al., 2021). Eggs maintain qualities better when stored for a short period of time (Jin et al., 2011). Prolonged length of egg

storage deteriorates egg quality of chicken eggs (Nasri et al., 2019; Melo et al., 2020). Tebesi et al. (2012) reported that eggs were able to maintain higher yolk height when stored within 7 days.

The naked neck (*Na*) and frizzle (*F*) genes are two mutant thermoregulatory genes that aid chickens to adapt to high ambient temperatures in, especially, the tropics (Asumah et al., 2022). Layer chickens carrying the *Na* or *F* alleles have been reported to record higher percentage of fertile eggs (Asumah et al., 2022), increased egg production (Fathi et al., 2013; Adomako et al., 2014) and improved egg shell quality (Salahuddin and Howlider, 1991). However, El-Rahman and Makled (2006) reported reduction in shell quality in birds carrying *Na* alleles compared to birds with only *na* alleles.

Whilst there have been several studies on egg quality traits of frizzle, naked neck and normal feathered birds in Sub-Saharan Africa and other parts of the world (Salahuddin and Howlider, 1991; Abou-Emera et al., 2017; Fathi et al., 2022), these studies have barely focused on the nutrient contents of the eggs produced by the birds. In addition, information is scanty on the interactions between feather genotype and egg storage methods on the egg quality traits of chicken eggs. The objective of this study therefore was to determine the influence of feather genotype, storage duration, storage temperature and storage method on external and internal egg quality characteristics, amino acid profile and proximate composition of chicken table eggs.

# MATERIALS AND METHODS

### Location and duration

The research was conducted at Akate Farms and Trading Company Limited (AFTC) at Saaman, Kumasi, Ghana and the Department of Animal Science, Kwame Nkrumah University of Science and Technology within a period of six months.

# Experimental birds and eggs

The experimental birds kept at the AFTC were offspring of crosses between naked neck and frizzle feathered cocks and hybrid commercial Lohmann hens. The naked neck and frizzle feathered, both heterozygotes, were bred with normal feathered Lohmann Brown classic layers in two separate matings to produce offspring which were heterozygous naked neck, heterozygous frizzle feathered and normal feathered chickens in the first filial (F1) generation. Eight hundred and sixty-four (864) table eggs were collected from the naked neck (*Nanaff*), frizzle (*nanaFf*) and normal feathered (*nanaff*) layer chickens (288 per genotype) kept as experimental birds by AFTC, Kumasi, Ghana. The layer birds were 28 weeks old at the start of the experiment. The external and internal egg qualities were determined after collection, using the procedures described by Fayeye et al. (2005).

#### **Experimental design**

A Completely Randomized Design in a 3x2x5x2 factorial was applied. Eggs were obtained from three genotypes being *Nanaff*, *nanaFf* and *nanaff*, stored at two storage temperatures (26°C and 5°C) for four storage durations with a control of 0 days (0, 7, 14, 21 and 28 days) using two storage methods (with or without the application of vegetable oil to the egg shells). For eggs which received oil treatment, Sunny vegetable oil manufactured in Ghana was applied by immersion.

The experiment was conducted in three phases and eggs were collected from the chicken genotypes which were housed in deep litter pens. The three chicken genotypes were placed into nine different pens, with each bird genotype put into three different pens labelled as treatments (T1, T2 and T3) with about 20 birds in each pen. A total of 864 table eggs from the three genotypes were further used in the study with 288 table eggs obtained from each genotype.

### Parameters studied and their measurement

a. The egg width and length was measured using a pair of vernier calipers in centimetres.

b. Egg weight was measured with a digital electric balance in grams.

c. Egg shell thickness was measured with a micrometer screw gauge in mm. Shell thickness was calculated from the average of three measurements taken at the middle, broad end and the small end of the eggs.

e. Yolk diameter was measured with a vernier caliper in centimeters.

f. Yolk colour was determined with the DSM yolk colour fan (formerly Roche Yolk Color Fan). Higher figures indicate deeper yolk colour while lower figures indicate lighter yolk colour.

g. Yolk weight was determined with a digital weighing scale in grams.

h. Yolk height was determined by the use of a tripod spherometer.

i. Albumen weight was also determined by the use of a digital weighing scale.

j. Albumen height was determined with a tripod spherometer in mm.

k. Egg weight loss was determined by subtracting the final weight from the initial weight and expressed as a percentage.

I. Haugh unit was determined using the formula,  $HU = 100 \times \log (H + 7.57 - 1.7W^{0.37})$  introduced by Haugh (1937).

where HU = Haugh Units; H = Observed albumin height (mm); W = Observed weight of egg (g) (Roush, 1981).

Twenty-four table eggs from each of the three genotypes were analysed on each storage period (0, 7, 14, 21 or 28), storage method (with or without vegetable oil application).

