

Full Length Research Paper

Preliminary identification of genetic mechanism of shank color in game chicken

G. B. Chang, H. Chang, R. Chen, T. Ma, A. Q. Dai and G. H. Chen*

College of Animal Science and Technology, Yangzhou University, Yangzhou 225009, China.

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This study aimed to explore a simple way to identify the genetic mechanism of shank color in chicken and provide an on-the-spot evaluation method for the trait without genealogy records. The shank color of 202 parental mating combinations and 489 offsprings were observed in Zhongyuan Game chicken and Snyder genetic ratios were calculated. The results showed that the cyan shank was dominant compared to the livid shank in Zhongyuan Game chicken, which was coincident with the characteristic of Mendelian trait in autosome. We concluded that Snyder genetic ratio was an effective way to identify the genetic mechanism of complex genetic trait.

Key words: Chicken, shank color, Snyder, genetic mechanism.

INTRODUCTION

Shank color is a complex genetic trait in Game chicken and controlled by multiple gene loci and other factors (such as species, nutrition, gender, age and so on) (Crawford, 1990). On one hand, because the genetic and physiological mechanisms of pigment formation remain unclear, we know a little about the chicken pigment-related traits (feather, skin, shank color, etc.) (Crawford, 1990; Yang et al., 2004; Chang et al., 2006). On the other hand, genealogy records of generations are needed to identify the genetic mechanism of pigment-related traits, which is difficult to achieve in practice especially for the rare traits. Nevertheless, as a part of the species genetic characteristics, the pigment-related traits are required to assess, protect and utilize the genetic resources. It is urgent to provide an on-the-spot evaluation method to identify the genetic mechanism of the trait without genealogy records. In chicken, the shank color is mainly affected by epidermal melanin, dermal melanin and carotenoids, including yellow, flesh-colored, red, black, blue, cyan and so on. Moore et al. (1972) have shown

that there were eight major genotypes of the shank color in chicken involved in four loci (E, I, W, and Id) and also negatively affected by the common reed flower gene. Yin et al. (2001) thought that the light shank was completely dominant to the dark shank with the effect of gender and age, which was slightly divergent from different mating combinations. Kang et al. (2002) observed the shank and plumage color and the feather growth in a slow-feathering pure line of Gushi chicken. They found that the shank color gradually changed from light to dark with the increasing of week-age while the plumage color did from dark to light. While both positive and negative cross-mating were carried out to produce F2 population using White Leghorn, Shouguang and Tibetan chicken, Wang et al. (2006) found that Id / id gene showed a sex-linked inheritance in the shank color and the id gene had expressed step by step in the recessive homozygous. And then they confirmed that the genotype of shank color was dominant homozygous in the white leghorn rooster. Recent studies have shown that there were multiple alleles in Id locus, five alleles were found up to now but their genetic mechanisms remained unclear. This study aimed to explore a simple method to identify the genetic mechanism of shank color and provide a new way to reveal the genetic mechanisms of shank color and other

*Corresponding author. E-mail: passioncgb@163.com. Tel: 86-514-87979034. Fax: 86-514-87350440.

Table 1. Primary observation on shank color of Zhongyuan Game chicken.

Parental combination	Number	Offspring		
		Livid shank	Cyan shank	Total
LividxLivid	28	48	0	48
LividxCyan	79	81	109	190
CyanxCyan	95	35	216	251
Total	202	164	325	489

Mendelian traits in the Game chicken as well as the wild animal population. It is true that the method of trait separation and dominant inheritance initially revealed by Snyder and Fisher is still one of the fundamental laws widely used in the population genetics theory. Herein, it was an exploration and attempts to discuss the genetic mechanism of shank color in the balance population with Snyder genetic ratios, which would provide bases to protect the genetic diversities effectively.

MATERIALS AND METHODS

Animals

In this study, 202 pairs of Zhongyuan Game chicken parents were selected and divided into three types of mating combination to produce the offsprings (Table 1).

Snyder genetic ratio

Definition: In the Hardy-Weinberg equilibrium population, the proportion of recessive offspring is the ratio of the frequency of recessive gene to the value plus 1 in the mating system of the dominant type with the recessive type (DxR), while it is the square of the former in the mating system of the dominant type with the dominant type (DxD).

$$\begin{aligned} \text{DxR: } S_1 &= q/(1+q) \\ \text{DxD: } S_2 &= q^2/(1+q)^2 \end{aligned}$$

Where

q = frequency of recessive gene;
D = the individual of dominant type;
R = the individual of recessive type.

Omitting the certification, the concrete steps are available in Wright (1953).

Application: When the genetic approach of the relative trait is unknown, an assumption is proposed with the relationship of dominant and recessive. Under this assumption, the frequency of recessive gene and Snyder genetic ration are calculated with the actual observation. According to this ratio, the number of the dominant and recessive individuals is deduced from two kinds of mating system respectively. Finally, the number is compared with the actual number and a suitability test is made to accept the assumption or not.

Requirement: The phenotypes are known for parent-child generations.

