Feather colour affects the aggressive behaviour of chickens with the same genotype on the dominant white ($I$) locus

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Abstract

Aggression in chickens is a serious economic and animal welfare issue in poultry farming. Pigmentation traits have been documented to be associated with animal behaviour. Chicken pecking behaviour has been found to be related to feather colour, with premelanosomal protein 17 ($PMEL17$) being one of the candidate genes. In the present study, we performed a genotypic and phenotypic association analysis between chicken plumage colour (red and white) and aggressive behaviour in an F1 hybrid group generated by crossing the autosomal dominant white-feathered breed White Leghorn (WL) and the red-feathered breed Rhode Island Red (RIR). In genetic theory, all the progeny should have white feathers because WL is homozygous autosomal dominant for white feathers. However, we found a few red-feathered female chickens. We compared the aggressiveness between the red and white females to determine whether the feather colour alone affected the behaviour, given that the genetic background should be the same except for feather colour. The aggressiveness was recorded 5 days after sexual maturity at 26 weeks. Generally, white plumage hens showed significantly higher aggressiveness compared to the red ones in chasing, attacking, pecking, and threatening behaviour traits. We assessed three candidate feather colour genes—$PMEL17$, solute carrier family 45 member 2 ($SLC45A2$), and SRY-box 10 ($SOX10$)—to determine the genetic basis for the red and white feather colour in our hybrid population; there was no association between the three loci and feather colour. The distinct behavioural findings observed herein provide clues to the mechanisms underlying the association between phenotype and behaviour in chickens. We suggest that mixing red and white chickens together might reduce the occurrence of aggressive behaviours.
Introduction
Aggressive behaviour in chickens is a widespread economic and animal welfare issue in poultry farming, and many factors such as food, mates, and social rank affect its occurrence [1, 2]. Such behaviour has different forms (threats and intense agonistic behaviour) and is divided into numerous types such as still threats, chasing, aggressive pecking, and attacks [3]. As a complex trait in chicken, aggressiveness has been found to be present, albeit with low heritability ($h^2 = 0.04–0.17$) in various studies [4–8], and aggressive pecking has been reported to be affected by a variety of environmental factors, including light intensity [9], stocking density [10, 11], food [12], feeding methods [13], group size [14], and male presence [1]. Moreover, appearance factors such as comb type [15], plumage pattern [16], and plumage colour [17] can also influence the behaviour of chickens. Therefore, aggressive behaviour in chickens is a function of interaction among genes, phenotype, and environment.

In terms of the genetics of aggressiveness, a few candidate genes, including premelanosome protein 17 (PMEL17, I locus), solute carrier family 45 member 2 (SLC12A9), G protein subunit gamma 2 (GNG2), calyntenin-2 (CLSTN2), brain-derived neurotrophic factor (BDNF), neurtensin (NTS), G protein subunit alpha o1 (GNAO1), and sortilin-related VPS10 domain-containing receptor 2 (SORCS2) have been identified to be correlated to the trait [18–20]. As a domesticated trait, colour phenotypes may also be selected as side-effects linked to behaviour during domestication [17]. Previous studies on pigs [21], silver foxes [22], Norway rats [23], deer mice [24], mink [25], and chickens strongly supported this point. In chickens, feather-pecking damage had a highly significant QTL that coincided perfectly with the dominant white locus (I) [26], and the PMEL17 gene was found to be responsible for this phenotype [27]. Moreover, the PMEL17 locus could also affect explorative and social behaviours in chickens [17]. However, whether PMEL17 is directly associated with behaviour or indirectly influences feather colour remains unknown. If the PMEL17 locus were consistent (II), whether there are other factors that might affect chickens’ feather colour and behaviour and how long the impact can last also remain subject to investigation. In this study, we used individuals with two different feather colours (white and red) and controlled the effect of the I locus to discover the behavioural diversity in mature chickens.

Materials and methods
All procedures and protocols (DOI: dx.doi.org/10.17504/protocols.io.ysgfwbw) involving animals were conducted in accordance with the Guidelines for the Care and Use of Experimental Animals established by the Ministry of Agriculture of China (Beijing, China). All the animal protocols were approved by the Animal Welfare Committee of China Agricultural University (Beijing, China, Permit Number: XK622).

