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Research Article

Determination Effects of Slow (K) and Fast (k+) Feathering Gene on Egg Production and Hatching Traits in Laying Hens

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Abstract

Background and Objective: The sex determination of pure lines of one-day-old chicks was achieved by vent-sexing which has some challenges and the risks, so it was decided that the sex determination of chicks by the feathering rate should be used. In the present study, fast- and slow- feathering of RIR, BAR and L54 genotypes were developed and investigated the effect of feathering rate on egg production and hatching traits. **Materials and Methods:** Data of body weight and age at first egg, egg number and egg weight up to 43 weeks age were collected from total of 10717 hens and analysed. The following genetic parameters were estimated from a three generation selection study: Age at first egg (AFE), Body Weight at First Egg (BWFE), Egg Number (EN) and Egg Weight (EW) by the Restricted Maximum Likelihood (REML) procedure. Statistical evaluation was performed using one-way analysis of variance and genetic parameters were estimated by the restricted maximum likelihood (REML) procedure for egg production and hatching traits. **Results:** Mean egg production trait values were determined as follows: 149.42-161.29 days (AFE), 1562.90-1748.10 g (BWFE) 117.54-129.36 eggs (EN) and 54.70-58.93 g (EW). Statistically significant differences ($p < 0.01$) were observed between the genotype groups according to the fertility, embryonic mortality and hatchability values. Predicted heritability's for egg production traits ranged from 0.11-0.59 and it was determined that AFE was positively correlated with BWFE except for L54-F and L54-S. The EN was negatively correlated with AFE, BWFE and EW in all genotypes. The AFE was negatively correlated with EW except for RIR-F and L54-S. The BWFE was positively correlated with EW except for RIR-F and RIR-F genotypes. **Conclusion:** The results of this study showed that one-day-old hybrid chicks from fast-and slow-feathering parent genotypes can be accurately sexed by feathering rate. It was observed that fast-feathering genotypes were generally higher than slow ones with respect to egg production and hatching traits, with the exception of egg weight.

Key words: Egg production, feathering rate, hatching traits, heritability, genetic correlations

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Poultry sector has developed very fast in recent decades and with this development egg producers prefer only female chicks, so sex determination has enormous commercial value. Several methods including feather sexing have been using practically for the sex determination of chicks. Superfluous day-old male chicks of laying hen strains have no commercial value so they are culled after sex determination¹. Luo *et al.*² suggested that high level of Prolactin receptor (PRLR) was caused the slow feathering genotype. They stated that growing rate and immunity system of fast feathering chickens were better than those of slow featherings. Sex determination according to feathering rate based on differences in the rate of feather growth provides a convenient and inexpensive approach². The sex-linked slow-feathering gene was widely using in layer sector for sex determination of chicks. A hen can transmit her sex-linked traits only to her son but a cockerel transmits its sex-linked traits to both sons and daughters³.

Mincheva *et al.*⁴ demonstrated that egg production traits of hybrids was not negatively affected by slow feathering (K) gene. One of the main prerequisites of using K locus genes was the lack of negative impact on production traits of hybrids and therefore, the knowledge on effects of K/k+ alleles on productivity was essential for breeding programmes. Khosravinia and Manafi⁵ stated that there were significant differences ($p < 0.01$) between fast-and slow-feathering chicks according to environmental stressors and slow-feathering chicks were more capable of withstanding holding periods. Male broilers, similarly as in guinea fowl, the fast feathering gene was associated with increased body weight⁶.

The purpose of the present study was to obtain parents of feather-sexable chicks and to investigate the effect of feathering rate on egg production and hatching traits. Parent males from fast feathering lines (kk) are crossed with parent females from slow feathering lines (K) for commercial hybrid chick production.

MATERIALS AND METHODS

This study was carried out between 2011 and 2014 years on 1 male (Rhode Island Red-1) and 2 female lines (Line-54 and Barred Rock-1) of Brown layers developed at Poultry Research Institute (PRI).