Proximate composition of the eggs from the three genotypes was determined by drying egg samples (albumen and yolk) in an oven at 65°C for 72 hours. The dried samples were transferred to the Crops and Soil Science Laboratory, Faculty of Agriculture, Kwame Nkrumah University of Science and Technology for the proximate composition analyses. The nutritional values of egg albumen from the three genotypes were analyzed by Evonik Nutrition South Africa Limited. Similar to the proximate analysis, the albumen was also dried in an oven at 65°C for 72 hours and later transferred to South Africa for the amino acid profile analyses.

# Data analysis

The data on external, internal quality characteristics of eggs, and proximate composition and amino acid profiles of the albumen using the general linear model procedure of GenStat (17th Edition). The model used for the analysis of the data collected is presented below.

 $Y_{ijklm} = \mu + G_i + T_j + D_k + M_l + GT_{ij} + GD_{ik} + GM_{il} + TD_{jk} + TM_{jl} + DM_{kl} + GTD_{ijk} + GTM_{ijl} + TDM_{jkl} + e_{ijklm}$ 

Where Y<sub>ijklm</sub> = measured or calculated variables;

 $\mu$  = overall mean;

G<sub>i</sub> = fixed effect of the i<sup>th</sup> chicken genotype (naked neck, frizzle or normal feathered);

 $T_j$  = fixed effect of the j<sup>th</sup> storage temperature (26°C and 5°C);

 $D_k$  = fixed effect of the k<sup>th</sup> egg storage duration (0, 7, 14, 21 or 28 days);

M<sub>i</sub> = fixed effect of the I<sup>th</sup> egg storage method (with or without cooking oil treatment);

GT<sub>ij</sub> = fixed interaction of the i<sup>th</sup> genotype and the j<sup>th</sup> storage temperature;

GD<sub>ik</sub> = fixed interaction of the i<sup>th</sup> genotype and the k<sup>th</sup> storage duration;

GM<sub>il</sub> = fixed interaction of the ith genotype and the I<sup>th</sup> storage method;

TD<sub>jk</sub> = fixed interaction of the j<sup>th</sup> storage temperature and the k<sup>th</sup> storage duration;

TM<sub>jl</sub> = fixed interaction of the j<sup>th</sup> storage temperature and the l<sup>th</sup> storage method;

DM<sub>kl</sub> = fixed interaction of the k<sup>th</sup> storage temperature and the l<sup>th</sup> storage method;

GTD<sub>ijk</sub> = fixed interaction of the ith genotype, jth storage temperature and the kth storage duration;

GTM<sub>ijl</sub> = fixed interaction of the i<sup>th</sup> genotype, j<sup>th</sup> storage temperature and the I<sup>th</sup> storage method;

TDM<sub>jkl</sub> = fixed interaction of the j<sup>th</sup> storage temperature, k<sup>th</sup> storage duration and l<sup>th</sup> storage method;

 $e_{ijklm}$  = random error term associated with each observation ~  $N(0, \sigma^2_e)$  where  $\sigma^2_e$  is residual variance.

Differences between means were separated using Tukey's Test at 5% probability level.

## **RESULTS AND DISCUSSION**

## Internal qualities of table eggs as influenced by genotype

The effect of chicken genotype on internal egg qualities of table eggs are presented in Table 1. There were no significant differences (P>0.05) among various genotypes in relation to albumen height, albumen weight, yolk diameter, Haugh unit and yolk height. The absence of significant differences for these parameters agrees with the findings of Rajkumar et al. (2009) who observed no significant differences in albumen height, albumen weight, yolk height (at 28 weeks old) and Haugh unit for *NaNa, Nana* and *nana* chicken genotypes in India. Udoh et al. (2012) also reported no significant difference (P>0.05) among three local genotypes in terms of yolk weight, albumen height and yolk height in Nigeria. Frizzle genotype recorded significantly (P<0.05) heavier yolk weight than normal feathered genotype, with the normal feathered showing the lowest value in this trait. The higher yolk weight for the frizzle eggs could probably be due to their efficient feed conversion ratio in converting protein for feather production into their eggs. The heavier yolk weight of eggs from frizzle feathered genotype in this study is contrary to the report of Yakubu et al. (2008) who reported that naked neck chicken eggs had heavier yolk weight compared to eggs from normal and frizzled feathered birds. However, Rajkumar et al. (2009) recorded a significantly (P<0.05) heavier yolk weight for normal feathered birds than naked neck ones in India, and noted that lower yolk weight in naked neck birds indicated lower fat percentage in these birds than their normal feathered counterparts. Non-significant (P>0.05) effect of feather genotype on yolk weight has also been reported by Udoh et al. (2012) and Ogundero et al. (2019) in Nigerian local indigenous chickens.

The yolk colour for naked neck and the normal feathered bird eggs were not significantly (P>0.05) different but both were significantly (P<0.05) different from the frizzle hens which recorded a lower yolk colour value (Table 1). Islam et al. (2011) recorded higher yolk colour values from Bangladesh naked neck chicken which is in agreement with the results of the current study. However, Rajkumar et al. (2009) reported higher yolk colour in normal feathered (8.00) and naked neck (7.49) than observed in the present findings. Yolk colour is probably controlled mainly by nutrition than genetics (Grashorn, 2016), hence the varying results for yolk colour as influenced by feather genotype in literature and this study.