RESULTS

In our experiment, we obtained 489 progeny chickens in all and then observed the phenotypes (livid or cyan shank) and numbers for two generations respectively (Table 1). We found that there was no individual with cyan shank in the mating system of LividxLivid.

According to the above phenotypic data, we proposed the following two assumptions to calculate the Snyder genetic ratios respectively.

Assumption 1: Livid shank is dominant; cyan shank is recessive.

$$q = \sqrt{R} = \sqrt{[(95 \times 2) + 79 + 325] \div [(2 \times 202) + 489]} = 0.816 \quad (1)$$

$$E(S_1) = q / 1 + q = \frac{0.816}{1.816} = 0.449 \quad (2)$$

$$E(S_2) = q^2 / (1 + q)^2 = 0.449^2 = 0.202$$

(3) Among the offsprings in the mating system of DxR (that is LividxCyan):

- i) The livid shank number is $(1-0.449) \times 190 = 104.69$;
- ii) The cyan shank number is $0.449 \times 190 = 85.31$.

Among the offsprings in the mating system of DxD (that is LividxLivid):

- i) The livid shank number is $(1-0.202) \times 48 = 38.30$;
- ii) The cyan shank number is $0.202 \times 48 = 9.70$.

(4) Making a suitability test between the corresponding numbers for dominant and recessive type from two kinds of mating system and the actual numbers.

$$\chi^2 = \frac{(81-104.69)^2}{104.69} + \frac{(109-85.31)^2}{85.31} + \frac{(48-38.30)^2}{38.30} + \frac{(0-9.70)^2}{9.70} = 24.10$$

(df = 3, P < 0.005).

(5) In practice, $S_1 = 109/190 = 0.573$; $S_2 = 0/48 = 0$.

Assumption 2: Cyan shank is dominant; livid shank is recessive.

Table 2. Comparison on the results between two supposed analyses.

Supposed	q	E(S ₁)	S ₁	E(S ₂)	S ₂	χ ²
Livid shank>Cyan shank	0.816	0.449	0.573	0.202	0	24.10 (P<0.005)
Cyan shank>livid shank	0.579	0.367	0.426	0.135	0.139	2.92 (P>0.30)

$$q = \sqrt{R} = \sqrt{(28 \times 2 + 79 + 164) \div (202 \times 2 + 489)} = 0.579 \quad (1)$$

$$E(S_1) = q / 1 + q = \frac{0.579}{1.579} = 0.367 \quad (2)$$

$$E(S_2) = q^2 / (1 + q)^2 = 0.135$$

(3) Among the offsprings in the mating system of D×R (that is Livid×Cyan):

- i) The livid shank number is $(1 - 0.367) \times 190 = 120.27$;
- ii) The cyan shank number is $0.367 \times 190 = 69.73$.

Among the offsprings in the mating system of D×D (that is Cyan×Cyan):

- i) The livid shank number is $(1 - 0.135) \times 251 = 217.12$;
- ii) The cyan shank number is $0.135 \times 251 = 33.88$.

(4) Making a suitability test between the corresponding numbers for dominant and recessive type from two kinds of mating system and the actual numbers.

$$\chi^2 = \frac{(81 - 69.73)^2}{69.73} + \frac{(109 - 120.27)^2}{120.27} + \frac{(35 - 33.88)^2}{33.88} + \frac{(216 - 217.12)^2}{217.12} = 2.92$$

(df = 3, P > 0.30).

(5) In practice, $S_1 = 81/190 = 0.426$; $S_2 = 35/251 = 0.139$.

Based on the result of a suitability test (Table 2), we accepted the assumption 2. It was indicated that the cyan shank was dominant to the livid shank and the livid shank was a recessive character.

Cyan shank, also known as bluish-green shank, was generally derived from the interaction between carotenoids and dermal melanin, related to W, Id, and E loci. In this study, the estimated genotype of the cyan shank was $W^+ W^+ id^+ id^+ e^+ e^+$ while that of the livid shank was $w^+ w^+ id^+ id^+ e^+ e^+$. The genetic mechanism of the shank color was similar to that of the Mendelian trait controlled by a pair of autosomal alleles. Of course, it was possible that there was a kind of allosomal inheritance, but it was normally inapplicable for a lack of reciprocal cross data.

DISCUSSION

Snyder genetic ratio is used to identify briefly the genetic

mechanisms of Mendelian traits in mammals without the genealogical records. It is applied to (1) semi-wild animals and wild animals; (2) rare traits; and (3) traits without systemic observation. As the Snyder genetic ratio is relatively simple and can be directly used on the farm only with the phenotypic data of two parent-child generations, the geneticists pay renewed attention to the Snyder genetic ratio. Jia et al. (1989) have proved that the inheritance of tongue-tied and non-tongue-tied was determined by a pair of autosomal alleles in human, and the tongue-tied was completely dominant compared to the non-tongue-tied. Otto et al. (1994) found the genetic mechanisms of the tongue-tied trait in Brazilians as well as other Mendelian traits with the different penetrance using Snyder genetic ratio. Snyder genetic ratio is the expanded application based on the Hardy-Weinberg equilibrium and also suitable for the sex-linked loci (sex-linked inheritance). For the sex-linked trait in the balanced population, the frequency of homozygote is the square of the corresponding gene frequency in female livestock and male birds while the frequency of heterozygote is twice the corresponding gene frequency product. The absolute difference value of gene frequency between male and female birds falls down by a half with the generation in random mating system, but the gene frequency in the whole population is constant, which is also the reason that our study left the gender out of consideration. In addition to identify the Mendelian traits in animal populations, Snyder genetic ratio can also provide a reference to remodel the inherent superior characteristics of endangered animal species and protect the species resources in our country.