Animals and management: In the present study, fast-and slow-feathering genotypes were used from RIR, BAR and L54. The fast and slow feathering of sire and dam genotypes were

coded as RIR-F, RIR-S, BAR-F, BAR-S, L54-F and L54-S, respectively. In the present project, a total of 6638 hatching eggs were used and 5076 chicks were produced. The mean hen number per genotype used for hatching eggs was 85.5 ± 0.88 and hatching eggs and chick number per hen were 12.94 ± 0.10 and 9.89 ± 0.10 , respectively.

Artificial Insemination (AI) was used to maintain the parent stocks at the PRI; however, before using sperm for AI, the sperm characteristics of cocks were examined for abnormalities under a microscope. For the duration of the study, hens were housed in individual cages and 9 hens were artificially inseminated with semen diluted with physiological saline [Phosphate-Buffered Saline (PBS)] (1:1) twice a week. It should be noted that cocks and hens were mated from the same feathering rate genotypes at the grandparent level, whereas different feathering rate genotypes were used for mating cocks and hens at the parent level, for example, RIR-F \times RIR-F or RIR-S \times RIR-S at the grandparent level and RIR-F \times RIR-S at the parent level. The shells of the eggs obtained from the hens housed in individual cages were marked with the cock and hen number and eggs were collected as hatching eggs from day 2 post-insemination for 14 days. All substandard eggs were rejected and good quality oval-shaped eggs weighing 54-65 g were selected for hatching. On the 18th days of incubation, the eggs were candled to detect fertilized eggs and any dead embryos. At the end of the incubation period, un-hatched eggs were separated and cracked to determine infertility or early (<7 days), middle (8-17 days) or late (18-21 days) periods of embryonic mortality. In the present study, single stage setters (Pas Reform TM6 115000 capacity) and hatchers (Pas Reform TM4 19200 capacity) were used. The incubator temperature was maintained between 36.7 and 38.0°C during incubation and 36.7°C during the hatching time. Humidity was maintained at 52% during incubation and 60-65% at hatching time.

At the hatching time, the wing feathering rate was determined in one-day-old chicks and those showing desirable feathering rates with respect to the chosen genotypic characteristics were selected as breeders. The tail feathering rates of chicks were also examined at 10 days of age and those which did not show matching tail and wing feathering rates with the desired characteristics were discarded.

During the rearing period chicks were kept in four-tier, group-cage batteries up to the 16th week. The dimension of the chick cages were; length 120 cm, wideness 63 cm, height 40 cm; feeder length 120 cm. Chicks were reared with 60 birds per cage ($126 \text{ cm}^2 \text{ bird}^{-1}$) until the 5th week then

30 birds per cage from the 6th-15th week (252 cm² bird⁻¹). During the chick rearing period temperature was gradually reduced from 35-22°C in the 1st-15th week and relative humidity was between 60 and 70%. At 16th week of age, female pullets were transported to 3 tier, single-cage batteries (dimension; length 29 cm, wideness 54 cm, height 62 cm, feeder length 29 cm) with 1566 cm² floor space per hen.

During the present study, quality drinking water and standard poultry rations with balanced nutrition were maintained as according to NRC⁷ and were provided *ad-libitum*. The birds were fed crumbled diets with 2,980, 2,950, 2,930 and 2,850 kcal of Metabolisable Energy (ME) kg⁻¹ of diet; each contained 20, 18, 15 and 16.5% Crude Protein (CP), respectively, from 0-20 weeks of age. During the 1st and 2nd egg production cycle, birds were fed diets containing 2,750 and 2,650 kcal of ME kg⁻¹ of diet, respectively; each contained 17 and 16% CP, respectively. The birds were exposed to natural daylight and were provided with artificial light to increase the length of daylight to up to 16 h day⁻¹, which was then kept constant and maintained until the end of the study.

Data collection: Data for the characteristics detailed below were recorded. Age at First Egg (AFE) was measured as the age at which a hen laid her first egg. The Body Weight at First Egg (BWFE) was determined by weighing hens individually on the first laying day. Egg Weight (EW) was calculated as the mean of consecutive values (g) of 3 eggs weighed on the 28th, 32nd and 36th week. Egg Number (EN) was calculated as the total number of eggs produced by an individual hen over 43 weeks. Incubation traits of hens were investigated after 43 weeks of age by collection of the eggs for 14 days. The fertility (%), embryonic mortality (%) and hatchability (%) rates were calculated using the following formulae⁸:

$$\text{Fertility rate} = \frac{\text{Number of fertile eggs}}{\text{Number of eggs placed into the hatching machine}} \times 100$$

$$\text{Embryonic mortality rate} = \frac{\text{Number of dead embryos}}{\text{Number of eggs placed into hatching machine}} \times 100$$

$$\text{Hatchability rate} = \frac{\text{Number of alive hatched chicks}}{\text{Number of eggs placed into hatching machine}} \times 100$$