Items	Albumen height (mm)	Albumen weight (g)	Haugh unit (%)	Yolk colour	Yolk diameter, (cm)	Yolk height, (mm)	Yolk weight, (g)	
Genotype								
Nanaff	4.52	33.74	60.36	<b>4.94</b> <sup>a</sup>	4.21	13.38	<b>17.37</b> ª	
nanaFf	4.52	33.69	60.47	<b>4.52</b> <sup>b</sup>	4.23	12.88	<b>17.48</b> ª	
nanaff	4.43	32.30	59.92	<b>4.93</b> ª	4.22	13.18	<b>17.15</b> <sup>b</sup>	
SEM	0.092	0.295	0.737	0.098	0.019	0.013	0.098	
P-value	0.544	0.512	0.855	0.003	0.822	0.116	0.042	
Storage duration (days)								
0	<b>4.82</b> ª	<b>34.57</b> ª	62.25ª	4.99	4.05°	<b>14.62</b> ª	17.36	
7	<b>4.49</b> <sup>b</sup>	34.28 <sup>ab</sup>	60.94 <sup>b</sup>	4.80	<b>4.19</b> ℃	13.15 <sup>b</sup>	17.33	
14	<b>4.44</b> <sup>b</sup>	33.75 <sup>b</sup>	59.95°	4.83	4.23 <sup>b</sup>	<b>12.91</b> °	17.25	
21	<b>4.40</b> <sup>b</sup>	<b>32.58</b> ⁰	59.80°	4.60	4.25 <sup>b</sup>	<b>12.93</b> °	17.31	
28	4.31°	32.70°	58.30 <sup>d</sup>	4.76	<b>4.41</b> <sup>a</sup>	<b>12.13</b> <sup>d</sup>	17.38	
SEM	0.084	0.354	0.873	0.116	0.022	0.015	0.112	
P-value	<0.001	<0.001	0.005	0.216	<0.001	<0.001	0.938	
Storage temperature								
Refrigeration (0°C)	<b>5.21</b> ª	<b>34.22</b> ª	68.27ª	5.07ª	4.00 <sup>b</sup>	15.13ª	<b>17.16</b> <sup>b</sup>	
Room temperature (26°C)	3.77 <sup>b</sup>	32.93 <sup>b</sup>	52.32 <sup>b</sup>	<b>4.52</b> <sup>b</sup>	<b>4.46</b> ª	<b>11.15</b> <sup>b</sup>	<b>17.51</b> ª	
SEM	0.247	0.059	0.617	0.082	0.015	0.144	0.079	
P-value	<0.001	0.001	<0.001	<0.001	<0.001	<0.001	0.008	
Storage method								
Vegetable oil	<b>4.79</b> ª	<b>33.60</b> ª	<b>63.77</b> ª	<b>4.89</b> ª	4.08 <sup>b</sup>	<b>14.08</b> ª	<b>17.44</b> ª	
No oiling	3.85 <sup>b</sup>	32.86 <sup>b</sup>	54.00 <sup>b</sup>	4.55 <sup>b</sup>	<b>4.39</b> ª	<b>11.75</b> <sup>b</sup>	17.15 <sup>b</sup>	
SEM	0.059	0.247	0.617	0.116	0.015	0.144	0.079	
P-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	

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## Internal qualities of table eggs as influenced by storage duration

The effect of storage duration on the internal qualities of table eggs is presented in Table 1. Storage duration of eggs did not have any significant (P>0.05) effect on yolk colour and yolk weight. However, Jin et al. (2011) reported significant effect of storage time on yolk colour of laying hens at peak production. Duration of egg storage significantly (P<0.05) influenced albumen height, albumen weight, Haugh unit, yolk diameter and yolk height. Albumen height, albumen weight, Haugh unit and yolk height largely decreased with increase in length of egg storage. The importance of storage duration on albumen height in this study corroborates the findings of Raji et al. (2009) and Santos et al. (2019) who observed decline in albumen height with increase in storage length. Akinola and Ibe (2014) and Abioja et al. (2021) also reported similar findings to the present study. Tebesi et al. (2012), however, reported different findings with eggs stored for 14 days showing higher albumen height. Yolk diameter significantly (P<0.05) increased with increase in storage length. This could be due to the expansion of yolk as storage length increases. This finding agrees with results of Abioja et al. (2021) in FUNAAB-a chicken eggs. The reduction in yolk height with increase in storage length could be attributed to loss in moisture from the yolk resulting in shrinkage of the yolk. The current result agrees with Raji et al. (2009) and Tebesi et al. (2012) who reported higher yolk height for day 7. Similarly, the decline in Haugh unit with increase in storage duration is an indication of deterioration of egg quality. USDA (2000) reported that higher Haugh unit determines the protein content and freshness of eggs. Several authors (Raikumar et al., 2009; Raii et al., 2009; Akinola and Ibe, 2014; Abioia et al., 2021) have presented results of higher Haugh unit with reduced storage duration of eggs which corroborate the findings from this work.

