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Effect of incorporation of heat-tolerant genes into Lohmann brown layers on egg production and quality under hot and humid environments

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ABSTRACT

A breed development strategy aimed at incorporating naked-neck and frizzle genes into Lohmann Brown layers to make them more productive even under warm and humid environment is underway at Akate Farms in Kumasi, Ghana. The present experiment was carried out to find out the effect of the genes on egg production in hot and humid environments. Three genetic groups comprising naked-neck, frizzle, their normally feathered sibs were obtained after 4 successive generations of crossing. In this present experiment, a total of 360, 16-week old pullets, 90 each of the 3 groups in addition to pure Lohmann Brown layer lines (used as the genetic control group) were selected randomly and assigned to a completely randomized design experiment with 3 replicates, with 30 birds in each replicate group and kept up to 72 weeks. The birds were kept in a partitioned open-sided deep-litter house constructed with sandcrete blocks with 30 pullets in each compartment. They were fed *ad lib* with layer diets containing 18% crude protein and 2800 Kcal ME/kg. Results obtained showed that the Lohmann brown layers (used as the control group) were significantly superior in terms of rate of lay, egg mass, egg weight and feed efficiency, but were however significantly inferior in egg quality as compared to their counterparts with the naked-neck and frizzle traits. Within the genetic groups with the heat-tolerant traits, the naked-neck layers out-performed their frizzle and normally feathered sibs in terms of the egg production parameters measured even though they all segregated from similar parents.

Key words: Crossbred, Frizzle, Lohmann Brown, Naked-neck, Normally feathered, Phenotypes

Under hot and humid conditions - a major characteristic effect of global warming- fully feathered birds do not reach their full genetic potential for growth, body weight, meat yield and egg production because dissipation of their excessively produced internal heat is hindered by the feathers (Cahaner *et al*. 2008). According to Garces *et al*. (2001) high environmental temperatures constrain birds' performance under intensive production systems. Dissipation of body heat is reduced, feed consumption is depressed and optimal reproductive and productive functions cannot be maintained. This according to Galal *et al*. (2007) has necessitated the use of major heat-tolerant genes like naked neck (*Na*) and frizzle (*F*) in breed improvement programmes to improve productivity especially in the tropics.

These genes diminish the insulating power of the bird's plumage and thus are associated with increased heat loss. The naked-neck condition is characterized by complete absence or reduced feathers in the neck region of chickens controlled by an autosomal dominant gene (Somes 1990). The heterozygotes have a tuft of feathers on the ventral side of the neck whereas homozygotes have no plumage on the neck (Somes 1990). The frizzle gene (F) curls the feathers and reduces their size, thus increasing the heat conductivity of the feather coverage (Somes 1990). Birds with the naked-neck and frizzle traits have been found to be thermal stress tolerant as compared to their normally feathered counterparts (Nwachukwu *et al*. 2006). The effects of frizzled feathers on the performance of layers were reported by Harren-Kiso *et al*. (1995). Due to the great potential of these genes, crossbreeding between local chickens with the heat-tolerant genes and exotic breeds would allow for the exploitation of the rusticity of the former and high productive performance of the latter in tropical environment so as to produce highly adaptable and superior genetic types.

In pursuance of this, a breed development strategy aimed at incorporating the naked-neck and frizzle genes (common in the local chicken types in Ghana) into Lohmann brown (most common layer breeds in the country) to make them more productive even under extremes of high and humid environment is underway at Akate Farms in Kumasi, Ghana. This breed development programme has reached an advanced

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stage and at the end of the fourth generation, 4 main phenotypes were obtained, based on the presence or absence of the heat-tolerant genes. The present study was therefore to study the effect of the introduction of the naked-neck and frizzle genes into Lohmann brown layers on egg production under hot and humid environments.

MATERIALS AND METHODS

Experimental population: The base population used in this experiment were offspring of a cross involving local chicken male lines heterozygous for the naked neck (*Na*) and frizzle (*F*) genes and a Lohmann brown female line homozygous for the normal alleles of the 2 genes. Due to multi-coloured nature of the male lines, colour sexing at day-old was not possible, therefore the birds were sexed using secondary sexual characteristics. Four different combinations of genes for feather distribution and morphology constituting 4 different phenotypic groups were segregated. These were combined naked-neck frizzle (Nana Ff), naked-neck only (Nana ff), frizzle only (nana Ff) and their normally feathered (nana ff) sibs. This population has been maintained under some intense selection pressure for egg production and kept at Akate Farms, Kumasi, Ghana. To avoid inbreeding, planned pedigreed mating was practiced in each generation**.** Due to the small numbers obtained for the combined naked-neck and frizzle birds, they were not considered for the purposes of this experiment.

