

RELATIONSHIP BETWEEN SELECTED BIOCHEMICAL  
COMPONENTS OF PLASMA AND EGGS IN THE NIGERIA  
INDIGENOUS HENS AND BLACK HARCO LAYER.

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Target Audience: Poultry breeders and physiologists, human nutritionist,  
egg consumers.

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ABSTRACT

Thirty (30) each of the Nigeria Indigenous (NI) hens and Black Harco (BH) layers were reared separately with cocks for a 12-week period to study the association between the plasma and egg concentrations of protein, cholesterol and calcium. Eggs were selected and blood samples collected at 2-weekly intervals. Both the eggs and plasma were analyzed for protein, cholesterol and calcium concentrations. Three randomly picked hens from each group were slaughtered in the middle of the study for histological sectioning of the ovary, magnum, shell gland and liver. Egg yolk cholesterol concentration was lower while the plasma calcium concentration was significantly higher ( $P < 0.05$ ) in the NI hens as compared to BH layers. Plasma calcium and cholesterol concentrations were positively correlated in the NI hens but negatively in the BH layers. However, there were generally no significant association between the plasma concentrations of protein, cholesterol and calcium and their corresponding levels in the eggs. Histological examination revealed that of the mucosal folds of the shell gland and magnum tissue were no longer in the layers as compared to that of the NI hens, which supported the higher laying performance of the BH layers. Eggs of the NI hens compared favourably with those of the commercial BH layers in terms of chemical qualities. There was also no association between the concentration of these chemical substances in the plasma and those of the eggs.

Key words: Laying chickens, eggs, plasma components.

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DESCRIPTION OF PROBLEM

Chickens egg is classified among the high cholesterol foods. A standard 60gm egg contains between 244-273mg of cholesterol depending on the strain of the layers (1). Plasma cholesterol level is positively associated with the incidence of coronary heart disease in human as well as in livestock. Consequently attention has been on ways to prevent hypercholesterolemia. The relationship between the concentrations of protein and cholesterol of plasma and those of eggs laid has been reported for several exotic chickens

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(2,3). This study was therefore aimed at determine such association in the Nigeria Indigenous (NI) hens and compared to the commonly reared commercial Black Harco (BH) layers.

### MATERIALS AND METHODS

The study involved 30 each of the NI hens 46 weeks old Black Harco (BH) layers in a completely randomized block design. The NI hens were obtained from local rearers around Ile-Ife township while the BH layers were obtained from a medium sized commercial poultry farm in the same locality. Thus the precise ages of the NI hens used from could not be determined, but were estimated to be between 26 and 60 week old at the beginning of the study, judging from their body weights which varied between 950 and 1100g. Both groups were replicated thrice and kept in separate deep together with cocks to simulate the free range system of the rural poultry. The cocks and the hens were in ratio 1.0. Layer's mash (Table 1) and water were given ad libitum. Eggs laid in the last 3 days of every two weeks were pooled for each replicate and 12 eggs were randomly picked per replicate. Six sets of selection were made over a 12 week experimental period. Eggs were broken into a flat disc and physically separated into the component parts using a pair of forcep to unwrap the albumen from the yolk. The yolk was immediately weighed while the shell was air-dried overnight before weighing. The weight of the albumen was obtained by difference. At the same interval, 4 hens were randomly picked from each replicate groups and 3ml of blood was collected with a sterilized syringe and needle through the wing vein into lithium heparinized tubes.

The protein concentrations of the albumen, yolk and plasma were determined by buret colourimetric method. Total cholesterol concentrations of the yolk and plasma were determined by the Zlatkis colourimetric method as described by Varley (4). Calcium concentrations of the plasma and egg shell were determined using the Clark and Collip titrimetric method also described by Varley (4).

At the middle of the experimental period, 3 hens each from both groups were selected and slaughtered for histological study of the liver, ovary, magnum and shell gland (uterus). Thin tissue slices (8-10 micron) were stained by haematoxylin and eosin method. Slided were viewed under the microscope for any histological differences.

Data obtained were subjected to t-test and correlation analysis using the Statistical Analysis System (SAS) computer software (5).

