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Effect of different mating methods on embryonic development of indigenous chicken eggs

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Additional keywords
Incubation period.
Moisture loss.
Embryonic weight.

SUMMARY

The study determined the effect of different mating methods on embryonic development of indigenous chicken eggs. Ninety, 27 weeks old normal feathered indigenous chicken breeders comprising 72 hens and 18 cocks were allotted to three mating methods: pen mating (PM), alternate males (AM) and stud mating (SM). Each group, comprising 24 hens and 6 cocks, was replicated thrice. A total of 135 hatching eggs laid within 4 days across treatments and replicates were used to determine embryonic development. The eggs were accordingly picked, labelled and stored in an air-conditioned room (18°C) before setting. At point of incubation, weight of each egg was determined. Yolk, albumen and embryonic weights were determined on days 1, 7, 10, 15 and 18. Data obtained were subjected to Analysis of Variance in a Completely Randomized Design. Mating methods had no significant (p>0.05) influence on embryonic weight throughout the incubation period. At day 7, eggs produced by hens in SM had significant (p<0.05) highest final (54.25 g) and albumen weights (34.13 g) than eggs from PM (47.92g and 27.19 g, respectively) whereas yolk weight was higher (p<0.05) in eggs from PM (13.45 g) than eggs from AM (11.31 g). Yolk weight on day 15 of incubation was significantly (p<0.05) heavier in eggs from SM (10.70 g) than eggs from AM (8.96 g) and PM (8.59 g). Percentage egg weight loss on day 18 of incubation in eggs from AM (14.36%) and PM (13.70%) were higher (p<0.05) than 11.91% found in eggs from SM. It was concluded that mating methods had no influence on the rate of embryonic development. However, mating methods had significant influence on albumen and yolk weights and they decrease with increasing day of incubation. SM appeared as most suitable method for indigenous chicken production.

Palabras clave adicionales
Período de incubación.
Pérdida de humedad.
Peso embrionario.

Efecto de los diferentes métodos de apareamiento en el desarrollo embrionario de huevos de gallina autóctona

RESUMEN

El estudio determinó el efecto de diferentes métodos de apareamiento en el desarrollo embrionario de huevos de gallinas autóctonas. Noventa, 27 semanas de edad criadores de pollos autóctonos con plumas normales, que comprendían 72 gallinas y 18 machos, se asignaron a tres métodos de apareamiento: apareamiento en corrales (PM), machos alternos (AM) y apareamiento en sementales (SM). Cada grupo, compuesto por 24 gallinas y 6 machos, se replicó tres veces. Se utilizó un total de 135 huevos para incubar puestos dentro de los 4 días entre tratamientos y réplicas para determinar el desarrollo embrionario. Los huevos fueron recogidos, etiquetados y almacenados en una habitación con aire acondicionado (18°C) antes del cuajado. En el momento de la incubación, se determinó el peso de cada huevo. Los pesos de yema, albumen y embriones se determinaron a los días 1, 7, 10, 15 y 18. Los datos obtenidos se sometieron a Análisis de Varianza en un Diseño Completamente al Azar. Los métodos de apareamiento no tuvieron una influencia significativa (p>0,05) sobre el peso embrionario durante todo el período de incubación. En el día 7, los huevos producidos por gallinas en SM tuvieron pesos finales (54,25 g) y de albúmina (34,13 g) significativamente más altos (p<0,05) que los huevos de PM (47,92 g y 27,19 g, respectivamente), mientras que el peso de la yema fue mayor (p <0,05) en huevos de PM (13,45 g) que en huevos de AM (11,31 g). El peso de la yema en el día 15 de incubación fue significativamente (p<0,05) mayor en los huevos de SM (10,70 g) que en los huevos de AM (8,96 g) y PM (8,59 g). El porcentaje de pérdida de peso del huevo en el día 18 de incubación en huevos de AM (14,36%) y PM (13,70%) fue mayor (p<0,05) que el 11,91% encontrado en huevos de SM. Se concluyó que los métodos de apareamiento no tenían influencia en la tasa de desarrollo embrionario. Sin embargo, los métodos de apareamiento tuvieron una influencia significativa en los pesos de la albúmina y la yema y disminuyeron con el aumento del día de incubación. SM apareció como el método más adecuado para la producción de pollo indígena.