Animals
The chickens used were the offspring of a cross between RIR (male) and WL (female). WL’s dominant white plumage and RIR’s sex-linked recessive red plumage were genetically determined by PMEL17 on an autosome (denoted as I) and SLC45A2 on a Z chromosome (denoted as Z$^s$), respectively. The genotypes of the two loci in WL are IIZ$^s$/IIZ$^w$, and for RIR, they are iiZ$^s$/is for male/female. Since white feathers in WL are autosomal dominant homozygous (II), the daughters or sons of WL are expected to have white plumage (I-). However, when we crossed female WL (II) and male RIR (Z$^s$Z$^w$), some exceptions were observed, with a few daughters (IiZ$^w$W) presenting with red feathers or red-coloured heads, although most of the daughters (IIZ$^w$W) were white. The genetic basis underlying these exceptions remains to be identified.
The WL and RIR chickens used in this study were maintained by a commercial company (Hehei Dawu Co., Ltd.) and had undergone selection for production traits for more than 30 generations. We believed that the I and Z′ locus of the F1 females ought to be the same; we separated the birds into those with white feathers, red feathers, and red-coloured heads. Therefore, these groups of birds formed an ideal population for investigating the association, without PMEL17’s influence, between pigmentation and the behaviour of chickens.

We crossed male RIR and female WL to obtain F1 hybrids (commercial line) and 150 red-feathered daughters, and 150 white-feathered daughters (Fig 1) were randomly selected for our experiments. All the hens were examined in further behavioural tests.

**Housing conditions of animals**

The eggs from the cross between RIR males and WL females were hatched at the same time. We collected red- and white-feathered female chickens (because all the males were white). We reared the selected females in the same chicken house, where the lighting, temperature, and ventilation were completely artificially controlled. The temperature was maintained at 23°C with ventilation fans and cooling pads. The chickens were housed in six pens with a plastic net ground (4.30 m long × 4.00 m width × 2.00 m high, 1.00 m above the floor). The pens were equipped with automatic drinkers (Fig 1), and clean water (fresh) was available ad libitum to the birds. The chickens were fed manually with standard commercial chicken feed. No adverse effects such as severe exhaustion, illness, or severe injury were observed to have resulted from the experiment. In addition, the birds were exposed to a 13.5L:10.5D photoperiod (from 0700 to 2030 hours) with a light power of 5 W (26 weeks).

**Behavioural tests**

Red and white hens were separately reared in chick cages (~30 chickens/cage) from 1–115 days and transferred to adult chicken cages (3 chickens /cage) after 115 days. At 20 weeks old, the chickens were transferred to the pens and were divided into six groups of 50 birds each. Groups 1 and 2 were replicates for the red-feathered hens, while groups 3 and 4 were the same, each containing 25 red and 25 white hens. Groups 5 and 6 were replicates for the white hens (Fig 1). All the birds were allowed to adapt to the new conditions for 5 weeks. After the adaptation period, aggressive behaviours were recorded using a monitor from day 177 to 181 (26 weeks), since the females were sexually mature and tended to be more aggressive at this time than before attaining sexual maturity. Moreover, body weight was recorded at the beginning (20 weeks) and end (25 weeks) of the adaptation period.

Behaviour was recorded between 1200 and 1230 hours by one person using the same standard (30 min × 5 days × 6 pens; S1 Table). Two main behavioural features, B1 and B2, were recorded in this study. B1 consisted of the chase and attack behaviour, while B2 comprised the peck and threat behaviour (Table 1). These behaviours were counted separately (attacker or victim) for the red- and white-feathered hens in the blended group (groups 3 and 4). The behavioural definitions were based on the ethogram of Väisänen [3]. Each distinct behaviour pattern occurring with the two same hens (attacker or victim) was counted as one event independent of its duration unless suspended by any other behaviour for more than 3 s. Due to the inability to identify individuals within a pen, we used the entire pen’s behavioural occurrence time as the standard unit.