#### Internal qualities of table eggs as influenced by storage temperature

Results of the influence of storage temperature on internal egg quality is presented in Table 1. Albumen height, albumen weight, Haugh unit, yolk height, yolk colour, yolk diameter and yolk weight were significantly (P<0.05) affected by storage temperatures.

Albumen height was higher for eggs stored in a refrigerator than those stored under room temperature. This finding corroborates the results of Scott and Silversides (2000) and Samli et al. (2005) who observed increased albumen height for refrigerated eggs compared to eggs stored at room temperature. This could be attributed to the fact that eggs stored in a refrigerator maintain better albumen quality than those stored at room temperature. Refrigeration of eggs enhance the ability to retard carbon dioxide loss and breakdown of carbonic acid to carbon dioxide leading to the maintenance of egg quality (Qin et al., 2024). The heavier weight of the albumen for refrigerated eggs could be due to the prevention of evaporation of moisture from eggs stored in a refrigerator as a result of low temperature. The retention of Mucin fiber in the albumen of eggs stored in a refrigerator could have prevented the albumen from becoming watery and losing weight (Mountney, 1976). Khan et al. (2013) noted that albumen quality deterioration could be due to the effect of evaporation of moisture and carbon dioxide from the egg when stored under room temperature.

The mean yolk weight of eggs stored in a refrigerator was significantly heavier (P<0.05) than those stored at room temperature. This could be due to the retention of moisture in the yolk of eggs stored in a refrigerator. Samli et al. (2005) observed that there was a decrease in yolk weight with increase in storage temperature. Eggs stored at room temperature showed significantly lower yolk colour value (4.52) than eggs stored in a refrigerator (5.07) and this agrees with Jin et al. (2011) who reported significant effect of storage temperature on yolk colour. Yolk diameter also exhibited a significant difference (P<0.05) with eggs stored at room temperature showing higher yolk diameter than the ones stored in a refrigerator. Yolk height was significantly higher (P<0.05) for eggs stored in a refrigerator than eggs stored at room temperature. This result was similar to the finding of Raji et al. (2009) who recorded higher yolk height for eggs stored in a refrigerator compared to those stored at room temperature. Haugh unit showed a significant difference (P<0.05) with eggs stored at room temperature. Haugh unit showed a significant difference (P<0.05) with eggs stored at room temperature. Haugh unit showed a significant difference (P<0.05) with eggs stored in a refrigerator recording higher Haugh unit (68.27) than eggs stored at room temperature (52.32). The higher Haugh unit indicates the freshness of eggs stored in a refrigerator as Haugh unit value determines the changes of the interior qualities of eggs. Park et al. (2003) and Grashorn et al. (2016) also recorded a decrease in Haugh unit for eggs stored under high temperature. The present result is similar to that of Dudusola (2009) and Raji et al. (2009) who recorded higher Haugh unit values for eggs stored in a refrigerator compared to eggs stored in a refrigerator compared to eggs stored at eremines the changes of the interior qualities of eggs. Park et al. (2003) and Grashorn et al. (2016) also recorded a decrease in Haugh unit for eggs stored under high t

#### Internal qualities of table eggs as influenced by storage method

The effect of storage method on internal egg quality traits is presented in Table 1. There were significant (P<0.05) differences in internal egg qualities between eggs coated with vegetable oil and those that were not coated with vegetable oil. This difference could be attributed to the fact that oil has the ability to seal egg pores, preventing evaporation of moisture and carbon dioxide from the eggs during storage. Eggs coated with vegetable oil had heavier albumen weight and higher albumen height values than those that were not coated with vegetable oil. This might be due to the retention of moisture within the albumen of oiled eggs in the absence of osmotic pressure as observed by Orji et al. (1981). Eggs coated with vegetable oil were significantly higher (P<0.05) in yolk weight and yolk height than those eggs stored without vegetable oil and these values could be due to increase of fat within the yolk through absorption or reduction in moisture evaporation from the yolk. Raji et al. (2009) also observed higher yolk height in eggs of laying hens in a dry climate stored with vegetable oil.

The yolk colour was significantly (P<0.05) higher for oiled eggs (4.89) compared to eggs stored without oil application (4.55). The significantly higher yolk colour value observed for oiled eggs indicates that eggs stored after oil application maintained better yolk colour than eggs stored without oil application. Yolk colour has effect on the nutritional value of eggs. Eggs stored without oil application showed significantly higher (P<0.05) yolk diameter than those stored after vegetable oil application. The higher yolk diameter indicates the spread of yolk as a result of moisture loss from the yolk. This could be attributed to the evaporation of moisture from the eggs during storage as a result of high temperature. Oil helps to seal the various pores on the eggs preventing evaporation of moisture during storage.