Table 1. Composition of Layer's Mash

Ingredient	% Composition
Maize	38.0
Corn bran	15.0
Wheat offal	3.0
Groundnut cake	11.0
Palm kernel cake	11.75
Cotton seed cake	5.0
Fish meal	6.0
Bone meal	1.50
Oyster shell	8.0
Methionine	0.15
Biomix layer's premix	0.40
Salt	0.20
Ferrous sulphate <sup>a</sup>	0.01
Total	100.01

Nutrient	Proximate Analysis:
Energy (KCalME/Kg)	2420.00 <sup>b</sup>
Crude Protein (%)	17.18
Crude Fibre (%)	6.84
Crude Fat (%)	2.63
Ash (%)	9.83
NFE (%)	56.07

<sup>a</sup> Ferrous Salute included to counter the antinutritional effect of gossypol in cotton seed cake.

<sup>b</sup> AMEN (KJ/gDM) = 31.97 (%EE) + 14.14 (% CP) - 18.37 (%CF) + 14.48 (% NFE)

## RESULTS AND DISCUSSION

The concentrations of protein, cholesterol and calcium in the eggs and plasma of both the Nigeria Indigenous (NI) hens and Black Harco (BH) layers are shown in Table 11. Concentration of cholesterol in egg yolk (13.94/ml) was lower while the plasma calcium concentration (21.14mg/dl) was higher ( $P < 0.05$ ) in the hens as compared to the BH layers (16.78mg/ml and 16.32 mg/dl respectively). Protein concentrations of the yolk, albumen, and plasma as well as the egg shell calcium content and plasma concentration were not different between the NI and BH layers.

Table 2. Concentration of Protein, Cholesterol and calcium in the plasma and eggs of NI and BH hen.

Parameter	BH hens	NI hens
Yolk protein (g/dl)	16.56 ± 0.53 <sup>a</sup>	17.32 ± 0.47 <sup>a</sup>
Yolk cholesterol (mg/ml)	16.78 ± 0.75 <sup>a</sup>	13.94 ± 0.87 <sup>b</sup>
Albumen protein (g/dl)	9.88 ± 0.23 <sup>a</sup>	10.0 ± 0.35 <sup>a</sup>
Shell calcium (g/100g)	24.99 ± 0.68 <sup>a</sup>	27.99 ± 2.93 <sup>a</sup>
Plasma protein (mg/dl)	4.94 ± 0.19 <sup>a</sup>	5.36 ± 0.20 <sup>a</sup>
Plasma calcium (mg/dl)	16.32 ± 0.57 <sup>b</sup>	21.14 ± 0.97 <sup>a</sup>
Plasma cholesterol (mg/dl)	96.88 ± 5.60 <sup>a</sup>	92.22 ± 4.69 <sup>a</sup>

Means on the same row with different superscripts differed significantly ( $P < 0.05$ ).

Genetic variation has been reported on the concentration of cholesterol in the egg yolk of chicken (1, 6) and among poultry species in general (7, 8). This variation has been attributed to the rate of egg production following the observation of more cholesterol in the egg yolk cholesterol concentration and rate of egg production (6). In contrary, the present study showed that the NI hens with lower hen-day production (Table 111) had lower concentration of cholesterol in the egg yolk compared to the productive exotic BH layers. Although, the ages of the NI hens used in this study could not be determined due to their source, the unknown age-in-lay difference between the NI and BH hens may have an effect in this observation. Reports on the effect of age-in-lay on the concentration of chemical components of chicken eggs has been inconsistent (9, 10, 11, 12).

Plasma calcium concentration was probably higher in the NI hens due to very low rate of egg production which does not facilitate constant removal for shell calcification, considering that the birds were fed *ad-libitum* on layer's mash of high content (3.5% Ca). Calcium level of the egg shell was low in both breed compared to the average of 37% reported by other authors (13). This could be traced to the titrimetric method of analysis employed in this study, as most reports are based on result from atomic absorption spectrophotometric method.

**Table 3. Production performance of the Nigeria indigeous (NI) and Black Harco (BH) layers.**

Parameters	NI	BH
Hen-day production (%)	26.69 ±1.93 <sup>b</sup>	59.99 ±1.98 <sup>a</sup>
Feed conv. ratio (gfeed/gegg)	6.03 ± 2.49 <sup>a</sup>	2.93 ±0.64 <sup>b</sup>
Feed intake (g/bird/ay)	56.30 ±1.00 <sup>b</sup>	96.08 ± 2.07 <sup>a</sup>
Body weight (g)	987.50 ±22.48 <sup>b</sup>	1601.39 ± 23.61 <sup>a</sup>
Egg weight (g)	38.19 ± 0.88 <sup>b</sup>	53.58 ±1.22 <sup>a</sup>
Shell weight (g)	4.43 ± 0.10 <sup>b</sup>	5.47 ± 0.13 <sup>a</sup>
Albumen weight (g)	20.60 ± 0.65 <sup>b</sup>	33.18 ± 0.88 <sup>a</sup>
Yolk weight (g)	13.07 ± 0.28 <sup>b</sup>	14.93 ± 0.29 <sup>a</sup>
Albumen: Yolk	1.58 ± 0.04 <sup>b</sup>	2.22 ± 0.04 <sup>a</sup>