**Genotyping candidate genes for feather colour**

To delineate the genotypes underlying the red and white feather colour and determine if they directly or indirectly influence behaviour, we selected locus 3 as the gene for genotyping and
association analysis. Since we already knew the genotype of locus 2, including dominant white feathers (I) and sex-linked recessive red feathers (Zs), we picked the SRY-box 10 (SOX10) gene for genotyping. SOX10 is responsible for the dark brown (DB) phenotype, which is caused by an 8.3 kb deletion upstream of the transcription start site [28].

A total of 60 females were selected, consisting of 29 and 31 with white- and red-feathered birds, respectively, from the F1 population. Wing vein blood was obtained from red- and white-feathered hens (without anaesthetic). Genomic DNA was extracted using standard phenol/chloroform protocols. The DNA concentration was determined using a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific Inc.).

Polymerase chain reactions (PCRs) were performed using a Veriti 96 Well thermal cycler (Applied Biosystems) according to the manufacturer’s protocol. The polymorph of the 8.3 kb deletion upstream of the SOX10 gene was amplified using PCR with the 1 forward (1F, 5'-CCTTTGTCTTAAGGCTCCTCTTT-3'), 1 reverse (1R, 5'-CCTTGTGGAGACCAGGTGTT-3'), and 6R (5'-TGCTGAGACATTTGCTGACA-3') primers from Gunnarsson et al. [28].

| Table 1. Two aggressive behaviour phenotypes in female chickens demonstrated in this study. |
|-----------------------------------------------|
| **Abbreviation** | **Categories** | **Behaviour description [3]** |
| B1 | Chase | Bird follows another; both birds run, jump, or fly. |
|     | Attack | Bird jumps, flies, runs, or takes fast steps when approaching another bird to give it an aggressive peck; birds stand or walk >1 m away from each other. |
| B2 | Peck | Bird rapidly pecks the anterior part of another bird. |
|     | Threat | Stiff body posture towards another bird; the birds stand <0.25 m from each other. The head is positioned above or below the receiver’s head. Feathers may be lifted. |

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Fragments of 611 (1F/1R) and 1257 bp (1F/6R) associated with the wild-type (db/db) and dark brown (DB/DB) alleles, respectively, were displayed by agarose gel electrophoresis.

Statistical methods
The descriptive statistics were analysed using the Statistical Package for the Social Sciences (SPSS) software using all available records [29]. The general linear model was used for analysing the main and interactive effects of feather colour and position (south or north; Fig 1) on the occurrence of behaviours. In addition, Tukey’s honestly significant difference test was conducted for pairwise comparisons (behaviours and body weight) between different groups.

Results
Body weight
In the present study, hens with two feather colours obtained from the cross were raised in the same facility under the same environmental conditions. To minimize body size differences, all chickens were randomly separated to six pans (20 weeks). Although body weight differed (G1 and G2 were significantly heavier than G3 and G5) in six groups at the age of 20 weeks (Table 2), no significant difference in body weight was found in the repeat groups (G1 vs. G2, G3 vs. G4, and G5 vs. G6). After 5 weeks of adaptation, there was no significant difference among the feeding pans of the three types (red, mingled, and white groups) (Table 2).

However, the mingled group showed an inconsistent result. There was no difference between the body weight of red- and white-feathered birds (group 3), but it differed after adaptation (Table 3). Thus, most individuals had a relatively uniform size in the behaviour tests.

Aggressive behaviours
All birds were healthy throughout the observation period, and we easily recorded their aggressive behaviours. Feather colour had a significant effect on the counts of both B1 and B2 aggressive behavioural traits measured (Table 4). The white hen group (G5 and G6) exhibited significantly (p < 0.001) higher aggressive levels of chase, attack, peck, and threat behaviours.

