

# The Genetic Diversity between Original Population and Breeding Population of Saba Pig Revealed by Microsatellite Variation

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## Introduction

Saba pig is mainly distributed in the mountain areas in western Yunnan, China. Saba pig is an excellent female parent for commercial hybrids because of its strong adaptability to crude feed, good meat quality and high reproductive performance (Lian et al., 1997). Saba pig breeding population has been constantly selected for 13 years complying with the breeding goal of specialized female parent. Saba pig original population is still grazed and distributed in geographically isolated villages. The intra-breed diversity is very important for selection and conservation. Mitochondrial DNA polymorphism was successfully used to determine the intra-breed diversity (Gou et al., 2006), and Microsatellite markers was broadly used as an effective intra-breed diversity index (Wilke et al., 1994; Martinez et al., 2000). In this work, 27 microsatellite markers were for the first used to investigate genetic variability of breeding population and original population of Saba pig.

## Material and methods

**The sources of animals.** A total of 101 individuals of Saba pig original population were randomly collected from 20 villages located in remote mountain areas in Yunnan province, China, to assure that they had been never crossbred with commercial breeds. A total of 74 individuals of Saba pig breeding group were collected from Yunnan Saba pig breeding and conservation farm.

**PCR amplification and genotyping.** 27 pairs of microsatellite primers recommended by FAO were chosen to maximize the genome coverage. When multiple markers were located in the same chromosome, there was a minimum distance of at least 30 CM to avoid linkage

disequilibrium. The markers were the following: CGA, S0155, SW240, SW72, S0227, S0005, IGF1, SW122, SW632, S0178, SW911, SW951, S0386, S0090, S0068, SW857, SW936, S0026, SW24, S0218, S0101, S0355, S0225, S0226, S0002, S0215, S0228. PCR-amplified products were resolved on 8% urea-PAGE denaturing sequencing gel. Allele visualization was achieved by silver staining according to manufacturer's standard protocol (Promega Corporation). The amplification product sizes were estimated using a 10-bp molecular weight ladder (Invitrogen, Life Technologies, CA, USA). Allele types and sizes at each locus were determined accurately by Bandscan5.0 (Glyko, USA). Genotypes of individual animal of two groups at 27 different loci were recorded by direct counting.

**Data analysis.** POPGENE32 (<http://www.ualberta.ca/~fyeh>) was used to test Hardy-Weinberg disequilibrium and to calculate allele frequencies, observed number of alleles ( $N_o$ ), effective number of alleles ( $N_e$ ), and heterozygosity ( $H$ ) at each locus in breeding population and original population. Polymorphism information content (PIC) value for each locus was calculated by using the method described by Botstein et al. (1980).

## **Results and discussion**

Relative genetic variability between the breeding population and original population were estimated using genotype data of 27 microsatellite markers. The observed and effective number of alleles, heterozygosity, and PIC values for each microsatellite locus in the two populations presented in Table 1. Allele frequency data (not presented) for each of the microsatellite marker is available with the authors. 208 and 127 alleles were identified across the 27 loci in original population and breeding population, respectively. Average number of observed alleles per locus was estimated to be 7.7, and 4.7 and 5.9 in original population and breeding population, respectively. Effective numbers of alleles in two populations were distinctly less than the observed values across all loci. The value for mean effective number of alleles was higher in original population (4.7) than in breeding population (3.0). In addition, the values for average heterozygosity and PIC among breeds were higher in original population (0.77 and 0.74, respectively) than in breeding population (0.64 and 0.60, respectively). All microsatellite loci in original population were deviated from Hardy-Weinberg equilibrium, while in breeding population, 9 loci were fitted to Hardy-Weinberg equilibrium, and 18 loci were deviated from Hardy-Weinberg equilibrium.

Table 1. Microsatellite alleles (No, observed; Ne, effective), heterozygosity (H) and polymorphism information content (PIC) at each locus in breeding and original population

Locus	Original population				Breeding population			
	No	Ne	H	PIC	No	Ne	H	PIC
CGA	9	4.5	0.78	0.76	5	2.7	0.64	0.57
S0155	6	3.8	0.74	0.70	5	3.5	0.72	0.68
SW240	7	4.2	0.76	0.74	4	2.5	0.60	0.55
SW72	5	3.4	0.73	0.70	4	2.7	0.72	0.68
S0227	6	4.6	0.78	0.76	3	1.4	0.28	0.26
S0005	7	5.5	0.81	0.81	4	2.8	0.65	0.64
IGF1	8	4.6	0.78	0.76	3	2.0	0.49	0.43
SW122	6	2.7	0.63	0.56	4	2.4	0.59	0.51
SW632	9	4.7	0.79	0.77	3	2.0	0.49	0.43
S0178	6	2.4	0.58	0.50	4	2.4	0.59	0.51
SW911	7	6.0	0.83	0.83	6	3.1	0.68	0.63
SW951	5	3.9	0.74	0.71	6	3.2	0.75	0.72
S0386	10	7.0	0.85	0.85	5	3.0	0.66	0.66
S0090	9	4.3	0.76	0.74	6	3.3	0.70	0.68
S0068	8	4.2	0.76	0.74	5	4.0	0.75	0.73
SW857	7	4.7	0.79	0.78	4	3.1	0.68	0.65
SW936	9	6.4	0.84	0.83	5	3.4	0.70	0.66
S0026	5	3.2	0.69	0.65	4	2.7	0.63	0.57
SW24	10	5.8	0.82	0.82	8	6.1	0.84	0.83
S0218	9	5.2	0.81	0.80	6	2.3	0.57	0.57
S0101	9	5.5	0.82	0.81	4	3.0	0.67	0.63
S0355	10	5.9	0.83	0.82	4	2.4	0.58	0.55
S0225	8	4.8	0.79	0.78	3	1.6	0.37	0.32
S0226	6	3.0	0.67	0.65	6	3.1	0.92	0.92
S0002	13	7.7	0.87	0.86	10	6.6	0.85	0.85
S0215	9	6.1	0.84	0.83	3	2.1	0.52	0.51
S0228	5	2.7	0.63	0.56	3	2.2	0.54	0.43

In present work, the observed number of alleles, effective number of alleles ( $N_e$ ), Polymorphism information content (PIC) and heterozygosity (H) at each locus of 27 Microsatellite Markers in breeding population were much lower than in original population. The result showed that Saba pig breeding population experienced serious loss of genetic diversity through constantly closed selection (Lian et al.2006). It is therefore necessary to open this population and introduce new individuals from the original population to maintain sufficient selection potentiality. All microsatellite loci were deviated from Hardy-Weinberg equilibrium in original population. The plausible reason is that only a little genetic exchanges occurred between geographically isolated subgroups in remote mountain areas, and it is suggested that Saba pig original population still retains sufficient subgroup diversity. Likely, 18 microsatellite loci were deviated from Hardy-Weinberg equilibrium in breeding population, but the reason possibly is that these loci were linked with breeding traits and were under selective pressure.

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