Study site and management of experimental birds: Ghana is a tropical country with its northern savannah zone between latitudes 8°302 N and 11°002 N of the equator and longitudes 2°302 W and 0°302 E of the Greenwich Meridian. The experiment was carried out at Akate Farms located at Saaman within the Kwabre district in the Ashanti region of Ghana. The experimental area lies in the transitional forest zone and is between latitude 6.35° and 6.40° and longitude 1.30° to 1.35°, an elevation which ranges between 250–300m above sea level. The average minimum temperature is about 21.5°C and a maximum average temperature of about 36.5°C. The average humidity is about 84.16% at 0900GMT and 60% at 1500GMT. The double maximum rainfall is about 214.3mm in June and 165.2mm in September. The birds were kept in a partitioned open-sided deep-litter house constructed with sandcrete blocks with 30 pullets in each compartment. They were housed in partitioned deep litter pens with stock density of 0.15m²/bird. The feed and water were supplied in 2.5kg capacity hanging feeders and 10 litre capacity plastic fountain drinkers respectively. The birds were fed layer diets containing 18% crude protein and 2800 Kcal ME/kg from the 18 weeks on. Feed and water were provided *ad lib.* The plastic drinkers were cleaned daily throughout the experimental period. The house was constructed in such a way that there was adequate ventilation. There were 8 nests (measuring 30cm × 30cm × 35cm each) in each pen. Egg collection was done twice a day, at 0900GMT and 1500GMT.

The maximum and minimum daily temperatures during the study period ranged between 26.5°C and 35.5°C and 20.1°C and 25.3°C, respectively while the relative humidity ranged between 55 and 80%. There were 12 h of natural light which was supplemented with 4 h of artificial lighting throughout the experimental period. This was achieved by providing 60 watts electric bulbs. This did not only enable the pullets to eat during the night but also stimulated the hypothalamus for proper endocrine communication resulting in continuous ovulation, which enhanced egg production. Fowl pox and third Newcastle vaccinations were carried out at 12 and 16 weeks of age respectively. A coccidiostat was added to their drinking water fortnightly to control coccidiosis. Treatment for worms and lice were done monthly using levasol (water medication) and ectomin (dipping). Miramed was given to them as a prophylactic treatment for chronic respiratory disease (CRD). Vitamin supplements (mixture) like Vitalytes was given to the birds after debeaking, transfering and weighing. All the birds were reared under the same environmental, managerial and hygienic conditions.

Data collection and parameter estimation

Egg production: Data on daily egg production were kept throughout the laying period on pen basis. In terms of egg production, the parameters taken were age at sexual maturity, rate of lay (hen-day), egg mass, egg weight, internal and external egg characteristics. Rate of lay was calculated as the number of eggs produced divided by the total laying period in days (from start of lay to end of experimental period) multiplied by 100. Age at sexual maturity was estimated as the age at which the birds laid their first egg. Egg mass was calculated as the product of egg weight (g) and hen-day egg production. Feed conversion ratio was calculated as the ratio of kilogram feed intake to the kilogram egg produced.

Egg quality estimation: At 30 weeks of age 15 samples of fresh eggs were collected from each of the phenotypes for internal and external egg quality test. This was done at the Department of Physics of the University of Cape Coast. Egg eggshell thickness, albumen height, yolk height, Haugh unit and yolk colour score were considered as determinants for egg quality. Fresh eggs were collected separately for the different groups and weighed using a 0.01g precision top-loading electronic balance. Albumen height was measured using a tripod micrometre. The eggs were broken on a metal plate and height of albumen was measured as the distance between the metal plate and the electrode placed on top of the thick egg white of the broken egg. In order to correct for difference in egg weight, the albumen height was converted into Haugh unit (HU). The Haugh unit was therefore estimated based on the formula by Haugh (1937):

The same tripod micrometre, which was used for albumen height, measured the yolk height; which was estimated as the distance between the metal plate and the electrode placed on top of the yolk of the broken egg. Shell thickness was measured by using a micrometer screw gauge. The shell was cleaned, washed and air dried at room temperature until constant weight was obtained and then thickness was measured along the equator lines.

Experimental design and statistical analysis: Pullets (300), 16-weeks-old, 90 each of naked-neck, frizzle, their normally feathered sibs and Lohmann brown (used as genetic control group) were selected randomly and assigned to a completely randomized design experiment with 3 replicates with 30 birds in each replicate group. The data obtained were subjected to one-way analysis of variance with phenotype effect using general linear model procedure of GenStat. Where significant differences among means were found, means were separated using least significant difference (LSD) test at 5% level of significance.