Haugh unit showed a significant difference (P<0.05) with eggs coated with vegetable oil showing higher value (63.77) than eggs without vegetable oil (54.00). Güçlü et al. (2008) observed Haugh unit similar to the current results in Table 1, with eggs stored with fish oil showing higher (P<0.05) Haugh unit than other storage methods. Dudusola (2009) reported results that were similar to the current study. The current result also agrees with the finding of Grobas et al. (2001) and Eke et al. (2013) who reported significantly (P<0.05) higher Haugh unit for eggs stored with oil.

# Internal qualities of table eggs as influenced by the two-way and three-way interactions of feather genotype, storage duration, storage temperature and storage method

The P values of the influence of the two and three-way interactions among genotype, storage duration, storage temperature and storage method on internal egg quality traits are presented in Table 2. No significant interaction of genotype by storage duration on internal egg qualities was observed except for yolk weight (0.008). This suggests that variation in yolk weight of chicken genotypes is dependent on the storage durations of the eggs. Significant interaction (0.034) of genotype and storage method on yolk colour of eggs was also observed.

Storage duration x storage temperature had significant effect on all the internal egg qualities studied except yolk colour. This is in agreement with other studies who have reported important storage duration x temperature influence on Haugh unit (Chung and Lee, 2014), albumen height (Samli et al., 2005), albumen weight and yolk weight (Jin et al., 2011). However, Jin et al. (2011) reported significant storage duration x storage time interaction on yolk colour which was contrary to the finding in this study. The variation between the two studies could be attributed to the differences in the breeds of chicken used.

Yolk height was significantly higher (P<0.05) for eggs coated with vegetable oil during storage; although the yolk height values slowly decreased with increase in storage time. Eggs stored without oil application rapidly deteriorated in yolk height as storage time increased. This result agrees with Tebesi et al. (2015) and Raji et al. (2009) who recorded higher yolk height values for eggs stored with oil application for shorter periods of time.

Storage temperature x storage method had significant (P<0.05) effect on albumen height, Haugh unit, yolk diameter, yolk height and yolk weight. There were significantly higher (P<0.05) albumen height values for oiled eggs stored in refrigerator as compared to eggs stored without oil application under room temperature. The eggs coated with oil stored at room temperature or refrigerator also recorded higher albumen height values than eggs stored at room temperature or refrigerator without oil application. This indicates that eggs coated with vegetable oil and stored in a refrigerator maintained better albumen quality possibly due to the prevention of moisture loss by evaporation thus retention of moisture in the albumen as the oil seals the egg pores. Dudusola (2009) reported that eggs coated with oil and refrigerated eggs did not lose much solvent as compared with those in polythene bag and uncoated. The significant variations in yolk weight and diameter due to storage temperature x storage method corroborate the findings of Dudusola (2009) and Orji et al. (1981) respectively who observed increased in yolk weight and diameter as a result of increase in storage temperature and storage time. The increased yolk weight during storage at room temperature could be due to movement of water from albumen to the yolk due to some high pressures. In addition, the significant variation in yolk height due to storage temperature x storage method agrees with Raji et al. (2009) who recorded higher yolk height values for oiled eggs stored at low temperature. Similarly, the important variation in Haugh unit due to storage temperature x storage method is an indication that changes in egg quality due to storage temperature is dependent on the presence or absence of oil application on the egg.

The three-way interactions of the factors studied were significant sources of variations for some of the internal egg parameters except yolk colour (Table 2). The explanation of some of these complex interactions could be quite complicated.

## External qualities of table eggs as influenced by chicken genotype

The effect of genotype on external egg qualities are presented in Table 3. Chicken genotype had significant (P<0.05) effect on all the external qualities of eggs studied except egg weight. The differences in the external qualities could be attributed to the differences in the alleles controlling the genotypes. These results corroborate the findings of Egahi et al. (2013) who also found significant (P<0.05) effect of genotype on all the external egg qualities studied including egg weight. Rajkumar et al. (2009) however observed no significant differences in shell weight and egg weight for *NaNa*, *Nana*, and *nana* genotypes in India. Udoh et al. (2012) also reported no significant difference (P>0.05) among three local genotypes in terms of shell thickness in Nigeria.

Naked neck showing higher shell thickness value could be attributed to the result of their feed intake and feed conversion ratio. As the naked neck take in more feed, calcium from the feed is being converted into the egg shell thereby making their shell thicker than the other birds. However, the frizzle and the normal feathered birds showed no significant difference (P>0.05) in terms of shell thickness. The current result for naked neck was similar to that of Nwachukwu et al. (2006), who also recorded shell thickness between 0.30 mm to 0.34 mm in naked neck, frizzle and normal feathered birds. Yakubu et al. (2008) observed 0.38 mm of shell thickness in naked neck chickens from Nigeria which was higher than the values realized in the present study (0.31 mm). Egahi et al. (2013) also reported shell thickness of 0.33 mm in naked neck, 0.36 mm in frizzle, and 0.32 mm in normal feathered birds.

