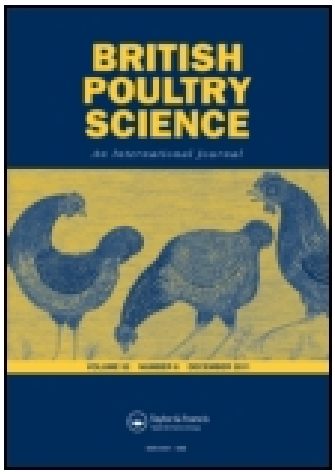

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Effect of the frizzle gene (*F*) on egg production and egg quality of laying hens kept in tropical villages

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Abstract 1. Two experiments were conducted to determine the influence of the frizzle gene (*F*) on the production and quality of chicken eggs kept on village farms in Ghana.
2. In the first experiment, 144 pullets, 72 each of *Ff* and *ff* pullets from F_1 birds, were compared in a randomised complete block design (RCBD), with three villages and 4 batches of hatch as blocks and the two feather genotypes (*Ff* and *ff*) as the treatments.
3. The *Ff* pullets were superior ($P < 0.05$) to their *ff* counterparts in terms of the number of eggs per clutch, egg mass and hen-housed and hen-d rates of lay, whereas age at first egg was lower ($P < 0.05$) in *ff* layers compared to *Ff* ones.
4. The eggs of the F_1 heterozygous frizzle (*Ff*) layers had higher values for albumen height, Haugh unit and yolk height compared with eggs from their normal feathered counterparts.
5. In the second experiment, 144 pullets, 48 each of *FF, Ff* and *ff* pullets, were compared in a RCBD, with three villages and 4 batches of hatch as blocks and the two feather genotypes (*Ff* and *ff*) as the treatments.
6. The F_2 birds with genotypes *Ff* and *FF* were better than their *ff* counterparts in terms of the number of eggs per clutch, egg mass, and hen-housed and hen-d rates of lay. Age at first egg was significantly lower in *ff* layers compared to *FF* and *Ff* ones.
7. The Haugh unit value was higher in the homozygous and heterozygous frizzles compared to the normal feathered ones.
8. The presence of the frizzle gene (*F*) in egg type chickens led to an improvement in egg production and egg quality traits in village chickens, and the cross-breeding scheme evaluated in this project could contribute to improved productivity.

INTRODUCTION

The frizzle phenotype is caused by a single autosomal incompletely dominant gene, *F* (Hutt, 1930; Landauer and Dunn, 1930). According to Anonymous (2007), chicks possessing the frizzle gene appear to be normally feathered when they are hatched, but the wing feathers soon start to grow and turn outwards. The frizzling gene is a feather structure gene (Horst, 1988) that causes a reduction in tropical heat stress by improving the bird's ability for convection, resulting in improved feed conversion and better performance (Merat, 1990). Benedict *et al.* (1932) found a considerable increase in energy metabolism for birds possessing the frizzle gene, implying that they will respond differently from normal feathered birds to high temperatures. The frizzle gene has favourable effects on production traits such as egg number, egg weight, egg mass, body weight and productivity index (Somes, 1988; Mathur, 2003).

Egg laying strains used in Ghana were bred and selected under temperate and optimal conditions and are therefore not able to perform optimally when raised under tropical and suboptimal conditions. In the advent of rising feed costs globally and more especially in Ghana, lower rates of lay make an egg production enterprise a non-lucrative venture

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for small- and medium-scale farms. Developing a breed that is well adapted to tropical and suboptimal conditions is therefore crucial if an egg production enterprise could be economically viable for small- and medium-scale farmers in the tropics who raise laying birds under suboptimal conditions. This could be achieved by either breeding and selecting a breed under tropical and suboptimal conditions for physiological adaptation or introducing (a) gene(s) into existing breeders for genetic adaptation. Since the frizzle gene has been found to improve meat and egg production in birds under tropical conditions (Adomako *et al.*, 2012), it is hypothesised that introducing the frizzle gene into existing egg laying strains would provide genetic adaptation under tropical and suboptimal conditions. The objective of this study therefore was to investigate the effect of the frizzle gene on the production and quality of eggs produced by chickens raised under tropical and suboptimal conditions.

MATERIALS AND METHODS

Location and duration of the experiment

The experiment was carried out at the Department of Animal Science, Kwame Nkrumah University of Science and Technology (KNUST), Kumasi (altitude 261.4 MSL, latitude 06°41′N and longitude 01°33′W) (Meteorological Services Department, Kumasi), and three selected villages/towns within the Asante-Akim South District of the Ashanti Region, Ghana, namely, Yawkwei, Juaso and Nkwanta. These villages were selected based on convenience and the availability of reliable chicken keepers. The experiment lasted for 20 m.