Means on the same row with different superscripts differed significantly ( $P < 0.05$ ).

In Table 4, plasma cholesterol (PCH) was positively correlated to plasma protein (PTP) in the BH pullets. While the association between plasma cholesterol (PCH) and plasma calcium (PCA) was negative in the BH pullets, it was positive in the NI hens. In both NI and BH layers there was generally no significant ( $P < 0.05$ ) association between the concentration of protein, cholesterol and calcium of the plasma and the corresponding levels in the eggs laid. The positive association between PCH and PCA in the NI hens agreed with an earlier report (14) where experimentally elevated plasma cholesterol led to an increase in the plasma calcium. The lack of association between the plasma cholesterol has also earlier been reported (2,3). An explanation given to the latter observation was that the concentration of cholesterol in the plasma of laying hens fed normal diet remain fairly stable between weeks and very high levels are only observed during period of rapid yolk development (15). High cholesterol diet is known to increase the plasma cholesterol level (3). Although the cholesterol content of the high diet fed in this study was not determined, the diet was a normal one containing ingredients of low cholesterol content. The same explanation goes for the association between the plasma calcium and shell calcium (16). Therefore, the upsurge in the plasma cholesterol concentration due to rapid yolk development will consequently trigger an increase in the plasma calcium concentration.

**Table 4:** Correlations between the concentration of protein, cholesterol and calcium of the plasma and of eggs in the BH (above diagonal) and NI hens (below diagonal).

	PTP	PCA	PCH	YTP	YCH	ATP	SCA
PTP		-0.04	0.43**	0.17	-0.08	0.24	-0.14
PCA	0.19		-0.37*	0.07	0.03	0.17	0.19
PCH	0.04	0.74**		0.13	-0.15	0.02	-0.21
YTP	-0.39	-0.32	-0.31		-0.63**	-0.09	0.05
YCH	-0.23	-0.21	-0.09	0.41*		-0.15	0.21
ATP	0.18	0.02	0.15-	-0.02	-0.05		-0.25
SCA	0.19	-0.08	-0.03	0.06	0.11	-0.13	

PTP=Plasma protein; PCA = Plasma calcium; PCH = plasma cholesterol; YTP = Yolk protein; YCH =Yolk cholesterol; ATP = albumen protein SCA = Shell calcium.

\* Significant correlation ( $P < 0.05$ )

\*\* Significant correlation ( $P < 0.01$ )

Histological sections revealed that the hepatic cell plates of the liver of both NI and BH hens were predominary two-cell thick and clearly separated by the sinusoids. However, artritic follicles were a more common feature on the surface of the ovary of the BH hens, while the mucosal folds of the oviduct (magnum and uterus) were also longer than that of the NI hens. Atretic and post ovulatory follicles are a normal features on the surface of an active ovary (17). Also the length of the oviduct is positively associated with laying capacity of the hen (18), while the area of secretory surface in the oviduct increases in proportion to the amount of mucosal folding (19). Thus more and frequent follicle scars and longer mucosal folds observed in the ovarian and oviduct sections respectively of the BH layers supported the higher laying capacity of this exotic breed over the NI hens.

### CONCLUSIONS AND APPLICATIONS

1. This study showed that eggs from the Nigerian Indigenous hens are as good as those from the exotic Haco layers in terms of protein content and are even better in the cholesterol concentration.
2. The Nigeria Indigenous hens can be used to evolve commercial layers of lower egg yolk cholesterol content.
3. Under normal diet, there is no significant association between the concentrations of protein, cholesterol and calcium of the plasma and their corresponding levels in the Nigeria Indigenous hens as has been observed in most exotic laying chickens.
4. This study recommends a more detailed trial which should last through the first year in lay of both the exotic and indigenous hens.

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