Table 2 - P-values of the 2 and 3-way interactions among the effects of feather genotype, storage duration, storage
temperature and storage method on internal egg quality characteristics

Source of variation	Albumen helght (mm)	Albumen weight (g)	Haugh unit (%)	Yolk colour	Yolk diameter, (cm)	Yolk height, (mm)	Yolk welght, (g)
Genotype*SD	0.994	0.384	0.505	0.986	0.099	0.112	0.008
Genotype*ST	0.937	0.408	0.945	0.936	0.958	0.715	0.856
Genotype*SM	0.467	0.467	0.580	0.034	0.397	0.583	0.568
SD*ST	0.052	<0.001	0.035	0.623	<0.001	<0.001	0.003
SD*SM	0.830	0.493	0.830	0.643	1.000	<0.001	0.126
ST*SM	0.017	0.696	<0.001	0.144	<0.001	<0.001	<0.004
Genotype*SD*SM	0.183	0.018	0.308	0.657	0.375	0.010	0.488
Genotype*SD*ST	0.077	0.140	0.111	0.423	0.506	0.020	0.173
Genotype*ST*SM	0.300	0.850	0.350	0.670	0.040	0.561	0.402
SD*ST*SM	0.043	0.824	0.008	0.655	<0.001	<0.001	0.008
<sup>1</sup> SD: Storage duration; ST: St	orage time; SM: S	torage method					

 Table 3 - External qualities of table eggs as influenced by genotype, storage duration, storage temperature and storage method

Footo		Shell thickness	Shell weight	Egg weight	Egg length	Egg width
гасц	<b>JIS</b>	(mm)	(g)	(g)	(cm)	(cm)
Geno	otype					
I	Nanaff	0.30a	6.01ª	61.46	5.90 <sup>a</sup>	<b>4.32</b> <sup>a</sup>
I	nanaFf	0.25 <sup>b</sup>	6.09 <sup>a</sup>	61.21	5.81 <sup>b</sup>	<b>4.33</b> ª
I	nanaff	0.22 <sup>b</sup>	5.95 <sup>b</sup>	60.95	5.80 <sup>b</sup>	<b>4.28</b> <sup>b</sup>
:	SEM	0.004	0.042	0.333	0.016	0.010
I	p-value	<0.001	0.043	0.569	0.035	0.003
Stora	age duration					
(	0	<b>0.27</b> ª	6.16ª	<b>63.21</b> ª	5.95ª	<b>4.37</b> ª
-	7	<b>0.27</b> ª	6.02 <sup>b</sup>	<b>61.45</b> ⁵	<b>5.88</b> <sup>a</sup>	<b>4.31</b> <sup>a</sup>
:	14	<b>0.27</b> ª	5.96 <sup>b</sup>	61.34 <sup>b</sup>	<b>5.87</b> ª	<b>4.31</b> <sup>a</sup>
:	21	0.26ª	5.96 <sup>b</sup>	60.04°	5.83 <sup>b</sup>	<b>4.28</b> <sup>b</sup>
1	28	0.23 <sup>b</sup>	5.99 <sup>b</sup>	59.98°	5.85 <sup>b</sup>	<b>4.28</b> <sup>b</sup>
	SEM	0.005	0.049	0.395	0.019	0.012
1	p-value	<0.001	0.043	<0.001	<0.001	<0.001
Stora	age Temperature					
I	Refrigeration	0.26	6.02	61.45	5.86	4.30
1	Room Temperature	0.25	6.01	60.96	5.86	4.32
	SEM	0.004	0.034	0.279	0.013	0.009
1	p-value	0.901	0.943	0.467	0.326	0.643
Stora	age method					
١	Vegetable oil	0.27ª	6.05	<b>61.50</b> ª	5.87	4.30
1	No oiling	0.24 <sup>b</sup>	5.98	60.40 <sup>b</sup>	5.86	4.31
9	SEM	0.003	0.035	0.300	0.013	0.009
	p-value	<0.001	0.176	<0.001	0.709	0.752
<sup>abc</sup> Me	ans within the same sub-colu	mn with different subscript	ts are significant at I	P<0.05.		

# External qualities of table eggs as influenced by storage duration

Storage duration significantly (P<0.05) influenced shell thickness and weight, egg weight, length and width (Table 3). Shell thickness was lower (P<0.05) for eggs stored for 28 days compared to all the other storage durations. The significant variation in shell thickness due to storage duration is in agreement with the report of Grashorn et al. (2016) but contrary to the finding of Lee et al. (2016) who reported non-significant effect of storage duration on shell thickness. Shell weight and egg weight significantly (P<0.05) decreased with increase in length of storage duration and this corroborates the findings of Samli et al. (2005), Jin et al. (2011), Akinola and Ibe (2014) and Lee et al. (2016). The loss in weight is attributed to water loss through evaporation from the pores in the egg shell and escape of carbon dioxide from the egg albumen (Samli et al., 2005). Dudusola (2009) also indicated that the loss of egg weight due to prolonged storage might be due to loss of carbon dioxide, ammonia, nitrogen, hydrogen sulphide gas and water from the eggs.

