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FERTILITY AND HATCHABILITY OF THREE-NIGERIAN INDIGENOUS CHICKEN STRAINS RAISED IN HUMID TROPICAL ENVIRONMENT

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Abstract

Background: The contribution of indigenous chickens to the livelihood of the rural poor in developing countries cannot be overemphasized. Fertility and hatchability of these species in determining a good breeder stocks along with other important dependent factors such as strain and environment constitute major factors to the nutritional and socio-economic sustainability of the people. Objective: Therefore, this study examines the effects of strain on fertility, hatchability and agility of chicks in three-Nigerian indigenous chicken strains. Methodology: Eighty-one birds comprising nine cocks and seventy-two hens namely: naked neck (NN), normal feather (NF) and frizzle feather (FF) were used for the experiment. Each strain was replicated three times at 9 birds per replicate with mating ratio of 1 cock to 8 hens. The cocks were allowed to run freely with the hens on a deep litter system between 30–32 weeks before collecting eggs for incubation in a 4-week period. A total of 1,269 eggs at 474, 417 and 378 for NF, NN and FF respectively were collected from the hens and set for incubation. Data collected were subjected to statistical analysis. Results: Results showed significant ($p<0.05$) differences in all parameters except for percentage fertile eggs in NN and FF. Percentage hen-day production was not significant ($p>0.05$) in NF and NN hens. Agility of the chicks was not significantly ($p>0.05$) influenced by the strain effect. Egg fertility ranged from 77.10% FF to 84.00% NF. Significant ($p<0.05$) strain effect was recorded for hatchability where NN (66.04%) had the highest percentage. The FF had the highest dead-in-shell of 115.29%, infertility was highest in NN (93.00%) while NF recorded the lowest value of 33.00%. Conclusion: It was concluded that continuous reduction in the population of indigenous chickens with major genes of frizzling and naked neck maybe attributed to greater loses of chicks due to poor fertility and embryonic mortalities.

Keywords: *Chicken eggs, Infertility, Major gene, Agility, Embryonic mortality*

INTRODUCTION

Fertility and hatchability are major factors in determining a good breeder stock, and also along with some important dependent factors such as strain, environment, age and age at first delivery (Godard*et al.*, 2007; Kingori *et al.*, 2010). Fertility and hatchability progresses with an increase in the age of breeders (layers) that are well nourished and retrogresses after the attainment of peak at laying (Wiggins, 2008). The health and well-being of any animal is highly correlated with their physical fitness (Ortega *et al.*, 2008). Low level of fitness usually disposes an animal into one health

challenge or the other (Rodrigues *et al*., 2013). The starting quality of day old chicks has a big effect on the starting of growing period and consequently on the final performance (Cervantes, 1997).

The Nigerian indigenous chickens are one of the major chickens known for their good laying ability and serves as a good source of protein to the Nigerian people (Ogbonna *et al.*, 2002). The importation of exotic breeds have overtaken these native chickens because of factors such as fast reproduction cycle, bigger eggs and higher body weight at the detriment of local chickens (Ogbonna *et al.*, 2002). The growth rate of indigenous chicken is generally slower compared to the commercial broilers (Pym *et al.*, 2006). This is a reflection of true genotypic differences and the effect of rearing environment (Sharma, 2010; Ajayi, 2010). These indigenous chickens are regarded as conservation breeds with divergent productive traits (Mtileni *et al*., 2012).

Due to technological development and inadequate information on the reproductive capabilities of indigenous chickens, this study was initiated to supply more information on the fertility, hatchability and agility of three Nigerian indigenous chicken strains raised under intensive management system and their eggs kept at room temperature.

MATERIALS AND METHODS

Experimental Site: The experiment was carried out at the Poultry Unit of the Teaching and Research Farm, the Federal University of Technology, Akure, Ondo State, Nigeria on latitude 07° 16^1 and 07° 18^1 N and longitude 05° 09^1 and 05° 11^1 E. The city of Akure falls in the tropical climate and belongs to the equatorial rainforest belt. It has a bimodal rainfall patterns which start from February – July and September – October with average of 1,556mm per annum. The range of ambient temperature is between30 – 32°C with relative humidity of 80% (Daniel, 2015)**.**