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INTRODUCTION

The process of producing hatching eggs in chickens is preceded by successful mating activity of the breeder birds. Success in mating is considered as the transfer of spermatozoa from the male to the female reproductive tract. The mating process in poultry is of several kinds and it includes flock mating, pen mating, stud mating, alternating males and artificial insemination (Lamidi, 2014, pp 13-14).

Fertile avian eggs contain embryos and allow their development to the point at which they are able to survive external environment when they hatch. Christensen and Bagley (1989, p. 69) described embryonic mortality as a non-random event with the chance of an embryonic death occurring not being equal on all days of incubation. Embryo development is a dynamic process determined by both the genetic background of the organism and the environment in which it develops. Environmental alterations during an organism's embryogenesis may induce changes in the development of some physiological regulatory systems thereby causing permanent phenotypic changes in the embryo (Druyan *et al.*, 2012, p. 987).

Development of the avian embryo begins immediately after fertilization in the infundibulum and continues as egg components are deposited over the next 25 -26 hours (Reijirink *et al.*, 2008, p. 581). Most embryonic development occurs while the egg is in the shell gland. This is possible as the body temperature of the hen (41.5^{0}C) is higher than the required temperature for embryonic development (Whiltow, 1986, p. 221). Avian embryos develop within the confines of an egg shell independent of maternal physiological functions (Tazawa, 2004, p. 478). According to Moran (2007, p. 1043), embryonic development can be divided into three major phases: establishment of germ, embryo completion and emergence. It is a dynamic process that requires a fine balance between several factors in order to achieve optimum hatchability and quality chick (Onagbesan *et al.*, 2007, p. 557).

However, several factors such as genetic line of the breeders, age of the breeders, egg weight, incubation temperature, humidity, gas levels and altitude and egg handling (turning) interact to determine embryonic growth and quality of the resultant chicks. Embryonic development is not achieved without mating (Levis, 1997. Pg 2). Different mating methods have been used in research, these include pen, flock, stud and shift mating while artificial insemination is now very common in industrial breeding system. Indigenous chickens have not been well integrated into efficient and modern poultry production. There is also a dearth of information on mating methods suitable for the chickens. The present work is aimed at determining the effects of different mating methods on embryonic development of normal feathered indigenous chickens.

MATERIALS AND METHODS

Site of the experiment

Incubation of eggs was done at the Hatchery Unit of UNAAB-LEVENTIS Agro-Allied Industry Limited, Kotopo, Abeokuta, Nigeria. Determination of embryonic development was carried out at the Laboratory of Department of Animal Production and Health, Federal University of Agriculture, Abeokuta, Nigeria. The site falls within the rain forest vegetation zone of South-Western Nigeria on Latitude 7^0 13″ 49.46″ N. Longitude 30 26″ 11.98″ E and altitude of 76 inches above sea level. The climate is humid with a mean annual rainfall of 1037 mm. The annual mean temperature and humidity is 34.7^0 C and 82% respectively (Google Earth, 2020).

Source of experimental materials

The hatching eggs were obtained from normal feathered indigenous chicken breeders (fifth generation progenies) sourced from Animal Breeding and Genetics Research Unit of Teaching and Research Farms, Federal University of Agriculture, Abeokuta, Nigeria.

Experimental design and egg management

Ninety (90), 27 weeks old normal feathered indigenous chicken breeders comprising 72 hens and 18 cocks, were allotted to three mating methods: pen mating (PM), alternate males (AM) and stud mating (SM) in a Completely Randomized Design. Each group, comprising 24 hens and 6 cocks, was replicated thrice with 8 hens and 2 cocks per replicate.