#### External qualities of table eggs as influenced by storage temperature and storage method during storage

The external egg qualities were not significantly (P>0.05) different between the two storage temperatures. This finding does not agree with report of Raji et al. (2009) who observed higher egg weight for eggs stored in the refrigerator than those stored under room temperature. Oil application during egg storage had important (P<0.05) effect on shell thickness and egg weight (Table 3). Eggs with oil application had thicker shells and heavier egg weight than those without oil application. The higher shell thickness of oil coated eggs is in agreement with Raji et al. (2009) who also reported higher shell thickness values for eggs coated with oil during storage. The high shell thickness of oiled eggs is due to the layer of oil applied on the shells. In addition, the heavier egg weights of oil applied eggs compared to non-oil applied eggs is probably due to the reduction in moisture loss through the pores on the shells.

# External qualities of table eggs as influenced by the two-way and three-way interactions of genotype, storage duration, storage temperature and storage method

All the two-way and three-way interactions of genotype, storage duration, storage temperature and storage method on the external qualities of eggs were not significant (P>0.05) except the interaction of storage duration x storage method on shell thickness, storage temperature x storage method on egg weight and genotype x storage duration x storage temperature on egg weight (Table 4). The significant (P<0.05) interaction of storage duration x storage method observed in this study corroborates the findings of Tebesi et al. (2012) and Akinola and Ibe (2014) but contrary to the report of Raji et al. (2009).

temperature and storage method on external egg qualities								
Factors	Shell thickness (mm)	Shell weight (g)	Egg welght (g)	Egg length (cm)	Egg width (cm)			
Genotype*SD	0.989	0.702	0.477	0.156	0.550			
Genotype*ST	0.870	0.876	0.939	0.577	0.045			
Genotype*SM	0.910	0.736	0.986	0.069	0.580			
SD*ST	0.513	0.841	0.401	0.256	0.034			
SD*SM	0.007	0.386	0.237	0.632	0.590			
ST*SM	0.100	0.279	0.031	0.765	0.502			
Genotype*SD*SM	0.390	0.500	0.420	0.464	0.886			
Genotype*SD*ST	0.756	0.763	0.009	0.164	0.142			
Genotype*ST*SM	0.820	0.930	0.460	0.402	0.511			
SD*ST*SM	0.453	0.142	0.166	0.685	0.642			
<sup>1</sup> SD: Storage duration; ST: Storage	time; SM: Storage method							

 Table 4 - P-values of the 2 and 3-way interactions among the effects of genotype, storage duration, storage temperature and storage method on external egg qualities

# Effect of genotype on the proximate composition of egg albumen and egg yolk (as-fed basis)

Table 5 shows the effect of chicken genotype on proximate composition of chicken egg albumen. There were no significant (P>0.05) difference among chicken genotypes for all the proximate compositions of egg albumen except ether extract (EE). The EE content of egg albumen from frizzle feathered hens were significantly lower (0.08%) than those of normal feathered and naked neck birds. There was no significant (P>0.05) difference among chicken genotypes with respect to the proximate composition of egg yolk except for ash content. Eggs from frizzle feathered hens recorded higher levels of ash compared to those from the naked neck and normal feathered birds.

# Effect of chicken genotype on the amino acid profile of table egg albumen and egg yolk

There were no significant differences (P>0.05) among the chicken genotypes with respect to amino acid profile of the egg albumen (Table 6) and egg yolk (Table 7). The absence of significant differences among frizzle, naked neck and normal feathered birds with regard to amino acid profiles in the albumen and yolk might be due to the similarity of diet fed to the birds and the same environmental conditions under which they were raised.

Egg part	Moisture (%)	NFF (%)	<b>Ash</b> (%)	FF (%)	CF (%)	CP (%)
Genotype			/ 10/1 (/0)	(//)	••• (,,•)	••• (,••)
Egg albumen						
Nanaff	89.04	5.14	0.27	0.18ª	0.02	5.35
nanaFf	89.39	4.93	0.19	0.08 <sup>b</sup>	0.03	5.38
nanaff	88.66	5.49	0.17	0.20 <sup>a</sup>	0.02	5.45
SEM	0.27	0.23	0.04	0.01	0.04	0.13
P-value	0.18	0.24	0.12	<0.01	0.21	0.83
Egg yolk						
Nanaff	57.06	6.78	<b>1.24</b> <sup>b</sup>	27.08	0.06	7.78
nanaFf	57.00	6.95	<b>1.58</b> ª	26.86	0.08	7.53
nanaff	57.17	6.74	<b>1.16</b> <sup>b</sup>	27.05	0.07	7.81
SEM	0.09	0.18	0.09	0.18	0.03	0.05
P-value	0.40	0.70	0.01	0.67	0.86	0.20

Table 6 - Effect of feather genotype on amino acid profile as a percentage of egg albumen