Table 2. Descriptive statistics of body weight (20 and 25 weeks) in six groups.

| Feather colour | Red | Mingled | White |
|----------------|-----|---------|-------|
| Group          | G1  | G2      | G3    |
| (n = 50)       | (n = 50) | (n = 50) | (n = 50) |
| Body weight (20 wks) | Mean ± SD (g) | 1434.10 ± 110.80<sup>a</sup> | 1426.92 ± 105.37<sup>a</sup> | 1354.22 ± 112.96<sup>b</sup> |
| CV (%)         | 7.38 | 7.38    | 8.34  |
| Body weight (25 wks) | Mean ± SD (g) | 1566.22 ± 132.75 | 1570.62 ± 101.37 | 1531.64 ± 129.63 |
| CV (%)         | 8.53 | 6.45    | 8.46  |

Table 3. Descriptive statistics of body weight (20 and 25 weeks) in mingled groups (group 3 and group 4).

| Mingled group | Group 3 | Group 4 |
|---------------|---------|---------|
| Feather colour | White (n = 25) | Red (n = 25) | White (n = 25) | Red (n = 25) |
| Body weight (20 wks) | Mean ± SD (g) | 1348.52 ± 115.02 | 1359.92 ± 112.94 | 1387.00 ± 130.32 | 1347.12 ± 120.16 |
| CV (%)         | 8.53 | 8.31    | 9.40  | 8.92  |
| Body weight (25 wks) | Mean ± SD (g) | 1586.40 ± 103.81<sup>a</sup> | 1476.88 ± 131.46<sup>b</sup> | 1536.56 ± 97.72<sup>ab</sup> | 1524.32 ± 106.02<sup>ab</sup> |
| CV (%)         | 6.54 | 8.90    | 6.36  | 6.96  |

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than the red hen group (G1 and G2) did (Fig 2). Moreover, the mean number of aggressive behaviours exhibited was lower among the mingled groups than the white groups (29.50 vs. 50.90 in B1, 116.10 vs. 239.60 in B2). Pairwise comparison between the red and mingled group did not reveal significant differences in counts of B1 and B2.

The results of each group are shown in Tables 5 and 6. Peck and threat behaviours (109.40 to 242.60) occurred more frequently than chase and fight (24.80 to 61.20) did in the three different types of hen groups. Behavioural traits were highly variable, and descriptive statistics of behaviours in each group showed very high coefficient of variation (14.16% to 59.09%) (Table 5).

Furthermore, we counted the number of attacks and victim experiences between the red and white hens in the mingled groups, respectively (Table 6). However, there was no

### Table 4. P- and F-values for the main effects of feather colour (red, mingled, and white), position (north and south), as well as the interactive effect of feather colour × position on the behavioural variables obtained by GLM.

| Variable | Feather colour | Position | Feather colour × position |
|----------|----------------|----------|----------------------------|
|          | P   | F      | P    | F   | P     | F      |
| Behaviour 1 | <0.001 | 10.825 | 0.093 | 3.055 | 0.205 | 1.696 |
| Behaviour 2 | <0.001 | 23.668 | 0.882 | 0.023 | 0.748 | 0.294 |

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### Fig 2. Aggressive behaviours count in different feather colours hens. (a) Red and white feather coloured groups (mean ± SE). (b) Mingled group (mean ± SE).

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significant difference for B1-attacker, B2-attacker, B1-victim, or B2-victim between the red and white hens (Fig 2).

Genotyping
The genotype of the SOX10 deletion for both red and white plumage hens is shown in Table 7. The homozygous DB deletion did not appear in any of the tested hens. Two types of SOX10 genotypes were recognised—heterozygous DB deletion and non-deleted wild-type—in both the red and white plumage hens, indicating that this locus was not linked to feather colour phenotypes.

Discussion
The domestication of animals is associated with feather or coat colour changes in various animal species [22, 30]. In mammals, these observations suggest that domestication has a co-evolutionary effect of inducing pigment loss and tame behaviour, which has also been proven in birds.

In the present study, the birds used were the offspring of the first generation of a cross between RIR (iiZsZs) and WL (IIZsW). However, a few daughters (IiZsW) presented with red feathers or red-coloured heads, although most were white (IiZsW). The emergence of red-feathered hens shows that the epistatic effect of PMEL17 on SLC45A2 was reduced. Moreover, behavioural observations of the red (groups 1 and 2) and white (groups 5 and 6) groups indicated that white plumage hens were more aggressive than the coloured hens were, although all the birds had the same I locus (I/i).