Statistical analysis: The data for early, middle and late mortality, fertility and hatchability rates were transformed into angles [angle = (arc sin)] prior to analysis⁹. The untransformed

data are also displayed in the results tables. Statistical evaluation of egg production and hatching traits were performed using one-way analysis of variance (ANOVA)⁹ in Minitab statistical software package¹⁰ (p<0.05). Genetic parameters were estimated on performance data from 2,162 BAR-F, 1,701 BAR-S, 2,127 L54-F, 1,458 L54-S, 1,789 RIR-F and 1,480 RIR-S (total 10,717 hens) from a three generation selection experiment for the traits: AFE, BWFE, EN and EW by the restricted maximum likelihood (REML) procedure¹¹. Differences between means were analysed at a significance level of 0.05 using the Tukey test.

RESULTS

The L54-F birds reached sexual maturity significantly (p<0.05) earlier than other genotypes and RIR-S birds reached sexual maturity later than other genotypes. Although the differences between BAR-S and RIR-F birds were not significant, differences between all the other genotypes were significant (p<0.01) with regards to AFE. The BWFE was found to be significantly influenced by feathering rate (p<0.01). Interestingly, L54-F birds had significantly (p<0.01) lighter BW than other genotypes, whereas the heaviest genotype was RIR-F. However, there was no significant difference between fast-and slow-feathering BAR with respect to BWFE. Mean egg production of L54-F hens was significantly (p<0.05) higher when compared to other genotypes during the 43-week study period. It was noted that hens from fast-feathering genotypes of BAR, L54 and RIR produced a greater number of eggs than those of slow-feathering genotypes (p<0.01).

The means of total EW over the 43 weeks study period were between 54.70-58.93 g and significant differences were found between genotypes (p<0.01). Results showed that the egg weights of RIR-F and RIR-S, which were similar, were significantly heavier than those of other genotypes, whereas the difference between the EW of BAR-F and BAR-S was also not significant. Interestingly, L54-S hens produced significantly (p<0.05) heavier eggs than L54-F hens (Table 1).

Significant differences (p<0.01) were observed in fertility, embryonic mortality and hatchability rates between the 6 genotypes. The BAR-F was different from L54-S in terms of fertility rate (p<0.05). Early period embryonic mortality rate of RIR-S was significantly higher than those of the L54-F, L54-S and RIR-F genotypes. In the BAR-F, the middle period embryonic mortality rate was significantly lower than that of the other genotypes (p<0.01). Late period embryonic mortality of RIR-S (6.53 %) was significantly higher than those of the RIR-F (4.24 %) and BAR-F (3.50 %) genotypes but there was no significant difference among BAR-S, L-54-F, L-54-S and

Table 1: Descriptive statistics values of egg production traits (X±SEM)

Genotypes	N (hen)	AFE (day)	BWFE (g)	EN (per hen)	EW (g)
BAR-F	2162	152.68±0.18 ^a	1674.00±2.99 ^{bc}	126.46±0.33 ^b	55.51±0.07 ^c
BAR-S	1701	155.24±0.20 ^b	1666.40±3.67 ^c	123.76±0.39 ^c	55.34±0.09 ^c
L54-F	2127	149.42±0.18 ^e	1562.90±3.10 ^e	129.36±0.36 ^a	54.70±0.08 ^d
L54-S	1458	153.90±0.20 ^c	1630.60±4.55 ^d	126.44±0.40 ^b	57.65±0.10 ^b
RIR-F	1789	156.22±0.43 ^b	1748.10±3.90 ^a	124.42±0.72 ^{bc}	58.93±0.09 ^a
RIR-S	1480	161.29±0.24 ^a	1694.90±8.78 ^b	117.54±0.46 ^d	58.51±0.15 ^a
P		0.001	0.001	0.001	0.001