Source of Experimental Birds, Sample Size and Layout: A total of eighty-one (81) birds comprising nine (9) cocks and seventy-two (72) hens belonging to three different strains namely: naked neck, normal feather and frizzle feather were obtained from the Federal University of Agriculture, Abeokuta, Ogun State, Nigeria at 14 weeks of age, raised to laying stage and were used for the experiment. Each strain was replicated three times at 9 birds per replicate with the mating ratio of 1 cock to 8 hens, making a total of 27 birds per strain (3 cocks with 24 hens). All necessary vaccinations and medications as presented in Table 1 were administered as appropriate during the period of the study. The dryness of the litter materials was ensured and changed as and when due during the period of the study. The experiment was carried out using completely randomized design (CRD) with the model given as: $X_{ij} = \mu + T_i + e_{ij}$, where: X_{ij} = overall observation (fertility, hatchability and agility), μ = population mean, Ti = effect of the i^{th} strain (i=1, 2, 3) and eij = random error.

Experimental Diet and Feeding: The birds were supplied with feed and clean drinking water at 125g of feed and water was supplied *ad-libitum* during the period of the study. The birds were fed grower and layer diets at their various physiological stages. The diets used were shown in Table 2.

Mating of Experimental Hens: The hens were mated naturally and the cocks allowed to running with the hens for a 2-week period at the age of 32 – 34 weeks when they were observed to have attained 50% egg laying production. Eggs were collected between weeks 35 – 38 (4 weeks period) where straight line mating was applied during the period.

Method of Egg Collection and Incubation: A total of 1,269 eggs comprising of 474, 417 and 378 eggs for normal feather, naked neck and frizzle feather hen strains respectively were collected during the period of the study. Eggs from the hens were collected thrice (11:00, 13:00 and 15:00 GMT) daily based on their strain for four consecutive weeks. The eggs were sorted and stored at room temperature with adequate ventilation for three days before the eggs were taken to the hatchery for incubation. The eggs were set in local incubator and the hatchery operations were done manually. The eggs were set based on their strain at a temperature range of 32– 37°C and relative humidity of 55– 56% for eighteen days. Then, the temperature was increased to 37.5– 38.5°C and relative humidity to 75 - 80% which was maintained throughout the remaining days of hatching. Eggs were turned manually for at least 5 times each day (Robertson, 1961; Wilson, 1991; Bergoug *et al*., 2013). Data on egg production were collected and kept throughout the laying and incubation periods. Weekly data collected during egg production was summed up and used to express weekly hen-day egg production. Hen day egg production was calculated as percentage of the number of the eggs laid to the number of hens housed (Ali, 2023).

Candling Process, Incubation and Hatching: Candling was carried out on the 10^{th} day of incubation to identify fertile eggs in a dark room using a Candler fixed with a 200 Watt white bulb. The base of the eggs was placed on the Candler for easy penetration of light through the eggs and eggs were viewed against the source of light. The fertile eggs were seen to be densely clouded and opaque with network of veins indicating development of embryo within the eggs while unfertile eggs were translucent under the light (Insko and Martin, 1933; Kaltofen, 1955). Number of infertile and embryonic mortality was recorded. After candling, the fertile eggs were transferred into the hatching tray according to their strain and hatched at11 to 12 days. After the chicks were hatched, they were left in the hatching machine until 90% were dried. On the 21^{st} – 22^{nd} day, the number of hatched chicks including the normal, weak, abnormal chicks and dead chicks after hatch were recorded.

Data Collection: Data were collected on the number of eggs laid and eggs set per strain, number and percentage of fertile eggs, and infertile eggs, number of eggs hatched and hatchability percentage, number of dead in shell and percentage and number of dead germ and percentage and weight of cock and hen at 32 weeks old when they started dropping including the agility test of their progenies. Egg weight was taken as soon as eggs were collected using a sensitive electron weighing balance (Campy model: EK5350) of 0.01 g sensitivity. Agility test was carried out on the three strains of chicks at day-old by placing their back on the floor and then calculate the number of seconds they would use to turn and stand on their feet. Agility was calculated in seconds using a stop watch. The data collected were used to calculate the following parameters according to the procedure of (Ali, 2023).

$$\% \text{ fertility} = \frac{\text{Total number of fertile eggs}}{\text{Total number of eggs set}} \times 100$$

$$\% \text{ Hatchability} = \frac{\text{Total number chicks hatched}}{\text{Total number of fertile eggs}} \times 100$$

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% Dead in shell = $\frac{\text{Total number of chicks dead in shell}}{\text{Total number of fertile eggs}} \times 100$