Experimental birds

Five indigenous frizzle males (25–52 weeks old) obtained from local towns (Yawkwei, Juaso and Nkwanta within the Asante-Akim South District of the Ashanti Region) and 50 Lohmann layers (40 weeks old) obtained from a commercial poultry farm in Ghana (Akate Farms and Trading Company Limited, Kumasi) were used for initial mating at the Department of Animal Science, KNUST. The indigenous frizzle males were feet-washed with disinfectant to avoid the spread of any possible infection. All the birds were housed separately for 2 weeks within which the birds were dewormed and vaccinated with Newcastle disease vaccine (Newcavac, Nobilis). All birds were weighed individually. The indigenous frizzle males were then crossed with the Lohmann layers in the ratio of 1:8.

Experimental design

The experimental design was a randomised complete block design (RCBD), with three villages and 4 batches of hatch as blocks and the two feather genotypes (*Ff* and *ff*) as the treatments. The total number of birds was 144 with 48 in each block and 24 for each feather genotype.

Management

The birds were allocated to 4 pens in a deep litter system for 3 months. They were fed on layer mash *ad libitum* (175 g CP/kg and 11.3 MJ ME/kg). They were also provided with freshwater *ad libitum*. Each of the 4 pens had two laying boxes measuring 15 cm × 15 cm. Eggs were collected twice daily, labelled and stored for not more than 7 d at room temperature before incubation. The birds were dusted with Malathion poultry dust (Kepro, Netherlands) against lice, soft ticks and mites. They were also dewormed and given Pen Strip (Kepro, Netherlands) as an antibiotic and vitamin source. The average monthly temperatures within the pens during the experimental period ranged from 21.6 to 25.9°C.

Incubation

The eggs were selected for artificial incubation by discarding very small (<40 g) or very large eggs (>70 g), broken shells, blood-stained or dirty eggs. The eggs were incubated and hatched at the hatchery of Akropong Farms, a commercial hatchery in Kumasi. The incubation started 10 d after introducing the females to males and continued weekly for 5 consecutive weeks in a multistage machine. The entire management of birds, egg production and collection and incubation was repeated in birds obtained from the second filial (F_2) generation.

Chick rearing

After hatching, each batch of chicks was brooded for 6 weeks. The chicks were wing-tagged and weighed individually. Glucose was administered via the drinking water to supplement their energy. Commercial chick mash from AGRICARE Ltd, Ghana (195 g CP/kg and 11.7 MJ ME/kg) and fresh drinking water were given *ad libitum*. The chicks were vaccinated against Newcastle and Gumboro diseases. Coccidiostat, antibiotics and vitamins were administered through the drinking water during the first month.

Parents for the second-generation birds

Twenty heterozygous frizzle males and 120 heterozygous frizzle females were selected after the

brooding period and were later mated *inter se* to produce the second generation (F_2). The males were kept separately from the females. The birds were fed commercial grower mash from AGRICARE Ltd, Ghana (150 g CP/kg and 11.1 MJ ME/kg) at 6 weeks of age and layer mash from AGRICARE Ltd, Ghana at week 17 (175 g CP/kg and 11.3 MJ ME/kg). Feed and freshwater were given *ad libitum*. They were vaccinated against fowl pox and Newcastle (Newcavac) diseases. Deworming and vitamin supplementation were done after every 3 months via their drinking water. Four weeks after the first egg had been laid, the males were introduced to the females in a ratio of 1:10, and then collection of eggs for incubation took place 2 weeks thereafter. Chicks were reared up to 6 weeks as described above. After the 6th week, these second filial generation (F_2) birds were transferred to the three villages as mentioned earlier.

Mating

This mating involved indigenous heterozygous frizzle and Lohmann commercial layers, and also F_1 heterozygous frizzles in the first and second mating, respectively. The offspring in both F_1 and F_2 were made of full-sib and half-sib sire families, but due to the difficulty in separating these, they were reared together as sire families. The two crosses are described in Figure 1.

Selection and training of chicken keepers

The chicken keepers used for the study were selected prior to the study. The selection was done during an earlier survey to assess the performance of local chickens in Ghana. One keeper was selected from each of the three villages (Yawkwei, Juaso and Nkwanta). The selection was based on the ability to read and write and also on the reliability and interest in keeping the local birds. Each keeper made a hen-coop or prepared a place for the birds and these were inspected prior to the transfer of the birds. A top-loader weighing scale was given to each keeper for periodic weighing. They were trained on simple poultry management practices such as feeding and giving water under sanitary conditions, culling, litter changing, maintaining a clean coop, recording age at first egg and counting of the number of eggs per clutch.