Keeling (2009) showed that feather damage due to feather-pecking by other birds was 3.34 times higher in pigmented birds (i/i) than that in white (I/I) birds. However, the tendency for red (pigment type, I/i) birds to experience more attacks compared to white birds (I/i) was not significant in this study (Fig 2), which might be explained in three possible ways. First, the

*Table 5. Descriptive statistics of two aggressive behaviours in six groups (5 days).*

| Feather colour | Red (n = 5) | M Mingled (n = 5) | White (n = 5) |
|----------------|------------|-----------------|--------------|
| Group          | G1         | G2              | G3 (n = 5)   | G4 (n = 5) | G5 (n = 5) | G6 (n = 5) |
| Behaviour 1 (chase, attack) | Mean ± SD  | 24.80 ± 12.89   | 25.40 ± 15.01| 32.20 ± 9.63| 26.80 ± 7.98| 61.20 ± 21.49| 40.60 ± 6.80 |
| CV (%)         | 51.98      | 59.09           | 29.9        | 29.78      | 35.11      | 16.76      |
| Behaviour 2 (peck, threat) | Mean ± SD  | 158.40 ± 45.57  | 144.20 ± 41.38| 109.40 ± 33.35| 122.80 ± 22.83| 242.60 ± 60.97| 236.60 ± 33.49 |
| CV (%)         | 28.77      | 28.7            | 30.49       | 18.59      | 25.13      | 14.16      |

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*Table 6. Descriptive statistics of two aggressive behaviours in mingled groups (5 days).*

| Group         | Group 3 | Group 4 |
|---------------|---------|---------|
| Feather colour| White (n = 5) | Red (n = 5) | White (n = 5) | Red (n = 5) |
| B1_attacker   | Mean ± SD  | 12.00 ± 3.74 | 20.20 ± 6.50 | 17.40 ± 7.27 | 9.40 ± 2.79 |
| CV (%)        | 31.18    | 32.16    | 41.76      | 29.71      |
| B2_attacker   | Mean ± SD  | 39.60 ± 11.15| 69.80 ± 24.10| 69.60 ± 17.46| 53.20 ± 12.28|
| CV (%)        | 38.15    | 34.52    | 25.08      | 23.08      |
| B1_victim     | Mean ± SD  | 16.40 ± 6.66 | 15.80 ± 5.81| 17.40 ± 4.22| 9.40 ± 4.72 |
| CV (%)        | 40.58    | 36.74    | 24.25      | 50.24      |
| B2_victim     | Mean ± SD  | 54.00 ± 18.34| 55.40 ± 15.87| 56.20 ± 14.79| 66.60 ± 11.17|
| CV (%)        | 33.97    | 28.64    | 26.31      | 16.77      |

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same genotype \((I/i)\) might lead to uniform behavioural effects. Second, a huge coefficient of variation might reduce the difference between red and white hens in the mingled groups (group 3 and group 4). Third, the small sample size \((n = 25)\) of the groups might have affected the results.

From the perspective of genetics, one or more genes might be controlling the different plumage colour formation in the present population. Unfortunately, consistent with the \(SOX10\) genotyping results, neither \(SOX10\) nor \(PMEL17\) caused the red/white phenotype or produced dramatically different behavioural changes in the present population [17, 26]. Meanwhile, it is still unclear how feather colours and pigments affect (directly or indirectly) each other.

Normally, two main pigments in birds, eumelanin and pheomelanin, are generated from \(\text{L-tyrosine}\) after a series of reactions [31], and dopa and dopa analogues are crucial mid products [32]. In addition, the serotonergic and dopaminergic systems also have possible roles in the aetiology of feather pecking [33–36]. \(\text{DRD4}\), encoding the dopamine receptor D4, has been determined to be associated with the behaviour of birds [37]. \(\text{GNG2}\), involved in monoamine signalling, particularly in postsynaptic signalling at serotonergic and dopaminergic synapses, was identified as a positional candidate gene in chicken pecking in a genome-wide analysis study [18]. These clues suggest that bird behaviour and pigment traits might be influenced by dopa analogues or other mid-products. However, further well-designed experiments are required to prove this hypothesis.