Barrows (1914) had pointed out that the cyan was caused by the appearance of dermal melanin in the cutex. The phenotype of shank and foot depends on the accumulation and interaction effects among key genes, as well as the enhancement or inhibition of dermal melanin caused by the uncertain modifier genes. The role of Id and id^+ alleles is limited to the derma, while E is related to the cutex and also affects the amount of dermal pigment. Dominant white (I) has an epistatic effect on epidermal melanin and dilutes dermal melanin. So the recessive white does. The major inhibitors of dermal melanin are common reed flower gene and dot gene which are both sex-linkage. In addition, the absence of W and w^+ alleles or the interaction between yellow carotenoid and dermal melanin lead to the blue or cyan shank.

Zhongyuan Game chicken includes Henan Game chicken, Wanbei Game chicken and Luxi Game chicken

and is distributed in the Yellow River alluvial plain of the Zhongyuan area. It has been one of the characteristics of local culture. Zhongyuan Game chicken shows the half-prism body, small head, fine feather, nodular comb, vague wattle as well as thin and tight shin. The hair color is miscellaneous and there is no feather on the shank with a wide distance among four toes. The shank is mostly flesh-colored and there are dark shanks, especially the cyan shank is more prevalent. Because the producing areas are closer and different populations mixes with each other, the phenotype of chicken shank become more and more complex and the relevant research results differ from each other (Liu and Niu, 1994). In this study, we analyzed the genetic mechanism of shank color by calculating Snyder genetic ratios of the phenotypic data of two generations in Zhongyuan Game chicken. Preliminary results provided a reference to carry out the more rigorous analysis after the population entering into the protecting track and having access to genealogy records. Our study was only one case.

Conclusion

Snyder genetic ratio is used to identify the genetic mechanisms of Mendelian traits in mammals without the genealogical records. In this study, we indicated that the cyan shank was dominant to the livid shank and the livid shank was a recessive character using Snyder genetic ratios in Zhongyuan Game chicken, which would provide bases to the further research. This case suggested that Snyder genetic ratio was an effective way to identify the genetic mechanism of complex genetic trait.

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REFERENCES

- Barrows HR (1914). The histological basis of the different shank colors in the domestic fowl. *Maine Agric. Exp. Sta. Bull.*, 232: 237-252.
- Chang H, Yu YM, Qian GS, Ji DJ, Wu YQ (2006). Studies on way of purifying Mendelian characters in *Tupaia belangeri*. *Sci. Agric. Sin.*, 39: 2582-2589.
- Crawford RD (1990). *Poultry Breeding and Genetics*. Elsevier Press, New York, pp. 109-169.
- Jia SB, Zhu HB, Li SY, Liang FX, He DP, Sun XJ, Li YZ, Xu QF, Wang SJ (1989). An Analysis of Human Tongue-rolling Heredity Population. *J. Sci. Teach. Coll. Univ.*, 4: 52-56.
- Kang XT, Wang YB, Huang YQ, Xiong YH, Zhu XJ, Yang YL (2002). Study on shank color, plumage color and feather growth of slow-feathering pure line of Gushi fowl. *J. Northwest Sci-tech Univ. Agric. For. Nat. Sci. Edit.*, 30: 57-59.
- Liu FZ, Niu ZY (1994). Preliminary study on the genetics of feather color and skin color in Lueyang chicken. *J. Yellow Cattle. Sci.*, 20(sup.): 29-30.
- Moore JW, Jeffrey FP, Smyth JR (1972). Sexually dichromatic shank melanization associated with the brichen (ER) allele. *Poult. Sci.*, 51: 332-334.
- Otto PA, Frota-Pessoa O, Polcan SP (1994). Snyder's ratios with incomplete penetrance. *J. Hered.*, 85: 331-335.
- Wang CF, Li N, Wu CX (2006). Genetic Analysis of Feather Color and Shank Color Traits Based on F-2 Resource Population in Tibetan Chicken. *Hereditas*, 28: 810-814.
- Wright S (1953). The genetic structure of populations. *Ann. Eugen.*, 15: 323-354.
- Yang YS, Deng XM, Li N, Fu Y, Zhu MY, Wu CX (2004). MC1R is the candidate gene regulating Melanin synthesis in chicken. *Prog. Biochem. Biophys.*, 31: 500-505.
- Yin HG, Zeng ZJ, Pan YM, Zhou KY (2001). The genetic expression of dark shank in chicken. *Sichuan Anim. Vet. Sci.*, 28: 17-18.