One-hundred and thirty-five (135) fertile eggs (45 from each mating method) were collected from the birds over a period of four (4) days and held in the hatchery for two (2) days in an air-conditioned room (18°C). Fumigation of the eggs was done by spraying with Potassium tetraoxo-manganate VII ($KMnO_4$) and formaldehyde (HCHO) in ratio 1:2 for 20 minutes in a closed chamber. The eggs were set in egg trays with broad ends upward to prevent rupture of the air cell. Individual eggs and egg trays were numbered and labeled respectively on the basis of treatment and replicates for ease of data collection.

The eggs were weighed individually before they were set in Petersime incubator (B-9870 model). Temperature (37.5^{0}C) and humidity (60%) were automatically regulated. Egg turning was automated on hourly basis to ensure that developing embryos receive even distribution of nutrients and heat.

DATA COLLECTION

Embryonic development

At days 1, 7, 10, 15 and 18 of incubation, nine (9) eggs from each treatment group (three from each replicate) were randomly picked and transported to the laboratory for the study of embryonic development. In the laboratory, each egg was weighed and gently broken inside a Petri dish. After breaking, the embryo was carefully detached from the yolk sac and other membranes. The yolk and albumen were separated from the embryonic fluid and weighed individually.

Determination of moisture loss or Hatching egg weight loss

The difference between the weight of the eggs at setting and different periods of determination of embryonic development was derived for the calculation of moisture loss using the formula below:

$$\text{Moisture loss (g)} = \text{Weight before setting (g)} - \text{Weight after setting (g)}$$

$$\textit{Egg weight loss during incubation} = \frac{\textit{Moisture loss(g)}}{\textit{Weight before setting}} * 100$$

ALBUMEN (%)This was determined by using the formula below:

$$Albumen(\%) = \frac{Albumen\ weight(g)}{Egg\ weight\ at\ thetime\ of measuring\ Albumen\ weight(g)} * 100$$

YOLK (%)

This was determined by using the formula below:

$$Yolk(\%) = \frac{Yolk\ weight(g)}{Egg\ weight\ at\ the\ time\ of\ measuring\ Yolk\ weight\ (g)} * 100$$

Relative embryonic weight (%)

This was determined by the formula below:

$$Relative\ egg\ weight(\%) = \frac{Embryonic\ weight}{Initial\ egg\ weight} * 100$$

Statistical analysis

Data obtained on all the parameters evaluated were subjected to Analysis of Variance (ANOVA) in a Completely Randomized Design (CRD) of SAS (1999). Significantly ($p<0.05$) different means among variables were separated using Duncan Multiple Range Test of the same software.

RESULTS AND DISCUSSION

Effect Of Different Mating Methods On Embryonic Development

Egg composition before incubation as shown in **Table I** revealed that the yolk was significantly ($p<0.05$) heavier in eggs from both AM (14.76 g) and SM (14.75 g) than PM (13.94 g). Embryo development is a dynamic process that requires a fine balance between several factors in order to achieve optimum hatchability and chick quality (Onagbesan *et al.*, 2007, p. 557) and begins immediately after fertilization (Reijirink *et al.*, 2008, p. 581). It therefore suffices to suggest that other factors could be responsible for the higher yolk weight recorded with eggs from alternate males and stud ma-