Meanwhile, interesting phenomena appeared in the comparisons among the mingled group and two pure colour groups (red or white groups). Compared with the white group hens (higher aggression level), hens in the mingled groups showed a significantly lower occurrence of aggressive behaviours. This might be explained by the existence of a “mixed-phenomenon” effect, which occurs in certain vertebrate species, e.g. fish schools, ungulate herds, and bird flocks. Mixed-species associations of birds are roving groups of individuals comprising at least two species searching for food together [38].

Considering these findings, we constructed a cross between WL and RIR, and all the progenies exhibited the same genotype on the \(PMEL17\) locus \((Ii)\). We observed behavioural differences between the red- and white-feathered female progenies after they attained sexual maturity. Our results showed that feather colours could affect the behaviour independent of the \(PMEL17\) locus and remained latent till sexual maturity at 26 weeks. However, it is not clear which gene controlled the pigmentation or behaviour in this population. To investigate the genetic bases for the pigmented offspring of WL and the relationship between the above behaviour-related genes and feather colour, further research is warranted.

### Table 7. Plumage colour and related genotype distribution in the F1 generation of the White Leghorn/Rhode Island Red cross.

| Phenotype | \(SOX10\) |  |  |
|-----------|-----------|---|---|
|           | \(DB/DB^1\) | \(DB/db^2\) | \(db/db^3\) |
| Red       | 0         | 15 | 14 |
| White     | 0         | 23 | 8  |

\(^1\text{DB/DB: dark brown allele}\)

\(^2\text{DB/db: heterozygote allele}\)

\(^3\text{db/db: wild-type allele}\).

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Conclusions

In the present F1 cross population, white-feathered hens were more aggressive than the red ones were. Compared with raising white chickens together, mixing red and white chickens together can reduce the occurrence of aggressive behaviours. Moreover, these differences in behaviour and phenotype were not caused by \textit{PMEL17}, \textit{SOX10}, or \textit{SLC45A2}, which provide a good model for behavioural research of individual birds with different feather colours. Taken together, the distinct behavioural findings observed in this study provide novel insights into the mechanisms underlying the association between phenotype and behaviour in the present population and other chicken breeds.

Supporting information

S1 Table. Counts of two aggressive behaviours in six groups (5 days).

(DOCX)

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References

1. Craig JV. Aggressive behavior of chickens: Some effects of social and physical environments. 1978.

2. Pagel M, Dawkins MS. Peck orders and group size in laying hens: ‘Futures contracts’ for non-aggression. Behavioural Processes. 1997; 40(1):13–25. https://doi.org/10.1016/s0376-6357(96)00761-9 PMID: 24897609

3. Väisänen J, Håkansson J, Jensen P. Social interactions in Red Junglefowl (Gallus gallus) and White Leghorn layers in stable groups and after re-grouping. British Poultry Science. 2005; 46(2):156–68. https://doi.org/10.1080/000716660500062638 PMID: 15957435
4. Grams V, Wellmann R, Preuß S, Grashorn MA, Kjaer JB, Bessei W, et al. Genetic parameters and signatures of selection in two divergent laying hen lines selected for feather pecking behaviour. Genetics Selection Evolution. 2015; 47(1):77.

5. Bennewitz J, Böglein S, Stratz P, Rodehutscord M, Piepho HP, Kjaer JB, et al. Genetic parameters for feather pecking and aggressive behavior in a large F2-cross of laying hens using generalized linear mixed models. Poultry science. 2014; 93(4):810–7. https://doi.org/10.3382/ps.2013-03638 PMID: 24706957

6. Rodenburg TB, Buitenhuis AJ, Ask B, Uitdehaag KA, Koene P, Jj VDP, et al. Heritability of feather pecking and open-field response of laying hens at two different ages. Poultry science. 2003; 82(6):861. https://doi.org/10.1093/ps/82.6.861 PMID: 12817438

7. Kjaer JB, Sorensen P, Su G. Divergent selection on feather pecking behaviour in laying hens (Gallus gallus domesticus). Appl Anim Behav Sci. 2001; 71(3):229–39. Epub 2001/03/07. PMID: 11230903.