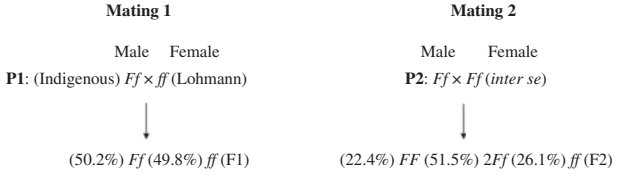

Figure 1. *Genetic crossing to produce F_1 and F_2 generations in the breeding programme.*

Transfer of chicks and rearing at the villages

In the F_1 generation, heterozygous frizzle (*Ff*) and normal feathered (*ff*) birds were given to farmers in the villages. In the second transfer, F_2 homozygous frizzle (*FF*), heterozygous frizzle (*Ff*) and normal feathered (*ff*) birds were involved. The chicks were brooded at the Poultry Section of the Department of Animal Science, KNUST, and transferred to the three selected villages at the end of the 6th week. One chicken keeper was selected in each of the three villages. Each keeper was first given 48 F_1 birds including 24 frizzles and 24 normal feathered birds. A total of 144 F_1 birds were used in the first trial. For the second-generation birds (F_2), each keeper was given the same number of birds (48), 16 from each of the three genotypes (*FF, Ff* and *ff*). A total of 144 F_2 birds were used in the second trial. The birds were raised under intensive system and were given commercial grower (7–16 weeks) and layer diets (from 17 to the end of the experiment) *ad libitum*.

Egg production

The average age at first egg was measured as the age at which pullets from each genotype laid their first eggs. The number of eggs per clutch was estimated by counting the number of consecutive eggs laid by individual layers before a pause in laying. All the eggs laid were recorded daily for a 6-m period. Egg production was calculated in terms of rate of lay i.e. percentage of the total number of eggs produced divided by the total number of hens alive per day (hen-d egg production), or divided by the total number of hens housed (hen-housed egg production). These methods were applied in F_2 birds.

Egg size and egg mass

Relative egg size was expressed as the ratio of egg weight to the body weight of the layer, and the egg mass was calculated by multiplying the hen-d rate of lay by the average egg weight.

Mortality

Mortality was calculated as the percentage of birds that died between one-d-old and the end of the experiment in the F_1 and F_2 generations.

Egg quality analysis

The egg quality analysis was conducted twice for F_1 heterozygous frizzle and normal feathered birds from each of the three villages. In each

village, 25 eggs from each phenotype were selected at random and used for the analysis. The analysis was done at the beginning of lay, during the 12th week after the first egg and in the 24th week after the first egg. In the F_2 generation, data were taken on homozygous frizzle (*FF*), heterozygous frizzle (*Ff*) and normal feathered (*ff*) birds in each village.

Statistical analysis

Analysis of variance (ANOVA) was performed using GenStat software (2007). Means with significant differences were separated by the least significant difference test at $P < 0.05$.

The data on F_1 and F_2 birds were analysed using the following linear model:

$$Y_{ijkl} = \mu + V_i + G_j + B_k + e_{ijkl},$$

where Y_{ijkl} = observation for a given variable, μ = overall mean common to all observations, V_i = environmental effect due to the ith village (i = 1, 2, 3), G_j = genetic effect due to jth phenotype (j = 1, 2), B_k = environmental effect due to the kth batch of hatch (k = 1, 2, 3, 4) and e_{ijkl} = random error effects peculiar to each observation (e = N(0,1)). Statistical differences were reported at $P < 0.05$ unless otherwise stated.

RESULTS AND DISCUSSION

Egg laying performance of F_1 and F_2 birds

The F_1 *Ff* layers took longer to come into lay compared to the *ff* layers (Table 1). Body weight at first egg was statistically higher in frizzle birds compared to the normal feathered ones in the F_1, but the same trait did not differ significantly between the two genotypes in the F_2 birds (Table 2). However 24 weeks after the first egg and also at the end of the experiment at 60 weeks, the body weight values did not differ significantly between the two phenotypes in both the F_1 and F_2 birds. Layers possessing the frizzling gene were superior ($P < 0.05$) to their *ff* counterparts in terms of the number of eggs per clutch, egg mass and rates of lay (hen-housed and hen-d) in the F_1 birds. However, egg size and mortality in the F_1 birds were not significantly different between the *Ff* and *ff* genotypes (Table 2).