The linear model below was used for the data analysis.

$$Yij = \mu + gi + \text{å}ij$$

where, Yij , performance of the jth pullet of the ith phenotypic group; μ , overall general mean common to all observations; gi , fixed effect due to ith phenotype (i = 1, 2, 3, 4); åij , random error effects peculiar to each observation

RESULTS AND DISCUSSION

The egg production performance of the naked-neck, frizzle, their normally feathered counterparts and the Lohmann Brown layers are presented in Table 1. The average rate of lay for the entire laying period showed the pure Lohmann Brown layers producing significantly ($P<0.05$) at a higher rates than their crossbred counterparts. This is not surprising because the Lohmann Brown strains have been intensively selected for egg production. Within the crossbred layers, those with the heat-tolerant genes laid significantly at a higher rate ($P<0.05$) than their normally feathered sibs, even though they all segregated from the same flock. This confirms the observation by Merat (1986), Yushimura *et al*. (1997), Barua *et al*. (1998), Abdel-Rahman (2000), El-Safty *et al*. (2006) and Mahrous *et al.* (2008) that when reared under hot and humid environments, naked-neck and frizzle layers outperformed their normally feathered sibs in terms of egg production and feed efficiency. The present results however disagree with that of Rajkumar *et al*. (2011) who recorded no significant differences ($P>0.05$) in part period egg production up to 40 weeks of age. There was also a significant difference ($P<0.05$) in terms of mortality with the naked-neck phenotypes recording the highest deaths as compared to the other phenotypes. One of the problems with the rearing of naked-neck and frizzle birds is the high incidence of pecking due to their exposed skin (Hagan 2010) and this present work confirms the observation. The naked-neck birds were found to be involved in pecking and had to be debeaked most frequently compared to the other phenotypes. The naked-neck and frizzle phenotypes consumed significantly ($P<0.05$) less feed and produced more eggs per kilogram of feed, confirming the observation by Mahrous *et al*. (2008) that the naked-neck and frizzle genes are associated with superior feed efficiency especially under stressful (high and humid) environments.

Table 1. Mean egg production performance of the crossbred naked-neck and frizzle phenotypes and the Lohmann brown layers

Parameters	Chicken phenotypes				
	Naked-neck	Frizzle	Normal	Lohmann	±SEM
Egg production/no.	267.2^{b}	260.4^{b}	229.4^{c}	294.8^{a}	3.5
Egg mass, g/HD	40.0^{b}	41.8^{b}	35.9^{c}	44.6^{a}	1.5
Feed intake, g/day	115^{a}	114^{a}	120^{b}	115^{a}	2.4
FCR	2.3^{a}	2.3^{a}	2.5^{b}	2.2^{a}	0.01
Rate of lay,%	70.7^{b}	68.9^{b}	60.7^{c}	78.0^{a}	2.7
Age @ 1st lay, days	129.3^{a}	127.0^{a}	128.0^{a}	121.0^{b}	1.9
Egg weight, g	55.7^{b}	54.4^{b}	49.4^{c}	59.8^{a}	2.3
Mortality,%	6.4^{a}	3.7^{c}	4.8^{b}	4.6^{b}	0.2

^{abc}Means in a row with different letters are significantly different at the 5% level.HDA is hen-day average, FCR is feed conversion ratio.

The superior performance of the naked-neck and frizzle phenotypes as compared to their normally feathered sibs is as a result of their reduced feathers, which provided a larger surface area for heat dissipation during periods of heat stress. Again, during periods of high temperature, because naked-neck or frizzle feathered birds are able to dissipate heat better, they tend to preserve more nutrient and energy that could otherwise have been used for heat dissipation, for other productive functions like egg production (Yalcin *et al.* 1997, Patra *et al.* 2002 and Rajkumar *et al*. 2011). This makes naked-neck and frizzle birds better able to tolerate low dietary protein as compared to the fully feathered counterparts. This is because normal birds use part of the protein for feather growth and development.

The egg quality characteristics of the phenotypes are presented and discussed under external (Table 2) and internal parameters (Table 3). The Lohmann brown layers laid significantly ($P<0.05$) bigger eggs than the third generation crossbred naked-neck and frizzle layers. This was because the former had been intensively selected for bigger egg sizes. With respect to the naked-neck and frizzle phenotypes, it was realized that there was a significant difference ($P<0.05$) on egg weight, with the naked-neck and frizzle phenotypes being superior to their normally feathered sibs even though they all segregated from the same flock. The results confirm the observation by Islam and Nishibori (2009) and Rajkumar *et al*. (2011) that under tropical environments naked-neck chickens laid bigger eggs than their fully feathered sibs. This observation also confirmed the results by Yushimura *et al*. (1997), Garces *et al*. (2001) and Younis and Galal (2006)

Table 2. Mean egg quality characteristics of the crossbred naked-neck and frizzle phenotypes and the Lohmann brown layers.

