

INFLUENCE OF SOURCES AND LEVELS OF INORGANIC AND ORGANIC IRON ON EGG QUALITY IN DOMINANT DARKSHELL DS109

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ABSTRACT

With the ever-increasing societal pressure on animal welfare, there is a gradual transition to alternative housing systems. A new development in the field of breeding of laying hens is the breeding of hybrids that can be used in alternative and extensive conditions on the parameters of eggshell colour and uniformity. Different feeding strategies are being tested to maintain the desired colour and uniformity, but these must also be tested in relation to egg quality parameters. The aim of this study was to test the effect of different iron sources and levels on egg quality parameters in the hybrid Dominant Darkshell DS109, which is specifically bred to produce table eggs with dark to chocolate brown eggshell colour. The study included 240 laying hens whose diets were supplemented with two different sources and levels of iron. Our

results showed a significant effect ($p < 0.001$) of increased organic and inorganic iron levels on the parameters of eggshell thickness (0.403; 0.402 mm vs. 0.395 mm), eggshell weight (5.7 and 5.6 vs. 5.4 g) and eggshell percentage (9.4 and 9.3 vs. 9.1 g). The results also indicate a positive effect of elevated 191 mg/kg Fe-Gly on egg weight (64.1 g vs 62.7 g). There was an effect ($p < 0.05$) of inorganic form of iron on yolk proportion parameter throughout the laying period.

Keywords: laying hens; ferrous sulfate; eggs quality; iron chelate, eggshell quality

INTRODUCTION

In the European Union, with increasing pressure on animal welfare, there is a gradual shift to various non-cage technologies for housing laying hens, which is also reflected in the field of breeding. In addition to the breeding of laying hens for alternative technologies, a new development in the world is the breeding of layers for eggshell colour to increase their attractiveness to both breeders and consumers. The breeding of laying hens for eggshell colour is taking place in hybrids intended for alternative housing systems, which presents the possibility of their increased use in the future. This trend is confirmed by statistics showing that by the end of 2023, almost 61 % of laying hens reared in the European Union (EU Commission, 2024) were housed in non-cage technologies.

One of the main objectives of breeding for eggshell colour is the requirement to maintain uniformity and persistence of eggshell colour throughout the laying period. Various nutritional strategies are used to enhance the production of pigments that cause eggshell colour. The most promising strategy appears to be the use of various macro and

microelements. In addition to testing these feed additives on eggshell colour, it is necessary to test their effect on egg quality parameters.

Therefore, this study is devoted to evaluating the effect of supplementation of inorganic and organic forms of iron at different levels on egg quality in the hybrid Dominant Darkshell DS109 intended mainly for alternative housing systems, extensive conditions and backyard production.

MATERIAL AND METHODS

A total of 240 laying hens of the genotype Dominant Darkshell DS109 at the age of 22 weeks were included in the experiment. The laying hens were randomly divided into four groups at this age. Each group had four replicates represented by a total of 60 laying hens. Laying hens were housed in 3-tier battery cages in the experimental facility.

Diet

All experimental groups were fed once a day the same feed mixture, which differed only in the source and amount of iron depending on the premix used, which was added to the feed mixture at 3% concentration. The distribution of laying hens into groups according to the premix used is shown in Table 1. Mixture N1 was fed until the 45th week of laying hens' age at a daily dose of 125 g/head/day with nutrient content of 11.17 MJ of ME and 165.03 g of crude protein per kg/mixture. Mixture N2 was fed until the 72nd week at a dose of 135 g/head/day with nutrient content of 11.7 MJ of ME and 151.51 of crude protein in accordance with the requirements of the breeder of the hybrids. Water were supplied ad libitum. The lighting regime consisted of 14 hours of light and 10 hours of darkness.

Table 2. Schematic division of laying hens into groups according to the premix used

Genotyp of hybrids	Name of group	Type of iron level	N1	N2	Number of repetition	Number of laying hen in repetition
			premix until 45 week	premix from 45 week		
Darkshell DS109	D-K- (K)	Standard	KON	KON	4	60
Darkshell DS109	D-K- O	increased ORG 45t	KON	ORG2	4	60
Darkshell DS109	D-O	increased ORG	ORG2	ORG2	4	60
Darkshell DS109	D-A	increased ANORG	ANORG2	ANORG2	4	60

Layers in the control group (D-K) were supplemented with an inorganic source, specifically ferrous sulphate monohydrate, $\text{FeSO}_4 \cdot \text{H}_2\text{O}$ was used in the premix at a level of 131 mg/kg. The premixes for the organic and inorganic groups of layers contained 45% more iron than the premix for the control group of layers. For the inorganic group (D-A), the same source of iron was used as for the control group, but its level was increased to 191 mg/kg of the feed mixture. Laying hens in the organic group (D-O) were supplemented with an organic source of iron, ferric chelated glycine Fe-Gly, at a level of 191 mg/kg of compound feed. Layers in the D-K-O group were fed the control diet until week 45. From week 45 they were supplemented with the same diet as the organic group.