	Genotype		nonoff	CEM	Duckus
Amino acid profile		nt nanart	nanam	SEM	P-value
ALA (%)	0.5	9 0.59	0.59	0.01	0.95
ARG (%)	0.43	3 0.44	0.45	0.01	0.41
ASP (%)	1.02	2 1.03	1.03	0.01	0.78
CYS (%)	0.1	5 0.14	0.15	0.04	0.58
GLU (%)	1.29	9 1.31	1.32	0.02	0.50
GLY (%)	0.3	5 0.35	0.35	0.03	0.90
HIS (%)	0.24	4 0.24	0.25	0.03	0.76
ILE (%)	0.5	0.50	0.50	0.01	0.86
LEU (%)	0.83	2 0.84	0.84	0.01	0.92
LYS (%)	0.54	4 0.53	0.56	0.01	0.38
MET (%)	0.3	1 0.31	0.31	0.06	0.94
MET + CYT (%)	0.4	5 0.45	0.46	0.06	0.57
PHE (%)	0.5	7 0.59	0.59	0.08	0.82
PRO (%)	0.34	4 0.34	0.35	0.04	0.32
SER (%)	0.64	4 0.64	0.65	0.08	0.62
THR (%)	0.43	3 0.42	0.43	0.03	0.75
VAL (%)	0.6	7 0.67	0.67	0.01	0.90

<sup>1</sup>SEM – Standard Error of Means; P-value: Probability Value; ALA: Alanine, ARG: Arginine; ASP: Aspartic acid; CYS: Cystine; GLU: Glutamic acid; GLY: Glycine; HIS: Histidine; ILE: Isoleucine; LEU: Leucine; LYS: Lysine; MET: Methionine; MET+CYS: Methionine+Cystine; PHE: Phenylalanine; PRO: Proline; SER: Serine; THR: Threonine; VAL: Valine.

	Table 7 - Effect	of genotype on	amino acid	profile of	egg volk
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	Genotype	Nonoff	n e n e 156	nonoff	CEM	Durahua
Amino acid profile		Nanaπ	nanart	nanaπ	SEM	P-value
ALA (%)		0.77	0.75	0.75	0.01	0.23
ARG (%)		0.56	0.56	0.57	0.01	0.72
ASP (%)		1.33	1.31	1.31	0.01	0.08
CYS (%)		0.19	0.18	0.18	0.01	0.66
GLU (%)		1.68	1.67	1.68	0.01	0.48
GLY (%)		0.46	0.46	0.45	0.01	0.45
HIS (%)		0.32	0.31	0.31	0.03	0.40
ILE (%)		0.65	0.64	0.63	0.01	0.63
LEU (%)		1.07	1.07	1.06	0.01	0.24
LYS (%)		0.70	0.68	0.71	0.01	0.39
MET (%)		0.40	0.39	0.39	0.01	0.48
MET + CYT (%)		0.59	0.57	0.58	0.01	0.31
PHE (%)		0.76	0.75	0.75	0.01	0.39
PRO (%)		0.45	0.43	0.45	0.03	0.19
SER (%)		0.84	0.82	0.83	0.01	0.39
THR (%)		0.55	0.55	0.55	0.03	0.08
VAL (%)		0.87	0.86	0.85	0.01	0.49
<sup>1</sup> SEM: Standard Error of Means; P-	value: Probability	Value; ALA: Alan	ine, ARG: Arginine;	ASP: Aspartic acid	; CYS: Cystine; GL	U: Glutamic acid;

PRO: Proline; SER: Serine; THR: Threonine; VAL: Valine.

# CONCLUSION

Naked neck and frizzle genes had positive influence on egg quality traits. Shorter storage duration had positive influence on egg qualities during storage. Eggs stored at low temperature showed positive results in terms of internal egg qualities. Eggs coated with vegetable oil also showed better egg quality during storage. Naked neck recorded heavier egg weight than frizzle and normal feathered in their interactions with storage duration and temperature. Refrigerator and vegetable oil showed better yolk quality in their interactions with storage duration. Information from this study could be used in the preservation of the internal and external qualities of table eggs from chicken.

# DECLARATIONS

This study is part of the thesis of the first author. The complete thesis is available in the library of of the Kwame Nkrumah University of Science and Technology (KNUST). No part of this thesis is however published in any journal or conference proceedings.

#### **Corresponding author**

Correspondence and requests for materials should be addressed to ; E-mail: bernard.hagan@uenr.edu.gh; ORCID: https://orcid.org/0000-0003-2902-5271

#### Data availability

The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

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#### **Ethics Approval**

All experimental procedures which involved the use of birds were conducted in compliance with the ARRIVE guidelines.

#### Authors' contribution

Kanasuah DN performed the field work, analysed the data and drafted the manuscript, Adomako K designed the study, edited that manuscript and approved the final manuscript, Hagan BA designed the study, analysed the data and wrote the manuscript. Olympio OS edited the manuscript.

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#### **Competing interests**

None of the authors of this article has any competing interests in the publication of this article.

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