Parameters	Chicken phenotypes				
	Naked-neck	Frizzle	Normal	Lohmann	±SEM
Shape index, mm	79.8^{a}	80.0^{a}	80.1^{a}	76.0^{b}	2.8
Shell weight, g	6.2	6.5	6.3	6.9	0.5
Egg height, mm	50.4	50.4	50.4	50.6	0.1
Egg width	40.2	40.3	40.4	40.5	0.1
Shell thickness, mm	0.5^{a}	0.5^{a}	0.4^{b}	0.3^{c}	0.01
Shell ratio	12.6^{a}	12.8^{a}	12.1^{b}	11.6^{c}	0.6
Albumen weight, g	27.5^{a}	27.2^{a}	21.0^{c}	24.4^{b}	2.8
Albumen height, mm	9.6^{a}	9.8^{a}	8.6^{b}	8.4^{b}	0.5
Albumen ratio	49.4^{c}	53.9^{bc}	59.1^{a}	40.7^{d}	4.3
Yolk weight, g	18.6^{b}	18.2^{b}	14.9^{c}	21.4^{a}	1.9
Yolk height, mm	6.9^{b}	7.9^{a}	6.4^{b}	3.5^{c}	0.9
Haugh Unit,%	86.6^{a}	87.0^{a}	81.5^{b}	81.2^{b}	5.1
Yolk ratio	38.1^{b}	32.3^{c}	28.1^{d}	45.8^{a}	5.8

^{abc}Means in a row with different letters are significantly different at the 5% level.

that the presence of the naked-neck or frizzle genes in chickens resulted in bigger egg weights of layers reared under tropical conditions as compared with their normally feathered sibs.

Shape index is the ratio of the width to length of the egg. There was a significant phenotype effect on shape index (Table 2) with the crossbred naked-neck and frizzle birds being superior to the Lohmann brown counterparts. The significantly ($P<0.05$) higher shape indices observed for the crossbred layers in this study is a positive sign, an indication that the birds were laying eggs of more uniform shape and size. This attribute could be harnessed and explored to develop the birds into layers producing high quality eggs. Albumen and yolk weight are directly related to the weight of the egg, hence the bigger the egg the higher the albumen and yolk weight. In this present study, albumen weight was significantly affected by phenotype with the naked-neck and frizzle phenotypes laying eggs of better albumen weight (Table 2) than the normally feathered phenotypes. The albumen weights and yolk weight values obtained for the naked-neck and frizzle phenotypes were comparable to those obtained by others (Chartejee *et al*. 2006 and Yakubu *et al*. 2008) for crossbred naked-neck birds reared under hot and humid environments.

Haugh unit is the measure of albumen quality, which determines the quality of the egg. Albumen quality, which is the most important egg quality criterion, is determined by its height. Hence, the larger the albumen height the better the albumen quality would be. Albumen height varied between 1.5 mm for low quality eggs and 11.5 mm for good and fresh eggs (TSS 1980). Again, according to Haugh (1937), Haugh unit values of 70% and above represent good quality eggs; hence the crossbred phenotypes in this present study produced high quality eggs. According to Crawford (1992) albumen quality is influenced by both genetic and non-genetic factors such as breed, age of hen, length of storage and season of lay. The naked-neck and frizzle phenotypes producing eggs with better Haugh units as compared to the normal phenotypes was a confirmation of the observation by Yakubu *et al*. (2008) and that Na/na and F/f genotypes are found to produce high quality eggs due to the association of the genes with heat tolerance (Rajkumar *et al*. 2010b).

According to Fisinin *et al*. (1990) the thicker shell helps in preventing the damage during handling and also improves the keeping quality of the eggs. The mean shell thickness obtained in this present study was higher than values of 0.30–0.34 in crosses involving naked-neck, frizzle and normal birds by Nwachukwu *et al*. (2006), 0.31 in naked-neck and White Leghorn crosses by Padhi *et al*. (1998) and 0.35 in crossbred naked-neck and frizzle genotypes by Hagan (2010) in Ghana. The phenotypes with the Na/na and F/f genotypes laid eggs with significantly ($P<0.05$) thicker shells than their normally feathered sibs, confirming the observation by Islam and Nishibori (2009). Again, due to frequent handling of eggs, thick- shelled eggs are likely to survive poor handling; hence the developed naked-neck and frizzle birds could be a good replacement for the exotic breeds in future.

The introgression of the naked-neck (Na) and frizzle (F) genes into the Lohmann Brown has been found to improve egg production and quality. This was evidenced by the superior egg production performance of the naked-neck and frizzle phenotypes even under moderately warm environments, as compared to their sibs which were normally feathered. It is therefore suggested that future breed development strategies should involve the incorporation of the naked-neck and frizzle genes for high egg production and quality. Again, to ensure the realization of the full genetic potential of exotic birds that are to be reared under hot and humid environments, the incorporation of Na and F genes is therefore recommended.

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