8. Su G, Kjaer JB, Sorensen P. Variance components and selection response for feather-pecking behavior in laying hens. Poultry science. 2005; 84(1):14–21. https://doi.org/10.1093/ps/84.1.14 PMID: 15685937

9. Mohammed HH, Grashorn MA, Bessei W. The effects of lighting conditions on the behaviour of laying hens. Archiv Fur Geflugelkunde. 2010; 74(3):197–202.

10. Pettit-Riley R, Estevez I, Russek-Cohen E. Effects of crowding and access to perches on aggressive behaviour in broilers. Applied Animal Behaviour Science. 2002; 79(1):11–25.

11. Estevez I, Newberry RC, Keeling LJ. Dynamics of aggression in the domestic fowl. Applied Animal Behaviour Science. 2002; 76(4):307–25.

12. Anderson KE, Adams AW. Effects of type of cage front and feed trough partitions on productivility and ingestive, agonistic, and fearful behaviors of egg-type hens. Poultry science. 1991; 70(4):770–5. https://doi.org/10.3382/ps.0700770 PMID: 1876556

13. Ventura BA, Frank S, Imma E. Access to Barrier Perches Improves Behavior Repertoire in Broilers. PloS one. 2012; 7(1):e29826. https://doi.org/10.1371/journal.pone.0029826 PMID: 2229026

14. D’Eath RB, Keeling LJ. Social discrimination and aggression by laying hens in large groups: from peck orders to social tolerance. Applied Animal Behaviour Science. 2003; 84(3):197–212. https://doi.org/10.1016/j.applanim.2003.08.010

15. Siegel PB, Dudley DS. Comb Type, Behavior and Body Weight in Chickens. Poultry science. 1963; 42(2):516–22.

16. Crawford DR, Smyth RJ. Social and Sexual Behavior as Related to Plumage Pattern in the Fayoumi Fowl. Poultry science. 1964; 43(5):1193.

17. Karlsson AC, Kerje S, Andersson L, Jensen P. Genotype at the PMEL17 locus affects social and explorative behaviour in chickens. British Poultry Science. 2010; 51(2):170–7. https://doi.org/10.1080/00071661003745802 PMID: 20461577

18. Lutz V, Stratz P, Preuss S, Tetens J, Grashorn MA, Bessei W, et al. A genome-wide association study in a large F2-cross of laying hens reveals novel genomic regions associated with feather pecking and aggressive pecking behavior. Genetics, selection, evolution: GSE. 2017; 49(1):18. Epub 2017/02/06. https://doi.org/10.1186/s12711-017-0287-4 PMID: 28158968.

19. Li Z, Zheng M, Abdalla BA, Zhang Z, Xu Z, Ye Q, et al. Genome-wide association study of aggressive behaviour in chicken. Scientific reports. 2016; 6:30981. Epub 2016/08/04. https://doi.org/10.1038/srep30981 PMID: 27485826.

20. Guo X, Fang Q, Ma C, Zhou B, Wan Y, Jiang R. Whole-genome resequencing of Xishuangbanna fighting chicken to identify signatures of selection. Genetics, selection, evolution: GSE. 2016; 48(1):82. Epub 2016/08/28. https://doi.org/10.1186/s12711-016-0239-4 PMID: 27565441.