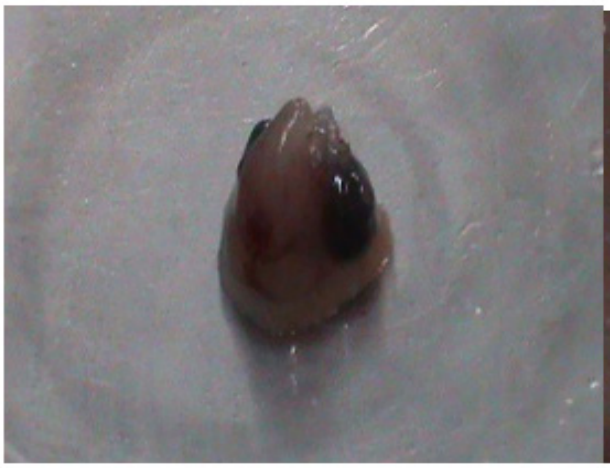

Plate 1:7th day of incubation

Plate 2:10th day of incubation

Plate 3:15th day of incubation

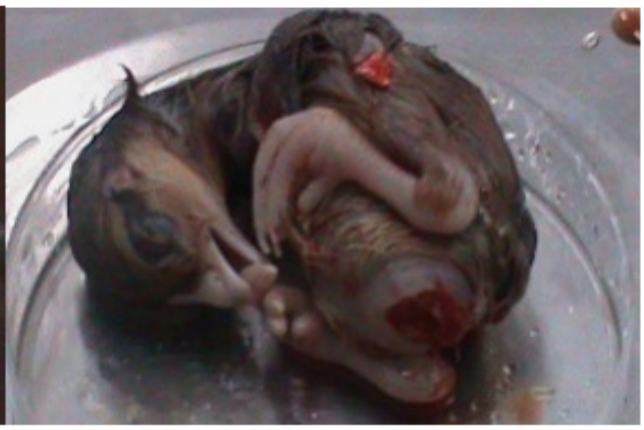

Plate 4:18th day of incubation

Plates from 1 to 4. Incubation progress.

Table I. Effect of different mating methods on embryonic development (Day 1) (Efecto de los diferentes métodos de apareamiento en el desarrollo embrionario (Día 1).

	Mating methods		
Parameters	Pen mating	Alternate males	Stud mating
Average egg weight (g)	50.05±2.58	52.83±1.48	55.78±1.50
Albumen weight (g)	31.18±3.02	32.63±1.34	35.64±1.32
Albumen (%)	62.002.95	61.72±0.94	63.87±0.71
Yolk weight (g)	13.94±0.08^{b}	14.76±0.73^{a}	14.75±0.12^{a}
Yolk (%)	28.01±1.56	28.00±1.04	26.47±0.57

^{a,b} Means on the same row with different superscripts differ significantly ($p<0.05$)

ting as genetic and environmental factors were same. However, the percentage of yolk in day 1 of embryonic development were similar. This also emphasis a probable individual variation in the egg which does not follow any trend.

Similarly, the proportions of albumen and yolk recorded for the hatching eggs across the different mating methods fell within the values reported by Fayeye *et al*. (2005, p. 87) and Sola-Ojo and Ayorinde (2011, p. 258) for Fulani chicken ecotype.

Table II shows effect of different mating methods on embryonic development on day 7. Final egg weight, albumen and yolk weights varied significantly ($p<0.05$) with mating methods. The final weight values ranged from 47.92 g (PM) to 54.25 g (SM) and was significantly ($p<0.05$) heavier in SM than PM but similar in AM and PM (Table II). The significant differences observed in albumen weights were in tandem with that of final egg weight. However, percentage yolk weight was significantly ($p<0.05$) superior in PM compared to the similar values noted in AM and SM (**Plate 1**); a sharp contrast to the trend recorded before incubation.

The differences recorded for final egg weight in 7th day of incubation could be largely due to mating method as SM by practice allows for moderate or controlled use of sire line. Females are brought in from the pen one after the other and introduced to the male kept in the coop. More quality offspring can be obtained from sire of high merit because SM method has been identified to result in higher fertility since males are not over used compared to PM and AM. Therefore, higher final weight in this case could be an attestation to the efficiency of SM in this study.

Also, moisture loss from the hatching eggs of birds in SM and AM groups compared to PM group (Figure 3). Similarly, Deeming (1994, p. 57) found egg weight loss to be less from larger and big birds eggs such as ostrich than small and medium eggs from small birds like chickens. Contrary to the current findings, Hassan *et al.* (2005, p. 1908) and Alibi *et al.* (2012, p. 718) reported that egg weight loss was higher in large-sized eggs compared to medium and small ones.

Albumen weight was also greater in eggs from SM and AM groups than eggs from PM after 7 days of incubation (**Figure 1**). This scenario suggested that albumen consumption by the developing embryo was proportional to the egg weight. The proportion of yolk available to the embryo on the 7th day of incubation was greater in PM than AM and SM groups (**Figure 2**). This is an indication that there was an indirect relationship between yolk mass and egg weight at this period of incubation. Consequently, SM appears suitable for better fertile egg and adequate nutrient supply for a

Table II. Effect of different mating methods on embryonic development (Day 7) (Efecto de los diferentes métodos de apareamiento en el desarrollo embrionario (Día 7)).

