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

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ABSTRACT

Aggression in chickens is a serious economic and welfare issue. Pigmentation traits have been well documented to be associated with animal behaviour. Previous studies have shown that chicken pecking behaviour is related to the feather colour and premelanosome protein 17 (PMEL17) is 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 an autosome dominant white feather breed, White Leghorn (WL), and a red feathered breed, Rhode Island Red (RIR). In genetic theory, the progeny should all have white feathers since the WL are homozygous autosome dominant white. However, we found a few red-feathered female chickens. We compared aggressiveness between the red and white females to determine whether the feather colour alone affected the behaviour since the genetic background should be the same except for the feather colour. The aggressiveness was recorded 5 days after sex maturity at 26 weeks. Generally, white plumage hens showed significantly higher aggressiveness than the red ones did in chase, attack, peck, and threatening behaviour. We also detected three feather colour candidate genes, PMEL17, solute carrier family 45 member 2 (SLC45A2), and SRY-box 10 (SOX10) in determining the genetic foundation for the red and white feather colour in our hybrids population and there was no association between the three loci and the feather colour. Additional studies should be conducted to elucidate further the genetic or phenotypic mechanisms underlying the feather colours and the behaviour.

EXTERNAL LINK

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Animals and Behavioural tests

1 The chickens used were the offspring of a cross between RIR (Rhode Island Red /male) and WL (White Leghorn /female).

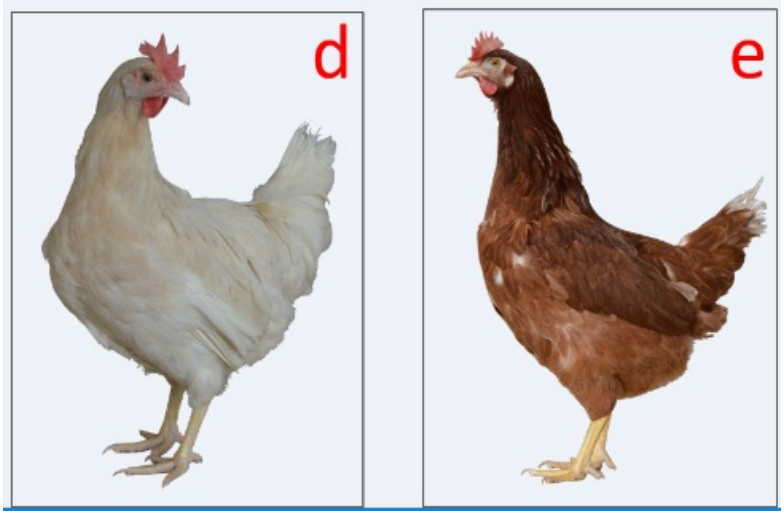
2 WL's dominant white plumage and RIR's sex-linked recessive red feather were genetically determined by PMEL17 on an autosome (denoted as I) and SLC45A2 on a Z chromosome (denoted as Zs), respectively. It is well knownthat the genotypes of the WL of the two loci areIIZSZS/IIZSWand for the RIR are iiZsZs/is for male/female.

Since white feathers inWL are autosomal dominant homozygous (II) and the daughters or sons of WLgenerally have white plumage (I-

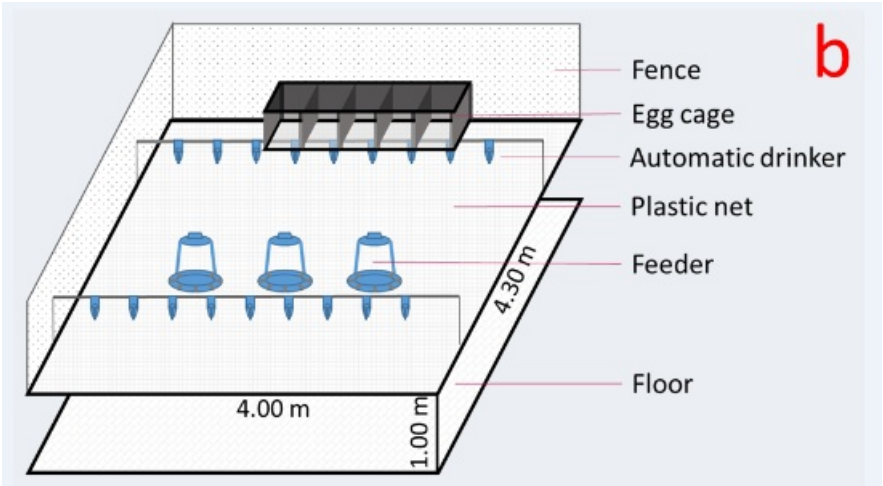
3).However, when we crossed the female WL (II)and male RIR (ZsZs),some exceptions were observed with a few daughters (IiZsW) that presented with red feathers or red-coloured heads, although most of thedaughters (IiZsW) were white. The genetic basement underlying these exceptions remains to be identified.