N: No. of hen, AFE: Age at first egg, BWFE: Body weight at first egg, EN: Egg number, EW: Egg weight, BAR-F: Barred Rock-1 fast, BAR-S: Barred Rock-1 slow, L54-F: Line-54 fast, L54-S: Line-54 slow, RIR-F: Rhode Island Red-1 fast, RIR-S: Rhode Island Red-1 slow, a, b, c, d, e: Means within a column with different letters are significantly different (p<0.05)

Table 2: Effect of feathering rate on fertility, embryonic mortality and hatchability rates (X±SEM)

Genotypes	Fertility (%)	Early period	Middle period	Late period	Hatchability (%)
		embryonic mortality (%)	embryonic mortality (%)	embryonic mortality (%)	
BAR-F	95.72±0.43 ^a	7.48±0.62 ^{ab}	2.68±0.38 ^b	3.50±0.41 ^c	82.06±0.81 ^a
BAR-S	93.60±0.58 ^{ab}	9.68±0.77 ^{ab}	4.77±0.47 ^a	6.21±0.64 ^{ab}	72.93±1.21 ^{cd}
L54-F	93.14±0.61 ^{ab}	7.71±0.84 ^{ab}	5.18±0.54 ^a	4.30±0.67 ^{abc}	75.93±1.38 ^{bc}
L54-S	93.10±0.61 ^b	7.37±0.78 ^b	5.42±0.47 ^a	5.41±0.52 ^{abc}	74.89±1.15 ^{bc}
RIR-F	94.64±0.52 ^{ab}	7.38±0.74 ^b	4.63±0.43 ^a	4.24±0.43 ^{bc}	78.38±0.99 ^{ab}
RIR-S	92.92±0.73 ^{ab}	11.61±0.98 ^a	5.60±0.61 ^a	6.53±0.62 ^a	69.17±1.52 ^d
P	0.004	0.001	0.001	0.005	0.001

BAR-F: Barred Rock-1 fast, BAR-S: Barred Rock-1 slow, L54-F: Line-54 fast, L54-S: Line-54 slow, RIR-F: Rhode Island Red-1 fast, RIR-S: Rhode Island Red-1 slow, a, b, c, d, e: Means within a column with different letters are significantly different (p<0.05)

RIR-F genotypes (Table 2). The lowest hatchability was observed as 69.17% in the RIR-S genotype, whereas the highest rate was as 82.06% in the BAR-F genotype (p<0.01).

Heritability is fundamental to the present study, because designs for breeding programmes are based on reliable estimations of genetic variation for relevant traits as well as for genetic correlations between the traits. It is speculated that heritabilities of egg production traits were generally lower in the RIR-F genotype than other genotypes and that EN showed a low-moderate heritability, ranging from 0.13-0.59. Heritabilities for AFE varied from 0.21-0.46, whereas EW was a highly heritable trait with values ranging from 0.26-0.59 as shown in Table 3. In this study, positive genetic correlations were observed between AFE and BWFE in all genetic groups, except L54-F and L54-S. The estimated genetic correlations of AFE with EN in all genotypes and AFE with EW, except L54-S and RIR-F, were negative. In all genotypes, EN was negatively correlated with BWFE and EW. Although BWFE was positively correlated with EW in BAR-F, BAR-S, L54-F and L54-S, a negative correlation was observed in the RIR-F and RIR-S genotypes.

The PRI offers specific crosses suitable for most defined study purposes but is also required to maintain a limited number of different genotypes to optimise utilisation. To improve egg production, selection has to be based on the genetic variation within genotypes. In our study, genetic

correlations were estimated to construct a selection index and to predict genetic changes in traits, resulting from more or less emphasis on egg production traits.

