Comparison of the W Chromosome-linked Female-specific Fragment (EE-0.6) in the Genus *Gallus*

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Abstract A female-specific DNA fragment (EE-0.6) exists in the chicken W chromosome and primers have been designed to amplify this area (USP-1, USP-3). Using these primers, we determined sequences of PCR products from the DNA of 4 species of jungle fowl and 2 varieties of domestic fowl as the template and carried out comparisons. We decoded 374 bp of the fragment (EE-0.6) and, comparing the decoded sequences, we found that *Gallus gallus*, domestic fowl (Gifu native fowl, Minorca) and *G. sonnerati* had the same sequence, while *G. lafayettei* and *G. varius* differed respectively at the 280th (C→G) and 281st (G→A) positions.

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Results and Discussion

Tone *et al.*²⁾ reported that the band pattern by means of *Msp* I of the W chromosome female-specific DNA fragment in three *Gallus* species (*G. gallus*, *G. sonnerati* and *G. varius*) is different among *Gallus gallus* • *G. sonnerati* and *G. varius*. It is thought that variations such as this control the potential for cross-breeding and we attempt to compare the sequences of the W chromosome-linked female-specific fragment (EE-0.6) in 4 species of jungle fowl and 2 varieties of domestic fowl classified in the genus *Gallus*.

Materials and Methods

We used total DNAs from blood samples of each one bird of the species *G. gallus*, *G. sonnerati*, *G. lafayettei*, *G. varius* and 2 varieties of domestic fowls (Minorca, Gifu native fowl). The primers used were USP-1 primer [5'-CTATGCCT ACCA (A/C) TTCCTATTTGC-3'] and USP-3 primer [5'-AGCTGGA (T/C)TTCAG (A/T) (C/G) CATCTTCT-3'] and, before initiating the PCR cycle, as preheating, we maintained the temperature at 95°C for 3 min. Conditions of the reaction were: 80 s at 95°C, 90 s at 60°C, and 60 s at 72°C per cycle, and 35 cycles were applied, which was then followed by single cycle reactions at 72°C for 9 min and at 4°C for 5 min¹⁾. The sequences were determined with an auto-sequencer (ABI) using the refined PCR products. We used DINASIS-Mac 3.6 to analyze the data that we obtained.

We determined the 374 bp sequence of the W chromosome female-specific DNA fragment (EE-0.6) in 4 species of jungle fowl and 2 varieties of domestic fowl (Fig.1). *G. gallus*, *G. sonnerati* and the 2 domestic fowl varieties (Minorca, Gifu native fowl) all had the same sequence, while *G. lafayettei* and *G. varius* were different at the 280th and 281st positions with C→G and G→A, respectively. The genus *Gallus* was therefore divided into three groups, *G. gallus*, *G. sonnerati* and two varieties of domestic fowl in one group, *G. lafayettei* in the second and *G. varius* in the third. In addition, the inclusion of *G. sonnerati* in the *G. gallus* group suggests that *G. sonnerati* may also have contributed, along with *G. gallus*, to the development of the domestic fowl.

References

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Comparison of the Female-specific Fragment

1	70
CTATGCCTACCACATTCCCTATTTGCTAGATGTCTTTGATGACTATAATCAAAACAACATGAAATACTAAG	
71	140
CATAAGCAATCATGTTAACATTAGGGTCACTGAATTTTACTTAAAAGTTTCAGTGCATTTATTTTACTGT	
141	210
GTATTTCCCTGTTTATCCACCCTAGATTGGTTAACCTATTTTCATTGACAATTTATCTATCTCCAGGGGAAA	
211	280
GCTGTATACAAGCAAGGAACTAAATCAGTGCCAACAACAACGATAAATGTTTTAGAATCACCTAATGTGC	
	<u>G</u>
281	350
GGAATGTCAATTTTAACTGAAATCCACTTCAGGTCAGATTATCTCTCAGACTCAACCTGAACCCATTACT	
<u>A</u>	
351	374
TAGAAGATGCNCTGAAATCCAGCT	<i>Gallus gallus</i>
	<i>Gifu native fowl</i>
	<i>Minorca breed</i>
	<i>Gallus sonnerati</i>
	<i>Gallus lafayettei</i>
	<i>Gallus varius</i>

Fig.1. Comparison of nucleotide sequences of genus *Gallus* EE-0.6.