% Dead in germ = $\frac{\text{Total number dead in germ}}{\text{Total number of fertile eggs}} \times 100$

% Weak in shell = $\frac{\text{Total number of chicks weak shell}}{\text{Total number of fertile eggs}} \times 100$

Hen-day production= $\frac{\text{Number of eggs laid}}{\text{Number of hen housed}} \times 100$

Statistical Analysis: The data generated were subjected to a one way analysis of variance (ANOVA) using SAS (2010) statistical package. Mean difference among treatments (strains) were deemed to be significant at $p<0.05$ and were separated using Duncan Multiple Range Test (DMRT) of the same statistical package.

RESULTS

Table 2 revealed the mean values and percentage of eggs set, fertility and hatchability estimated in the three Nigerian indigenous chicken strains. The numbers of: eggs collected, eggs set, fertile eggs, eggs hatched and normal chicks hatched showed significant ($p<0.05$) strain effects. The normal feather strain chickens had the highest values of 474.00 ± 3.48, 450.00 ± 1.74, 378.00 ± 3.48, 234.00 ± 3.48 and 231.00 ± 0.48 for the above parameters respectively, while frizzle feather recorded the lowest values (378.00 ± 3.48, 377.01 ± 2.64, 294.00 ± 1.74, 141.00 ± 1.74 and 129.00 ± 1.74) respectively for the above parameters. Significant difference ($p<0.05$) was also observed in the number of non-fertile eggs among the chicken strains. The naked neck had the highest value (93.00 ± 3.48), while the frizzle feather strain had the lowest value (33.00 ± 1.74). In the percentage fertility, there was no significant difference ($p>0.05$) between naked neck (77.40 ± 3.45) and frizzled feather (77.10 ± 2.40) chickens. There was numerical difference in their values where naked neck recorded a higher value compared to frizzle feather. Furthermore, significant difference ($p<0.05$) was observed in the percentage fertility of normal feather strain (84.00 ± 3.45) compared with the naked neck and frizzle feather counterparts. The percentage of hatched eggs was significantly different ($p<0.05$) among chicken strains. The naked neck recorded the highest (66.04 ± 3.45%) percentage hatchability, while frizzle feather strain (47.32± 1.98 %) had the lowest percentage hatchability. The number of dead in shell and percentage dead in shell was significantly different ($p<0.05$) among chicken strains. The frizzle feather chickens had the highest values (115.29 ± 1.74 and 38.43 ± 2.22%) and normal feather strain (75.00 ± 1.74 and 19.84 ± 1.74%) recorded the least value, both in terms of number and percentage. Number dead-in-germ and percentage dead-in-germ were equally significantly different ($p<0.05$) among Nigerian chicken strains. The normal feather chicken strain (69.00 ± 1.74 and 18.25 ± 1.74%) recorded the highest value, while naked neck strain (21.00 ± 1.74 and 6.60 ± 0.03%) recorded the least value. Abnormality in chicks was also significantly different ($p<0.05$). Frizzle feather recorded the highest value of 12.00 ± 1.74, while normal feather chicks recorded the least value of 3.00 ± 0.03. In the chicks' agility, there was no significant difference ($p>0.05$) seen in the liveliness and activity of the chicken strains. Numerically, chicks from naked neck recorded the highest smartness (2.00 ± 0.21 seconds), while chicks from normal feather recorded the least alertness value of 2.08 ± 0.20 seconds.

DISCUSSION

Hen Day Production: The result on hen day production revealed that there were no differences between normal feather and naked neck chicken strains, but hen day production of both strains was different from that of frizzle feather. This implies that both normal feather and naked neck chicken strains are good layers. Numerically, normal feather layers had superior egg laying performance than its counterparts which showed that normal feather chickens possessed greater ability to convert feed into egg production. This study was in accord with the findings of Adomako et al. (2013) who observed that the naked neck hens performed better than their frizzle feather counterparts in body weight, number of eggs laid per clutch and hen rate of lay. In addition, Merat (1990) and Horst and Rauen (2008) reported that at moderate temperatures there were no significant difference ($p>0.05$) in laying potentials between the normal feather strain compared to their naked neck counterpart. This shows that the humid tropical climate favours the normal feather laying rate than the chickens with modified body structures such as naked neck and frizzle feather. The findings of this study is in agreement with that of (Peters *et al.* 2000) who observed that the strains of normal feather hens in South-West Nigeria laid higher number of eggs than the frizzle feather strain. However, in spite of the no difference in the number of frizzle feather housed, they still recorded the lowest values for both hen-day-production and percentage hen-day-production which implies that number of hens housed does not determine the hen-day performance. However, the result of this study was at variance with that of Mathur and Horst (1992) and Gowe and Fairfull (2008) as they opined that when frizzle feather strains were raised under high ambient temperatures they performed better in terms of egg production as compared to other indigenous species. The poor performance of frizzle feather hen could be attributed to what was reported by Mathur (2003) that under natural conditions there are large variations in the performance of frizzle feather hens in terms of egg number, egg weight, body weight and production index at different geographical locations (Turkey, Egypt, Cuba, Burundi, Bolivia and Malaysia).