DISCUSSION

In the present study, the effects of feathering rate on egg production and hatching traits were examined on the basis of fast-and slow-feathering genotypes derived from the same pure lines. From this point of view, fast-feathering pullets matured significantly (p<0.01) earlier than slow-feathering genotypes. BAR-F, L54-F and RIR-F genotypes reached sexual maturity an earlier age than BAR-S, L54-S and RIR-S genotypes, by 3, 4 and 5 days, respectively. Results of this study agree with those of Lowe and Garwood¹² and Harris *et al.*¹³ who reported that fast-feathering pullets matured earlier than slow-feathering pullets. The BWFE of fast-feathering genotypes was significantly (p<0.01) higher than that of their slow-feathering counterparts, except for the BAR strain. These results agree with Lowe and Garwood¹⁰ and Durmus *et al.*¹⁴. According to egg number, there was a significant (p<0.01) difference between fast-and slow-feathering genotypes and fast-feathering genotypes produced more eggs than slow-feathering genotypes. It has been reported that the slow-feathering trait has in fact reduced egg production¹³, a finding compatible with this study results. It was also

Table 3: Genetic correlation values among egg production traits and estimated heritabilities for the egg production traits (X±SEM)

Traits	r _G	p-value	Traits	h ²
BAR-F				
AFE-BWFE	0.63±0.071	0.103	AFE	0.29±0.050
AFE-EN	-0.01±0.134	0.001	BWFE	0.43±0.053
AFE-EW	-0.27±0.109	0.006	EN	0.40±0.059
BWFE-EN	-0.67±0.083	0.001	EW	0.56±0.054
BWFE-EW	0.20±0.102	0.004	-	-
EN-EW	-0.35±0.103	0.001	-	-
BAR-S				
AFE-BWFE	0.85±0.052	0.001	AFE	0.38±0.077
AFE-EN	-0.02±0.198	0.003	BWFE	0.35±0.075
AFE-EW	-0.08±0.147	0.012	EN	0.21±0.061
BWFE-EN	-0.54±0.144	0.001	EW	0.54±0.078
BWFE-EW	0.15±0.152	0.008	-	-
EN-EW	-0.43±0.157	0.001	-	-
L54-F				
AFE-BWFE	-0.39±0.076	0.755	AFE	0.45±0.046
AFE-EN	-0.16±0.087	0.902	BWFE	0.51±0.048
AFE-EW	-0.37±0.073	0.001	EN	0.59±0.048
BWFE-EN	-0.84±0.026	0.004	EW	0.57±0.047
BWFE-EW	0.90±0.015	0.001	-	-
EN-EW	-0.75±0.038	0.008	-	-
L54-S				
AFE-BWFE	-0.21±0.122	0.001	AFE	0.34±0.062
AFE-EN	-0.31±0.116	0.009	BWFE	0.51±0.061
AFE-EW	0.10±0.120	0.638	EN	0.46±0.058
BWFE-EN	-0.86±0.036	0.484	EW	0.59±0.057
BWFE-EW	0.79±0.043	0.001	-	-
EN-EW	-0.85±0.033	0.012	-	-
RIR-F				
AFE-BWFE	0.01±0.277	0.001	AFE	0.21±0.057
AFE-EN	-0.09±0.255	0.005	BWFE	0.11±0.004
AFE-EW	0.01±0.184	0.023	EN	0.13±0.034
BWFE-EN	-0.82±0.074	0.001	EW	0.26±0.052
BWFE-EW	-0.06±0.225	0.007	-	-
EN-EW	-0.28±0.179	0.001	-	-
RIR-S				
AFE-BWFE	0.40±0.286	0.571	AFE	0.46±0.128
AFE-EN	-0.71±0.146	0.001	BWFE	0.44±0.130
AFE-EW	-0.28±0.278	0.277	EN	0.58±0.133
BWFE-EN	-0.61±0.209	0.683	EW	0.45±0.159
BWFE-EW	-0.36±0.246	0.001	-	-
EN-EW	-0.24±0.241	0.531	-	-

r_G: Genetic correlation, h²: Heritability, AFE: Age at first egg, BWFE: Body weight at first egg, EN: Egg number, EW: Egg weight, BAR-F: Barred Rock-1 fast, BAR-S: Barred Rock-1 slow, L54-F: Line-54 fast, L54-S: Line-54 slow, RIR-F: Rhode Island Red-1 fast, RIR-S: Rhode Island Red-1 slow