21. Fang M, Larson G, Ribeiro HS, Li N, Andersson L. Contrasting Mode of Evolution at a Coat Color Locus in Wild and Domestic Pigs. PLoS genetics. 2009; 5(1):e1000341. https://doi.org/10.1371/journal.pgen.1000341 PMID: 19148282

22. Trut LN. Early Canid Domestication: The Farm-Fox Experiment. American Scientist. 1999; 87(2):160–9.

23. Cottle CA, Price EO. Effects of the nonagouti pelage-color allele on the behavior of captive wild Norway rats (Rattus norvegicus). Journal of Comparative Psychology. 1987; 101(4):390. PMID: 3691061

24. Haysen V. Effects of the nonagouti coat-color allele on behavior of deer mice (Peromyscus maniculatus): a comparison with Norway rats (Rattus norvegicus). Journal of Comparative Psychology. 1997; 111(4):419–23. PMID: 9419866

25. Keeler C, Moore T. Psychosomatic synthesis of behavior trends in the taming of mink. 1961.

26. Keeling L, Andersson L, Schutz KE, Kerje S, Fredriksson R, Carlborg O, et al. Feather-pecking and victim pigmentation. Nature. 2009; 431:645–6.
27. Kerje S, Sharma P, Gunnarsson U, Kim H, Bagchi S, Fredriksson R, et al. The Dominant white, Dun and Smoky color variants in chicken are associated with insertion/deletion polymorphisms in the PMEL17 gene. Genetics. 2004; 168(3):1507–18. https://doi.org/10.1534/genetics.104.027995 PMID: 15579702.

28. Gunnarsson U, Kerje S, Bed'hom B, Sahlqvist AS, Ekwall O, Tixier-Boichard M, et al. The Dark brown plumage color in chickens is caused by an 8.3-kb deletion upstream of SOX10. Pigment cell & melanoma research. 2011; 24(2):268–74. Epub 2011/01/08. https://doi.org/10.1111/j.1755-148X.2011.00825.x PMID: 21210960.

29. Inc S. IBM SPSS Statistics for Windows, Version 20.0. 2011.

30. Klungland H, Vage DI. Pigmentary switches in domestic animal species. Annals of the New York Academy of Sciences. 2003; 994(1):331–8.

31. Sharma S, Wagh S, Govindarajan R. Melanosomal proteins—role in melanin polymerization. Pigment Cell Research. 2002; 15(2):127–33. PMID: 11936270

32. Cieslak M, Reissmann M, Hofreiter M, Ludwig A. Colours of domestication. Biological reviews of the Cambridge Philosophical Society. 2011; 86(4):885–9. Epub 2011/03/30. https://doi.org/10.1111/j.1469-185X.2011.00177.x PMID: 21443614.

van Hierden YM, Koolhaas JM, Kostal L, Vyboh P, Sedlackova M, Rajman M, et al. Chicks from a high and low feather pecking line of laying hens differ in apomorphine sensitivity. Physiology & Behavior. 2005; 84(3):471–7. https://doi.org/10.1016/j.physbeh.2005.01.015 PMID: 15763586

34. van Hierden YM, Korte SM, Ruesink EW, van Reenen CG, Engel B, Korte-Bouws GAH, et al. Adrenocortical reactivity and central serotonin and dopamine turnover in young chicks from a high and low feather-pecking line of laying hens. Physiology & Behavior. 2002; 75(5):653–9. https://doi.org/10.1016/s0031-9384(02)00667-4

35. Dennis RL, Chen ZQ, Cheng HW. Serotonergic mediation of aggression in high and low aggressive chicken strains. Poultry science. 2008; 87(4):612–20. https://doi.org/10.3382/ps.2007-00389 PMID: 18339980

36. Kjaer JB, Hjarvard BM. Effects of haloperidol, a dopamine D2 receptor antagonist, on feather pecking behaviour in laying hens. Applied Animal Behaviour Science. 2004; 86(1):77–91.

37. Flisikowski K, Schwarzenbacher H, Wysocki M, Weigend S, Preisinger R, Kjaer JB, et al. Variation in neighbouring genes of the dopaminergic and serotonergic systems affects feather pecking behaviour of laying hens. Animal Genetics. 2009; 40(2):192–9. https://doi.org/10.1111/j.1365-2052.2008.01821.x PMID: 19120086

38. Colorado Zuluaga GJ. WHY ANIMALS COME TOGETHER, WITH THE SPECIAL CASE OF MIXED-SPECIES BIRD FLOCKS. Revista EIA. 2013:49–66.