Fertility of Hens: The result of fertility in this study revealed significant cock/hen interactive effect for fertility and hatchability of strains. These two major parameters are of importance to the reproductive performances of birds which are most sensitive to environmental and genetic influences (Dunga, 2013). Fertility was highest in normal feather strain and least in frizzle feather, while naked neck recorded an intermediate value. Fertility was 6.60% lower in naked neck and 6.88% lower in frizzle feather than their normal feather counterparts which are in contrast with earlier reports (Horst, 1989; Peters *et al*., 2008).

Hatchability of Hens: There were differences in the hatchability traits of naked neck, normal and frizzle feather hens. Eggs from the naked neck hens had highest hatchability percentage than the normal and frizzle feather strains. These findings are in agreement with Ahmed *et al.* (2012) in Gazipur and Mymesingh (Bangladesh) who reported that hatchability in naked neck was significantly ($p<0.05$) higher than normal feather chickens. Yakubu *et al.* (2008) mentioned that normal feather chickens had higher hatchability compared to naked neck chickens. Higher hatchability percentages recorded in the study of Ahmed *et al.* (2012) compared to the values obtained in this study. The differences in the results of Ahmed *et al.* (2012) and the present study may be attributed to differences in locations, inseminated method, management system, method of incubation and eggs storage method prior to incubation.

Birds in the study of Ahmed *et al.* (2012) were reared under extensive/scavenging system, while this study was carried out under intensive system. Moreso, the storage system of eggs collected in the study of Ahmed *et al.* (2012) were carried out in a conventional method, while in this present study, storage of eggs were done in the prevalent environmental temperature with local incubation technique. The finding on hatchability in this study disagreed with the studies of Peters *et al.* (2008), Adeleke *et al.* (2012) and Bobbo *et al.* (2013) in Nigeria. In the current study, hatchability of naked neck and normal feather strains were lower compared to the values reported by (Ahmed *et al*. (2012). The author reported hatchability values for naked neck and normal feather chickens to be 87.40 and 86.98% respectively compared to 66.04% for naked neck chickens and 61.90% for normal feather chickens in this study. The differences in the results of the present study compared to that of Ahmed *et al.* (2012) maybe ascribed to differences in management system, locations and ambient temperature. The usually high ambient temperature of Akure in the first quarter of the year could have also contributed to the hatchability estimates obtained in this study.

Dead-in-Shell of Chickens' Embryos: Dead-in-shell of embryos were significantly different ($p<0.05$) with the frizzle gene having the highest value which could be as a result of the effect of frizzling genes. Similar observations were made by Bobbo *et al.* (2013) in Nigeria. According to Kalita *et al.* (2013) various causes of high dead-in-shell embryos include genetic factors, breeds, frequent power failure leading to fluctuations in temperature and humidity which are key factors in hatchability, incorrect turning of eggs and lack of proper hygiene during storage and incubation. Also, Deeming (1995) stated that high embryonic mortality can be observed in poultry due to microbial contamination. Besides, hatchability is adversely affect if eggs are not collected and cooled down to storage temperature, pre-incubation and embryo hatchability (COBB, 2008). Similarly, Ernst *et al.* (2004) stated that high dead-in-shell embryos maybe due to improper incubation temperature, humidity, improper turning and infected eggs. This study agreed with Deeming (1995) findings because eggs were kept at room temperature which might have eventually contributed to the number of dead-in-shell. Also, more dead-in-shells were recorded in frizzle feather eggs which may be due to lethal gene. Sharifi *et al*. (2010) reported that under moderate temperatures the homozygous F-gene on reproductive performance is inconsistent and that the number of eggs and number of chicks are distinctly reduced due to the depressive effect of the homozygous gene.