Egg quality

Egg quality parameters were monitored at regular four-week intervals from 24 - 72 weeks of age of the laying hens. The analysed eggs were

collected on the same day each time to avoid evaluating eggs from the same hen twice within the evaluated week of age.

The egg quality parameters monitored included egg weight, eggshell strength, yolk weight, yolk colour, eggshell weight and eggshell thickness. From the data obtained, the albumen weight, % albumen proportion, % yolk proportion, % eggshell proportion of egg weight were calculated.

Each egg was weighed to determine the egg, yolk and eggshell weight. Each egg was weighed on a KERN KB 1000-2 electronic balance (Germany) to the nearest 0,1 g. The above-mentioned scales were also used to determine yolk weight and eggshell weight. Eggshell strength was determined by the destructive method using an Egg Force Reader (Orka Food Technology, Ltd.). Eggshell thickness was measured on washed and dried eggshells, including the eggshell membranes, at three points - the equator, the sharp and the blunt end of the egg. Measurements were made using a micrometre. Yolk colour was assessed subjectively using a 15-grade Yolk Colour Fan (Hoffman La Roche).

Statistical evaluation

The individual egg quality characteristics were described using the mean. Two-factor analysis of variance was used to evaluate the effect of genotype, age and their interaction on each of the parameters studied. Scheffe's test was used to subsequently test the significance of differences between means for the group factor. Statistical evaluation was performed using Unistat 5.1 software (Unistat Ltd., ENGLAND).

RESULTS AND DISCUSSION

There was a significant effect ($p < 0.001$) of source and level of iron on eggshell weight, eggshell thickness and eggshell proportion there was a significant effect ($p < 0.001$) of source and level of iron. Significant effect of source and level of iron supplemented in the feed was also found for the parameters egg weight, albumen weight, colour and yolk proportion. The effect of age on all egg quality parameters was also demonstrated. The parameters eggshell thickness and yolk colour showed an effect ($p < 0.001$) of the interaction of group and age.

Table 1 shows the effect of the level and source of iron in each experimental group on egg quality parameters over the entire laying period. Over the period of interest, the heavier eggs (61.2 g vs. 60.6 g and 60.2 g) were conclusively ($p < 0.01$) produced by the D-O group layers, which is also confirmed by Xie et al. (2019) for Hy-Line White hybrids when supplementing the diet with an organic iron source, specifically Fe-Gly, which was also used in our experiment. Also Sharlak et al. (2021) found a significantly ($p < 0.05$) higher egg weight in the iron supplemented compared to the non-supplemented experimental group (60.5 g vs 55.3 g). However, no significant difference was found between the iron supplemented groups (Sharlak et al., 2021). The findings of Sharlak et al. (2021) are in agreement with those of Park et al. (2004).

Table 2. Egg quality parameters for the whole laying period

Parameter	Units	Group			Factor		
		D-K	D-A	D-O	Group	Age	Group x Age
Egg weight	g	60,2 ^b	60,6 ^{ab}	61,2 ^a	<0.01	<0.001	NS
Eggshell strength	N	36,5	38,0	38,0	NS	<0.001	NS
Yolk weight	g	16,4	16,6	16,5	NS	<0.001	NS
Albumen weight	g	38,3 ^b	38,4 ^b	39,0 ^a	<0.01	<0.001	NS
Eggshell weight	g	5,4 ^b	5,6 ^a	5,7 ^a	<0.001	<0.001	NS
Eggshell thickness	mm	0,395 ^b	0,402 ^a	0,403 ^a	<0.001	<0.001	<0.001
Yolk colour	-	11,6 ^a	11,9 ^b	11,7 ^{ab}	<0.05	<0.001	<0.001
Yolk proportion	%	27,3 ^a	27,3 ^a	26,9 ^b	<0.05	<0.001	<0.05
Albumen proportion	%	63,7	63,4	63,8	NS	<0.001	NS
Eggshell proportion	%	9,1 ^b	9,3 ^a	9,4 ^a	<0.001	<0.001	NS

NS means no statistically significant differences ($p > 0.05$); a,b,c means different letters indicate statistical significance ($p < 0.05$)

Egg weight in the D-O group was affected by the conclusively ($p < 0.01$) higher egg albumen weight also in this group (39 g vs. 38.4 g and 38.3 g). The weight, thickness and proportion of eggshell was significantly higher ($p < 0.001$) in the groups whose diet contained elevated levels of iron. Similar conclusions were reached by Sharlak et al. (2021). For the parameter of yolk colour, there was a significant difference in the D-K group compared to the other groups. This finding is in contrast to the study of Seo et al. (2010), who found no conclusive difference ($p > 0.05$) between groups of Hy-Line Brown laying hens given iron in the form of iron-soy proteinate. Also, Xie et al. (2019)

showed no effect of iron source and level on yolk colour in eggs from HY-Line White-produced layers, although the same iron source was used in the Fe-Gly group as in our D-O group.