	Mating methods		
Parameters	Pen mating	Alternate males	Stud mating
Initial egg weight (g)	51.82±0.80	52.92±1.24	57.28±2.79
Final egg weight (g)	47.92±0.45^{b}	49.63±1.08^{ab}	54.25±2.58^{a}
Moisture loss (g)	3.91±0.73	3.30±0.17	3.02±0.21
Moisture loss (%)	7.51±1.30	6.22±0.17	5.27±0.10
Albumen weight (g)	27.19±0.46^{b}	31.47±0.93^{ab}	34.13±2.29^{a}
Albumen (%)	56.74±0.91^{b}	63.39±0.86^{a}	62.80±1.19^{a}
Yolk weight (g)	13.45±0.56^{a}	11.31±0.20^{b}	12.21±0.36^{ab}
Yolk (%)	28.07±1.10^{a}	22.81±0.40^{b}	22.54±0.44^{b}
Embryonic weight (g)	1.25±0.11	1.29±0.10	1.44±0.03
REW (%)	2.43±0.27	2.44±0.23	2.54±0.16

^{a,b} Means on the same row with different superscripts differ significantly ($p<0.05$)
REW = Relative embryonic weight

Table III. Effect of different mating methods on embryonic development (Day10) (Efecto de los diferentes métodos de apareamiento en el desarrollo embrionario (Día 10)).

	Mating methods		
Parameters	Pen mating	Alternate males	Stud mating
Initial egg weight (g)	53.88±2.32	56.51±0.97	55.29±0.86
Final egg weight (g)	49.47±2.06	51.08±0.34	50.90±1.06
Moisture loss (g)	4.41±0.32	5.42±0.69	4.39±0.19
Moisture loss (%)	8.17±0.34	9.57±0.95	7.95±0.47
Albumen weight (g)	22.70±1.81	22.94±0.14	24.10±1.53
Albumen (%)	45.73±1.76	44.91±0.38	47.35±2.90
Yolk weight (g)	7.35±0.89	8.93±0.57	9.41±0.40
Yolk (%)	14.88±1.78	17.48±1.07	18.46±0.40
Embryonic weight (g)	4.14±0.10	4.22±0.07	4.16±0.13
REW (%)	7.70±0.36	7.48±0.24	7.53±0.27

REW = Relative embryonic weight

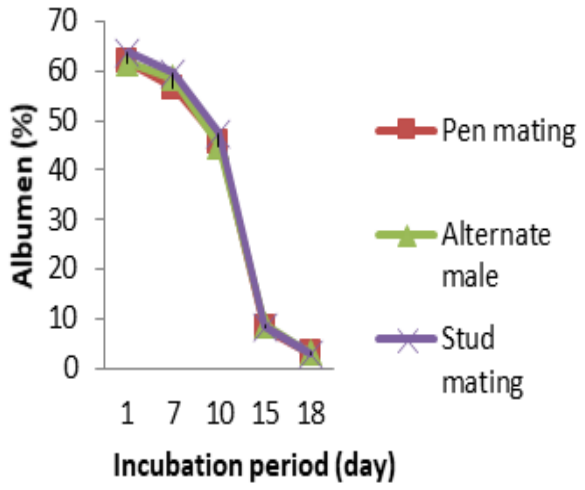

Figure 1. Rate of albumen consumption by developing embryos during incubation (Tasa de consumo de albúmina por embriones en desarrollo durante la incubación).

more efficient embryonic development particularly at early stage of embryogenesis as indicated in day 7 of incubation.

The non-significance of parameters of embryonic development as observed in day 10 of incubation could be due to the fact that most of the germ cells are being formed and there is consolidation which is independent of the mating methods.