Alive in Shell and Abnormal Chicks: Alive in shell and abnormal chicks were not affected significantly ($p>0.05$) by strains in this study. High number of dead in shell could be attributed to incorrect turning, improper egg handling and storage (Hananeh *et al.* 2021) while abnormal chicks were attributed to size of eggs, lethal genes and temperature fluctuations. Deeming (1995) stated that failure to turn poultry eggs properly during incubation causes problems with the formation of extra-embryonic fluids. According to COBB (2008) chicks' deformity could be ascribed to temperature variation within incubator due to fluctuations in power. In this study, the number of abnormal chicks could be due to negligence of the hatchery personnel in turning the eggs and maintaining a fairly constant incubation temperature and humidity.

Infertile Eggs: Strains had significant effect ($p<0.05$) on the infertility parameter. The numbers of infertile eggs recorded were highest in naked neck, followed by normal feather and least in frizzle feather chickens. This finding is in agreement with Peters (2005) and Bobbo *et al.* (2013) in Nigeria where naked neck chickens recorded significantly ($p<0.05$) higher number of infertile eggs than in normal feather chickens. The possible cause may either be genetic or environmental factor. The cocks could be overfed and be unable to carry out their mating exercise. However, over feeding results in obesity in some hens leading to infertility which could in turn lead to inability to fertilize the egg during ovulation. Genetically, naked neck cocks have been reported to produce a high population of semen volume but low parked sperm volume, low motility, higher abnormal sperms (Galal, 2000; Mahrous *et al*., 2008). The abnormalities in sperms were significantly ($p<0.05$) affected by an interaction between strains and gene.

Normal Chicks: There was no significant ($p>0.05$) difference between the Normal feather chicks in terms of liveliness and response to stimuli (alertness). However, the feeding and growth rate of the Normal feathered chicks showed less feed intake and reduced growth rate compared to their counterparts; frizzled feather and naked neck and this finding was supported by (Fathi et al., 2013; Davenport., 1914; Warrant., 1933) that the presence of the recessive autosomal gene and their feather orientation prevents the normal chicks from heat dissipation. Moreso, the absence of lethal gene in the normal feathered chicks significantly ($p<0.05$) help their survivability as compared to their counterparts; frizzled feather and naked neck that recorded a high mortality rate despite the exhibition of range of structural and physiological adaptations (Taylor *et al., 1997*)

Agility of Hatched Chicks: It was observed that there were no differences among the chicken strains with regards to their liveliness and agility. This implies that any of the strains might be selected and raised to maturity for production or breeding purposes with similar alertness. This findings corroborated that of Da *et al*. (2020) who reported similar output in their study. However, selection should be done based on environment in which they are to be reared since the naked neck and frizzle feather chicken strains performed better in hot ambient temperature and humid areas while the normal feather strain are good in the rain forest zones of Nigeria (Sharifi, 2006).

Conclusion: This study has shown that the normal feather strain was superior to the other indigenous strains in the number of egg set, fertile eggs, percentage of fertile eggs, number of hatched eggs and least in the number of percentage of dead-in-shell while naked neck strain took the lead in the percentage of hatched eggs and number of non-fertile eggs. Moreover, the continuous reduction in the population of indigenous chicken with major gene of frizzling and naked neck maybe attributed to greater losses of chicks due to poor fertility at natural incubation and embryonic (lethal gene) mortality before hatching. The difference in hatchability among strains was highly significant, fertility of eggs and embryonic mortality during hatching due to strain difference. Therefore, there is need for a shift in incubation process with conscious conservation of these rare genes in order to prevent them from going into extinction. This would enable humanity to use them for research and other purposes.

It is recommended that the genetic make-up of the Nigerian indigenous chickens should be manipulated through genetic engineering to reduce the lethality of their genes and improve their

fertility and hatchability traits. Management of these chickens in controlled environment may further help to advance their potentials in terms of productivity and reproduction. Production of indigenous chickens at intensive level should be encouraged so that larger number of eggs can be collected for incubation using modern hatching technologies. It is also recommended that their eggs should be kept in cold storage soon after collection so as to avert the greater impact of ambient temperature on the eggs collected especially from breeder farms and hatcheries. Management in the breeder farms as well as hatcheries should adjust according to chicken strains because every strain responds differently to hatchability. Management at the breeder farms and hatcheries should use recommendations that are applicable to each strain in making decisions and adjustments by assessing their performance at specific time.