determined that the L54-S genotype produced a higher EW than the L54-F, BAR-F and BAR-S genotypes. Although the highest EW was produced by L54-F and RIR-F and RIR-S hens, the lowest was produced by L54-F. Interestingly, there was no significant difference between the EWs of genotypes derived from the same lines, with the exception of the L54 hens. Similar results were reported by Lowe and Garwood¹² and Harris *et al.*¹³. Ledvinka *et al.*¹⁵ stated that the age of laying hens, genotype and the feather growth-rate gene incidence within the population significantly affected average egg weight, eggshell quality indicators and eggshell colour. Further study by Ledvinka *et al.*¹⁶ reported that the

feather growth-rate gene affected egg weight, the ratio of yolk, white and eggshell, Haugh unit, eggshell strength and colour. According to Durmus *et al.*¹⁷, fast-feathering hens had superior production traits compared to slow-feathering hens. In laying hens, the K gene has been shown to have adverse effects on egg production and mortality in chickens^{12,13}. Mincheva *et al.*⁴ investigated the effects of sex-linked feathering alleles on laying performance, hatchability and body measurements in hens from 2 White Plymouth Rock lines. Their results showed that the K locus alleles had no significant effect on egg production traits (p>0.05). In the present study, the sire genotypes and the mean EWs of

RIR-F and RIR-S were significantly heavier ($p < 0.01$) than those of other genotypes.

Although the mean EW of RIR-S was significantly heavier ($p < 0.01$) than those of other genotypes, with the exception of RIR-F, the lowest hatchability and EN were obtained in RIR-S. Flock *et al.*¹⁸ stated that high egg weight correlated with low egg production and poor hatchability, thereby agreeing with our findings. To obtain the desired EW using hybrid chickens, EW was maintained near the optimum for hatchability and chick quality in parent dam genotypes, whereas in parent sire genotypes, EW was higher. Gowe *et al.*¹⁹ reported that the important reproductive traits in poultry breeding were the proportions of fertile and viable eggs. In contrast, Nurgiartiningsih *et al.*²⁰ reported that genetic parameters, such as heritability and genetic correlation, were the most important considerations in determining appropriate animal evaluation methods. Pym²¹ described sex-limited traits with low heritability. Higher hatchability was observed in the fast-feathering genotypes than the slow-feathering genotypes, with the exception of Line-54. The high early embryonic mortality rate during egg incubation was significant ($p < 0.01$) and resulted from a decreased hatchability in RIR-S genotypes. The reason for the increased early embryonic mortality rate associated with the feathering rate was not ascertained and requires further study.

All egg production traits have moderate to high heritabilities and should be responsive to change through individual phenotypic selection. In the current study, positive genetic correlations were observed between AFE and BWFE in all genetic groups, except for L54-F and L54-S. All the estimated genetic correlations of AFE with EN and AFE with EW, except for L54-S and RIR-F, were negative. In all genotypes, EN was negatively correlated with BWFE and EW. Although, BWFE was positively correlated with EW in BAR-F, BAR-S, L54-F and L54-S, this relationship was negative in the RIR-F and RIR-S genotypes.

Xie *et al.*²² developed one fast-and one slow-feathering pure line of Anyi tile-like grey chickens and as a result of mating two pure lines, 94.96% auto-sexing accuracy rate of the progenies was obtained. Therefore, as a result of this study, sex identification of chicks was achieved with an average of 96% accuracy in hybrids from both fast- and slow-feathering genotypes. It can be concluded that this value can be further improved by further experimental studies on chickens.

CONCLUSION

Mean egg production and hatching traits in genetic groups of laying hens were significantly affected by the

slow-feathering gene (K). It was determined that the K-locus alleles adversely affected fertility, embryonic mortality, hatchability and egg production traits except for EW. Despite some adverse effects of sexing one-day-old chicks according to feather growing rate, it has been widely used in practise as in the Poultry Research Institute. If one of the traits accepted in selection studies for sexing one-day-old chicks is the feathering rate, the effect of the K gene on the studied traits should be taken into consideration.

SIGNIFICANCE STATEMENTS

Results of this study showed that the K-locus alleles of laying hens adversely affect fertility, embryonic mortality, hatchability and egg production traits, except for EW. Although the slow feathering gene (K) has some negative effect on egg production and hatching traits, it has a great commercial value in sexing day-old chicks. Findings of this study will guide the researchers to work on a poultry population carrying K-gene.

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