In the parameter yolk proportion, a significantly higher ($p < 0.001$) proportion of yolk was found in groups D-K and D-A. For the characteristics of eggshell strength, protein percentage, there was no significance ($p > 0.05$) between the tested groups. Our findings correspond in the case of eggshell strength parameter with the results of Xie et al. (2019) and Cao et al. (2023). In contrast, authors Park et al. (2004) found that the effect of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ at 200 ppm increased eggshell strength in ISA Brown, which is inconsistent with our results. Also the study of Tu, Zou and Tang (2004) confirmed the beneficial effect of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ and Fe-Gly on eggshell strength as well as weight and thickness in Roman hens.

Effect of elevated and organic iron levels on egg quality after 45. week of laying hens' age

Table 2 shows the comparison of the control group with the group in which the inorganic to organic iron source and iron levels were changed in the second phase of the laying period, i.e. after 45 weeks of age. Table 2 shows that the increased amount of iron in the organic form from 45 weeks of age onwards had a conclusive effect on egg weight, albumen, eggshell, thickness eggshell thickness and yolk proportion.

Table 3. Egg quality parameters after 45 weeks of age after changing the source and increasing the iron level

Parameter	Units	Group		Factor		
		D-K-K	D-K-O	Group	Age	Group x Age
Egg weight	g	62,7 ^b	64,1 ^a	<0.01	<0.001	NS
Eggshell strength	N	35,4	33,8	NS	<0.05	NS
Yolk weight	g	17,8	17,9	NS	<0.001	<0.05
Albumen weight	g	39,2 ^b	40,4 ^b	<0.01	NS	NS
Eggshell weight	g	5,6 ^b	5,8 ^a	<0.01	<0.001	NS
Eggshell thickness	mm	0,403 ^b	0,413 ^a	<0.01	<0.001	<0.001
Yolk colour	-	11,8	11,9	NS	<0.001	<0.01
Yolk proportion	%	28,5 ^a	28,0 ^b	<0.05	<0.001	<0.01
Albumen proportion	%	62,5	62,9	NS	<0.001	<0.05
Eggshell proportion	%	9,0	9,1	NS	<0.01	NS

NS means no statistically significant differences ($p > 0.05$); a,b means different letters indicate statistical significance ($p < 0.05$)

Characteristics such as egg, yolk and eggshell weight, eggshell thickness, yolk colour, yolk and eggshell proportion were significantly influenced by the age of the laying hens ($p < 0.001$). The characteristics of yolk proportion were also statistically significantly influenced by the age of the laying hens eggshell ($p < 0.01$) and eggshell strength ($p < 0.05$). Kraus et al. (2020) also conducted an experiment with Lohmann Brown Classic and Hisex Brown laying hens enrolled in the experiment at 46 weeks of age and found a conclusive effect of age on egg quality parameters.

For the eggshell thickness parameter, a statistically significant ($p < 0.001$) effect was found interaction of group and age on this parameter of egg quality. Significant effect of the interaction of group and age of laying hens was also confirmed for yolk weight and egg albumen proportion ($p < 0.05$) and also for the parameters colour and yolk proportion ($p < 0.01$).

For the egg weight characteristic, a significant difference ($p < 0.01$) was found, with the D-K-O group laying heavier eggs (64.1 g) than the D-K-K group (62.7 g), which was influenced by the higher eggshell weight (5.8 g vs. 5.6 g) of the D-K-O group compared to eggs produced by D-K-K group hens. Similarly, these eggs also had a thicker eggshell thickness (0.413 mm vs. 0.403 mm) compared to eggs from D-K-K group layers. On the other hand, D-K-K hens produced eggs with a significantly ($p < 0.05$) higher yolk weight (28.5 g vs. 28.0 g) than those from the D-K-O group.

CONCLUSION

In our experiments, a significant effect ($p < 0.001$) of increased supplementation with inorganic and organic iron sources on eggshell quality parameters was found, namely eggshell thickness (0.403; 0.402 mm vs. 0.395 mm), eggshell weight (5.7 and 5.6 vs. 5.4 g) and eggshell percentage (9.4 and 9.3 vs. 9.1 g). It was also found that significantly ($p < 0.01$) heavier eggs (61.2 g vs. 60.6 and 60.2 g) were produced by the D-O group layers due to the significantly higher egg white weight (39 g vs. 38.4 and 38.3 g). The effect ($p < 0.05$) of iron source and level on the qualitative parameters of egg colour and yolk proportion was also found. Increase in organic form of iron after 45 weeks of age had a significant effect ($p < 0.01$) on egg weight (64.1 vs.

62.7 g), eggshell thickness (0.413 vs. 0.403 mm), eggshell weight (5.8 vs. 5.6 g).

ACKNOWLEDGEMENT

The project was supported by the grant 235/9510/SV2220181.

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