The trend in residual yolk on day 15 of incubation (**Table IV**) depicted that mating methods had significant ($p<0.05$) variation on yolk weight. Eggs from birds under SM recorded ($p<0.05$) heavier (10.70 g) than eggs from AM (8.96 g) and PM (8.59g). The proportion of yolk mass was probably the consolidation of the observation on day 7. Yolk weight varied significantly ($p<0.05$) with mating methods and decreased with increasing day of incubation. This was necessary because the embryos had started the process of abdominal internalization of remaining yolk. In the process of hatching, yolk is utilized by the chick either through endocytosis of its contents into circulation or

Table IV. Effect of different mating methods on embryonic development (Day 15) (Efecto de los diferentes métodos de apareamiento en el desarrollo embrionario (Día 15)).

	Mating methods		
Parameters	Pen mating	Alternate males	Stud mating
Initial egg weight (g)	52.02±0.65	54.35±1.75	56.83±1.07
Final egg weight (g)	45.69±1.03	47.68±1.96	49.91±1.41
Moisture loss (g)	6.33±0.02	6.67±0.39	6.93±0.42
Moisture loss (%)	12.19±1.26	12.32±0.99	12.22±0.90
Albumen weight (g)	3.69±0.14	4.14±0.17	4.10.14
Albumen (%)	9.08±0.33	8.70±0.39	8.23±0.44
Yolk weight (g)	8.59±0.09^{b}	8.96±0.20^{b}	10.70±0.56^{a}
Yolk (%)	18.81±0.30	18.89±0.16	21.49±1.43
Embryonic weight (g)	17.69±0.91	18.57±0.16	18.60±0.33
REW (%)	33.99±1.51	34.21±0.91	32.72±0.15

^{a,b} Means on the same row with different superscripts differ significantly ($p<0.05$)
REW = Relative embryonic weight

Table V. Effect of different mating methods on embryonic development (Day 18) (Efecto de diferentes métodos de apareamiento en el desarrollo embrionario (Día 18)).

	Mating methods		
Parameters	Pen mating	Alternate males	Stud mating
Initial egg weight (g)	53.49±1.57	51.41±1.07	53.10±0.90
Final egg weight (g)	46.15±1.26	44.02±0.81	46.77±0.79
Moisture loss (g)	7.33±0.45	7.39±0.31	6.32±0.14
Moisture loss (%)	13.70±0.60[a]	14.36±0.36[a]	11.91±0.17[b]
Albumen weight (g)	1.49±0.23	1.95±0.02	1.94±0.02
Albumen (%)	3.21±0.47[b]	4.43±0.12[a]	4.16±0.10[ab]
Yolk weight (g)	6.82±0.23	6.17±0.16	6.50±0.35
Yolk (%)	14.78±0.38	14.01±0.12	13.88±0.53
Embryonic weight (g)	31.56±1.96	29.03±0.92	29.84±0.49
REW (%)	58.92±2.18	56.51±1.98	56.21±0.46

[a,b] Means on the same row with different superscripts differ significantly ($p<0.05$)
REW = Relative embryonic weight

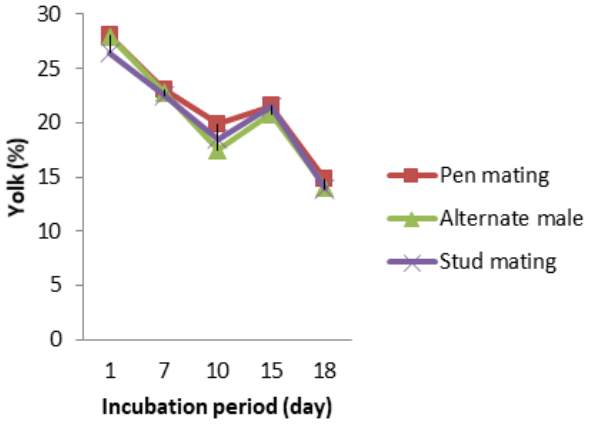

Figure 2. Rate of yolk consumption by developing embryos during incubation (Rate of yolk consumption by developing embryos during incubation).

by transportation through yolk stalk into small intestine (Sklan, 2003, p. 117). Appreciable embryonic growth was observed on the 15th day of incubation (**Figure 4 and Plate 3**).