Normal feathered (*ff*) F_2 birds had a lower age at first egg than the homozygous frizzle (*FF*) and the heterozygous frizzle (*Ff*) birds. The number of eggs per clutch was significantly higher for *Ff* birds followed by *FF*, and *ff* birds had the lowest clutch size. Hen-housed and hen-d rate of lay and egg mass were higher in the homozygous and heterozygous frizzle genotypes compared with those with the normal feathered genotype. Egg size and mortality did not differ significantly between layers from the three genotypes (Table 3).

The higher egg number in frizzle birds may be caused by the genotype. This has been reported earlier in which under high ambient temperature conditions (above 27°C), the frizzle gene had favourable effects on production traits such as egg number and productivity index

Table 1. *Laying performance of F_1 heterozygous (Ff) frizzle and homozygous normal feathered (ff) birds*

Trait	*Ff*	*ff*	SEM
Age at first egg (d)	147.3[a]	144.5[b]	0.425
Body weight at first egg[1] (g)	1583[a]	1467[b]	7.4
Body weight at 24 weeks[2] (g)	1644	1639	4.5
Body weight at the end of trial (g)	1657	1653	6.6
Number of eggs per clutch	16.8[a]	15.2[b]	0.32
Egg size (as % of body weight)	3.50	3.80	3.310
Egg mass (g)	26.5[a]	24.9[b]	0.39
Hen-housed rate of lay (%)	54.8[a]	51.5[b]	0.35
Hen-d rate of lay (%)	60.3[a]	57.9[b]	0.41
Mortality (%)	11.1[a]	9.7[b]	–

[a,b]Mean values within the same row sharing a common superscript letter are not statistically different at $P < 0.05$.

[1]Body weight at the beginning of the egg quality analysis.

[2]Body weight at 24 weeks after first egg (at the end of the egg quality analysis).

Table 2. *Laying performance of F_2 homozygous (FF), heterozygous (Ff) frizzle and normal feathered (ff) birds*

Trait	*FF*	*Ff*	*ff*	SEM
Age at first egg (d)	133.1[a]	133.57[a]	130.0[b]	1.43
Body weight at first egg[1] (g)	1651	1670	1666	6.6
Body weight at 24 weeks[2] (g)	1672	1681	1685	8.2
Body weight at the end of trial (g)	1761	1776	1768	15.7
Number of eggs per clutch	16.8[b]	17.5[a]	15.5[c]	0.30
Egg size (% of body weight)	3.52	3.57	3.61	2.480
Egg mass (g)	31.9[a]	32.5[a]	29.5[b]	0.52
Hen-house rate of lay (%)	59.1[a]	59.9[a]	55.7[b]	1.24
Hen-d rate of lay (%)	63.9[a]	64.3[a]	60.8[b]	0.83
Mortality (%)	10.42	8.33	10.42	–

[a,b,c]Mean values within the same row sharing a common superscript letter are not statistically different at $P < 0.05$.

[1]Body weight at the beginning of the egg quality analysis.

[2]Body weight at 24 weeks after first egg (at the end of the egg quality analysis).

Table 3. *Egg quality parameters of F_1 heterozygous (Ff) frizzle and homozygous normal feathered (ff) birds*

Trait	*Ff*	*ff*	SEM
Albumen height (mm)	6.60[a]	5.91[b]	0.200
Haugh unit (%)	80.3[a]	77.2[b]	0.98
Shell thickness (mm)	0.31	0.31	0.002
Yolk height (mm)	15.5[a]	15.0[b]	0.22
Yolk diameter (mm)	39.6	39.4	0.51
Yolk colour score	8.24	8.09	0.094

[a,b]Mean values within the same row sharing a common superscript letter are not statistically different at $P < 0.05$.

compared with normal feathered birds (Somes, 1988; Mathur, 2003).The frizzled gene curls the feathers of chickens and this enhances heat loss. This in turn improves feed efficiency and egg production (Somes, 1988; Mathur, 2003). Horst (1987) found that under high temperatures, the frizzle gene caused an increase in egg numbers. Sharifi *et al.* (2010) stated that under high ambient temperatures, the frizzle gene in heterozygous form (*Ff*) had a positive effect on laying performance, but added that this advantage was not significant under moderate temperatures. Missohou *et al.* (2003) studied frizzle, sex-linked dwarfism, normal feathered, normal size and combined frizzle and sex-linked dwarfism birds under Senegalese natural conditions and found that the interaction between the two genes was positive for egg number. Sharifi *et al.* (2010) showed that the presence of the *F* gene especially in the homozygous dominant form (*FF*) delays age at first egg as shown in the current research.