DECLARATIONS

Ethical approval and consent to participate: All animals used for the experiment were handled according to the ARRIVE and EU Directives 2010/63/EU for animal experiments and the National Institutes of Health guide for the care and use of laboratory animals (NIH Publications No. 8023, revised 1978). The strain on the results of the present study were included in the texts as appropriate.

Consent for publication: The Author wrote and purposed to publish this manuscript.

Availability of data and materials: All data used for the preparation of this manuscript is contained therein.

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Table 1: Vaccination/medication schedule

Age (week)	Vaccine/Drug	Route
14	Multivitamin + Antibiotics	Oral
15	Coccidiostat	Oral
16	NDVK (Booster dose)	Intramuscular
17	Fowl Pox vaccine	Wing web
18	Dewormer	Oral
19	Multivitamin	Oral

NDVK = Newcastle disease vaccine komorov. Birds were equally debeaked at week 15.

Table 2: Gross composition of the experimental diet (g/100g)

Ingredients	Growers mash	Layers mash
Maize	54.00	50.00
SBM	10.00	12.00
Wheat offal	22.00	17.00
PKC	3.50	3.50
GNC	4.50	6.50
Fish meal	1.00	1.00
Bone meal	2.60	2.60
Limestone	1.50	6.50
Methionine	0.25	0.25
Lysine	0.10	0.10
Premix	0.25	0.25
Salt	0.30	0.30
Total	**100**	**100**
Calculated Analysis:		
Crude protein (%)	15.50	16.70
ME = (KCal/kg)	2600.00	2550.00

Grower's diet was fed between16 – 20 and layer's mash fed between 21- 40 weeks respectively.
SBM = Soybean meal; PKC = Palm kernel cake; GNC = Ground nut cake; ME = Metabolizable energy

Table 3: Effect of strains on fertility, hatchability and agility of experimental birds

Strain	Normal Feather	Naked Neck	Frizzle Feather
Hen Day Production	16.92 ± 0.66^{a}	14.88 ± 0.72^{b}	11.79 ± 1.05^{c}
Percentage Hen Day Production	70.54 ± 2.44^{a}	70.91 ± 2.68^{a}	43.62 ± 3.18^{b}
Number of eggs collected	474.00 ± 3.48^{a}	417.00 ± 3.48^{b}	378.00 ± 3.48^{c}
Number of eggs set	450.00 ± 1.74^{a}	411.00 ± 3.48^{b}	377.01 ± 2.64^{c}
Number of fertile eggs	378.00 ± 3.48^{a}	318.00 ± 3.48^{b}	294.00 ± 1.74^{c}
Number of non-fertile eggs	72.00 ± 1.74^{b}	93.00 ± 3.48^{a}	33.00 ± 1.74^{c}
Percentage of fertile eggs	84.00 ± 3.45^{a}	77.40 ± 3.45^{b}	77.10 ± 2.40^{b}
Number of hatched eggs	234.00 ± 3.48^{a}	210.00 ± 3.48^{b}	141.00 ± 1.74^{c}
Percentage of hatched eggs	61.90 ± 1.74^{b}	66.04 ± 3.45^{a}	47.32 ± 1.98^{c}
Number of dead in shell	75.00 ± 1.74^{b}	29.00 ± 0.58^{c}	115.29 ± 1.74^{a}
Percentage of dead in shell	19.84 ± 1.74^{c}	27.36 ± 1.74^{b}	38.43 ± 2.22^{a}
Number of dead in germ	69.00 ± 1.74^{a}	21.00 ± 1.74^{c}	39.00 ± 1.74^{b}
Percentage dead in	18.25 ± 1.74^{a}	6.60 ± 0.03^{c}	13.18 ± 1.74^{b}

germ			
Number of normal chicks	231.00 ± 0.48[a]	201.00 ± 0.48[b]	129.00 ± 1.74[c]
Number of abnormal chicks	3.00 ± 0.03[c]	9.00 ± 1.74[b]	12.00 ± 1.74[a]
Agility (Seconds)	2.08 ± 0.20	2.00 ± 0.21	2.02 ± 0.96

a,b,c means within a row having different superscripts are significantly different ($p<0.05$)

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