Table V revealed that egg weight loss was significantly ($p<0.05$) influenced by mating methods (**Table V and Figure 3**). The values of 14.36% and 13.70% obtained for percentage moisture loss in AM and PM respectively were significantly ($p<0.05$) higher than 11.91% found in SM. The heavier eggs lost lesser amount of moisture than the small ones. Yolk weight was still appreciable while albumen was almost exhausted by the embryos on the 18thday of incubation. Percent egg weight loss on the last day of incubation (Plate 4) could be due to the advanced naturity of the embryo while awaiting emergence. This followed the trend already established in Figure 3. Embryonic weight was appreciable and proportional to the Final egg weight obtained across the mating methods in day 7. The progressive growth experienced by the embryos was largely due to prevailing optimum incubation conditions. The trend in embryonic growth observed in this study was consistent with that reported by Rahn *et al.* (1979, p. 297).

The decline in albumen weight (Figure 1) observed across the mating methods during incubation could be premised on its uptake by the developing embryo and also by moisture loss via the pores in the egg shell. The moisture lost from the egg had its origin largely from the albumen since the latter had water (about 85%) as its major constituent. Reduction in mean yolk mass

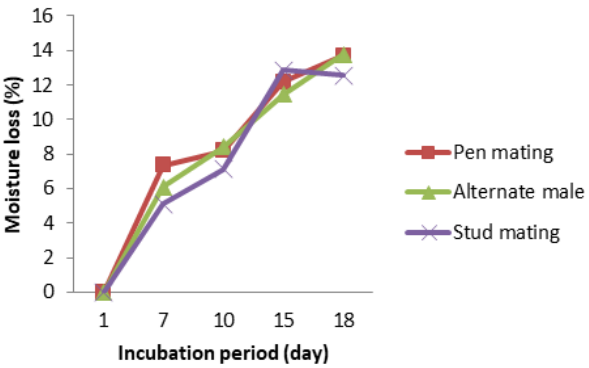

Figure 3. Rate of moisture loss from eggs during incubation (Tasa de pérdida de humedad de los huevos durante la incubación).

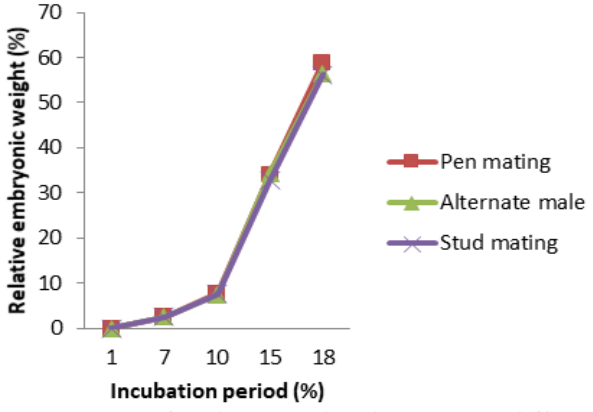

Figure 4. Rate of embryonic development at different stages of incubation (Tasa de desarrollo embrionario en diferentes etapas de incubación).

(Figure 2) observed across the treatments could also be attributed to its supply to the developing embryo. Yolk had been identified as an important nutritional component of the avian egg because it contributes 75% of the joules and provides all the lipids and thus the energy for the developing embryo (Noble *et al.*, 1996, p. 753) as well as being an important source of protein (Deeming, 2002, p. 43). Embryonic development was found to be directly proportional to the egg weight thereby showing that there was consistency in growth (**Plates 1, 2, 3 and 4**).

It was observed that albumen consumption by the embryo was very rapid between the 7th and 15days of incubation (Figure 1). This development was not unexpected because the embryos were undergoing a phase characterized by fully developed chorioallantois, which ensured adequate oxygen-carbon dioxide exchange that would support rapid embryonic growth (Moran, 2007, p. 1043).

CONCLUSION

The findings of this study revealed that mating methods had no influence on embryonic weight. However, mating methods had significant influence on albumen and yolk weights and they decrease with increasing day of incubation while stud mating appeared as most suitable method for indigenous chicken production.

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