The frizzle gene may have a positive influence on survivability. Mortality in the normal birds (*ff*) was better than the frizzles in the F_1 generation were associated with inadequate heating during brooding resulting from electrical power failures that decreased brooding temperatures (heat from an 100-watt bulb), and the frizzle birds suffered more because of their curled feathers. In comparison with the F_1, in the F_2 the positive effect of the frizzle gene on survivability became pronounced and confirmed what has been reported by others (Njenga, 2005; Adomako *et al.*, 2013).

The heterozygous frizzle birds had a higher average number of eggs per clutch than the homozygous frizzles under the conditions of this study (average ambient temperature of 27°C). Their feathers were extremely curled and parts of their skin were exposed leading to damaging pecking among themselves and by other phenotypes. This slightly affected their feed intake and feed utilisation and eventually the number of eggs per clutch. Furthermore, since the heterozygous frizzle birds performed better than their two homozygous counterparts (*FF and ff*), this result could be due to overdominance.

Egg quality of F_1 and F_2 birds

The eggs of the F_1 heterozygous frizzle (*Ff*) layers had higher albumen height, Haugh unit and yolk height compared with eggs from their normal feathered counterparts (*ff*). However, shell thickness, yolk diameter and yolk colour score values were not significantly different between eggs from layers of the two genotypes (Table 4).

Eggs from the layers of homozygous frizzle, heterozygous frizzle and normal feathered F_2 birds did not differ in albumen height, yolk height, yolk diameter and yolk colour score (Table 4). Haugh unit value was higher in eggs from the layers of the homozygous and heterozygous frizzle genotypes compared to those from the layers of the normal feathered ones. Eggs from layers with homozygous frizzle phenotype had a higher shell thickness than the heterozygotes which also had significantly thicker shells than the normal feathered ones (Table 4).

Table 4. *Egg quality parameters of F_2 homozygous (FF) and heterozygous (Ff) frizzle and normal feathered (ff) birds*

Trait	*FF*	*Ff*	*Ff*	SEM
Albumen height (mm)	7.39	7.23	7.11	0.184
Haugh unit (%)	79.4[a]	79.2[a]	76.9[b]	1.14
Shell thickness (mm)	0.35[a]	0.34[b]	0.32[c]	0.003
Yolk height (mm)	16.6	16.6	16.2	0.21
Yolk diameter (mm)	40.5	40.3	40.0	0.83
Yolk colour score	8.27	8.00	7.80	0.211

[a,b,c]Means with different superscripts are significantly different ($P < 0.05$).

F_1 frizzle (*Ff*) birds had higher values for albumen height, Haugh unit and yolk height than the normals (*ff*) while in the F_2 generation, the frizzles (*FF, Ff*) had higher values in Haugh unit and shell thickness than the normals (*ff*) which indicate that the frizzle gene has a positive influence on egg quality. According to Isikwenu *et al.* (1999), Haugh unit is one of the best indicators of internal egg quality, and the higher the Haugh unit the more desirable the egg quality is. The superior Haugh unit in the frizzle phenotype in this study is contrary to the finding of Missohou *et al.* (2003) who studied frizzles, sex-linked dwarfs, normal feathered and normal-sized birds and combined frizzle and sex-linked dwarf birds under Senegalese conditions and found that the frizzle gene did not significantly influence egg quality. However, Mathur (2003) studied the frizzle phenotype under natural conditions in different countries and stated that under fluctuating high ambient temperature conditions (above 27°C), there are large differences in the performance of frizzle birds in terms of productivity index at different locations.

The frizzle birds performed better in egg quality in this study because the frizzle gene may have a positive linkage with genes controlling egg quality (Benedict *et al.*, 1932). Additionally, the high ambient temperatures during the experiment could have caused stress in laying birds and affected egg formation and egg quality negatively. However, birds showing the frizzle phenotype have been found to respond differently from normals under heat stress due to their curled feathers which help in thermo-regulation to reduce heat stress and alleviate its eventual consequence on egg formation and quality. The homozygous frizzles had higher values in shell thickness than the heterozygous ones, suggesting

that the frizzle gene has a positive influence on eggshell thickness. The contrary results reported by Sharifi *et al.* (2010) might have been due to the variability in the performance of frizzle genotypes at different locations (Mathur, 2003).

Results on yolk diameter and yolk colour score showed that the frizzle genotypes do not influence these traits significantly. This is in agreement with the findings of Missohou *et al.* (2003). However, the most important egg quality traits are the Haugh unit and shell thickness which the frizzle phenotypes influenced positively in this study. It can therefore be concluded that the presence of the frizzle gene (*F*) in egg type chickens improves egg production and egg quality in the village conditions represented in these experiments.

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