4 The eggs from the cross between WL males and RIR females were hatched at the same time, and we collected red- and white-featheredfemale chickens since all the males were white.

5 we crossed male RIR and female WL to obtain theF1 hybrids and 150 red-feathered daughters, and 150 white-featthereddaughters were randomly selectedfor our experiments. All the hens were examinedin further behavioural tests.

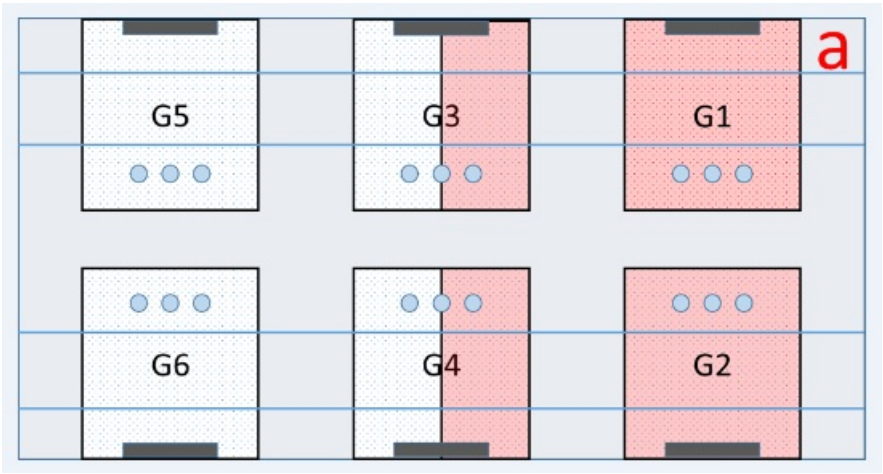


6 We reared the selected females in the same chicken house where the lighting,temperature, and ventilation were completely artificially controlled. Thetemperature was maintained at 23°C with ventilationfans and cooling pads. The chickens were housed in six pens with a plastic net ground (4.30 m long × 4.00 mwidth × 2.00 m high, 1.00 m above the floor). The pens were equippedwith automatic drinkers, and cleanwater (fresh) was available ad libitumto the birds. The chickens were fed manually with standardcommercial chicken feed. In addition, the birds wereexposed to a 13.5L: 10.5D photoperiod (from 7:00 to 20:30h) with a light powerof 5W (26 week).



7 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 dividedinto six groups of 50 birds each. Moreover, we have been weighted all birds at 20 and 25 weeks age.

8 Groups 1 and 2 were replicates for the red feather hens, while groups 3 and 4 were the same and each contained 25 red and 25 white hens. Group 5 and 6 were replicates for the white hens.



9 All the birds were allowed to adapt to the new conditions for 5weeks. After the adaptation period, aggressive behaviours were recorded using a monitor from day 177 to 181 (26 week), since the females were sexually matured and tended to be more aggressive at this time than before they were sexually matured.

10 Behaviours werecounted between 12:00 and 12:30 by one person using the same standard (30mins* 5 days* 6pens, Figure 1a). Two main behaviouralfeatures, B1 and B2, were recorded in this study. B1 consisted of the chase andattack, while B2 was the peck and threat behaviours (Table 1).

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 stands < 0.25 m from each other. Head positioned above or below the receiver's head. Feathers may be lifted.

11 These behaviours were counted separately (attacker or victim) for the red and white featherStiff body posture towards another bird, the birds stands < 0.25 m from each other. Head positioned above or below the receiver's head. Feathers may be lifted. hens in the blended group (groups 3 and 4).

Genotyping candidate genes for feather colour

12 we selected loci 3 as the gene for genotyping and association analysis. Since we already knew the genotype of loci 2 including dominant white (I) and sex-linked recessivered feather (Zs), we picked the SRY-box 10 (SOX10) gene for genotyping. The SOX10 is responsible for the dark brown(DB) phenotype and was causal by an 3 kb deletion upstream of the transcription start site.

13 A total of 60 females were selected, consisting of 29 and 31 with white and red feathers, respectively from the F1 population. Wing vein blood was obtained from red and white feathered hens (without anesthetic). Genomic DNA was extracted using standard phenol/chloroform protocols.

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