



2010 Annual Research Report

13. Subtropical Livestock Research

(1) Detection method of heterozygote with black-spots in white pig breed

This study was carried out to elucidate a diagnostic molecular marker for heterozygote with black-spots in pure white pig breed (i.e. Landrace and Large White). Using pedigree data previously analyzed, we selected heterozygote (I/IRN) in KIT gene. After mating between heterozygotes of KIT gene, we examined the phenotype of offspring. The genotypes of offspring were determined using RFLP, pyro-sequencing and conventional sequencing methods. In case of colored pig in white pig breed, they have no CNV (copy number variation) and splice mutation in intron 17 in KIT gene. Also, colored-pigs (IRN/IRN) were perfectly identified when compared with SNP site of KIT gene known in white pig breed. In conclusion, we could discern accurate genotype using CNV and splice mutation site and also develop diagnostic genetic marker for heterozygote with I/IRN genotype of KIT gene.

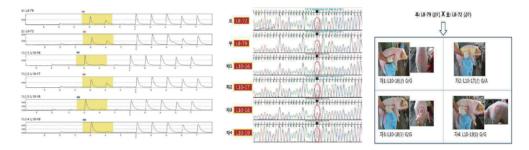


Fig. 13-1. Causative mutation of white pig breed with black-spots

(2) Identify genetic variation of paternal lineage in Jeju native black pigs(KNP)

To identify genetic variation of paternal lineage in Jeju native black pigs (KNP), six genes assigned to the Y-chromosome were selected. Also, eight pig breeds (Landrace, Yorkshire, Duroc, Berkshire, Korean wild boar (KWB) and three breeds of Chinese native pig were used for comparison with KNP. In this study, we constructed a linkage map and quantitative trait loci (QTL) analysis of genes affecting growth, carcass and blood traits. First, to confirm the correctness of the reference population crossed between KNP and Landrace, all pigs were genotyped using 13 microsatellite (MS) and 2 sexing markers. A linkage map was constructed by genotyping with multiplex-PCR using 168 MS markers



National Institute of Animal Science

and by using CRI-MAP version 2.4 (Green et al., 1990) with the build option. As a result, the total length of the 18 autosomal chromosomes was estimated at 2244.7 cM, and the average length between two MS markers was shown to be 13.4 cM. QTL analysis for growth, carcass and blood traits was performed by using QTL Express program. For blood traits, QTLs for mean corpuscular volume (MCV) were located in pig chromosomes 8 (70~111 cM) and 13 (70~99.5 cM). For carcass traits, four QTLs for backfat thickness were found in chromosomes 1, 4, 5 and 8. QTLs for 70-days and 140-days body weights were located in chromosomes 8 and 1, respectively. In addition, for reproduction traits, a QTL for teat numbers was estimated to be located in chromosome 7.

In conclusion, these results should be important for preservation and application of KNP. Especially, QTLs for economic traits may be used for identification of related genes and genetic markers, and moreover, for application of marker-assisted selection programs.

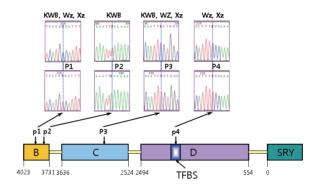


Fig 13-2. Conservation of SRY gene 5' area and SNP position jeju native black pig

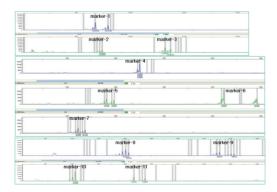


Fig. 13-3. Multiplex-PCR results using MS maker in jeju native black pig





2010 Annual Research Report

(3) Relationship between MC1R and ASIP Genotypes and Basic Coat Colors in Jeju Horses

This study was undertaken to reveal the relationship between genetic variations and the basic coat color classification system in Jeju horses. Genetic variations of the melanocortinreceptor 1(MC1R) and agouti signaling protein(ASIP) genes were investigated using pyrosequencing technique, A nucleotide substitution mutation for MC1R g,901C)T and an ASIP 11-bp deletion mutation were screened. Black horses had MC1R E-/- (E+/ E+ or E+/Ee) and ASIP Aa/Aa genotypes. In contrast, chestnut horse genotypes were MC1R Ee/Ee and ASIP -/-. Therefore, black and bay horses have at least one dominant MC1R allele, E+, whereas chestnut horses have homozygous recessive alleles Ee/Ee. This suggests that the MC1R genotypes determine chestnut or black/bay coat color, regardless of the genotype distribution of ASIP. In addition, the horses with MC1R E+/- and a dominant ASIP AA/- allele showed bay coat color, but not black, suggesting that the ASIP AA allele represses black coat color development in the hairs of the body, but not in the mane and all four legs. Pedigree analysis showed a consistent relationship between the genotype distribution of the MC1R and ASIP genes and basic coat color patterns, even in the F1 progeny. The results of this study revealed the relationship between the coat color phenotype and genetic background and suggested that useful information may be provided for molecular breeding of Jeju horses.

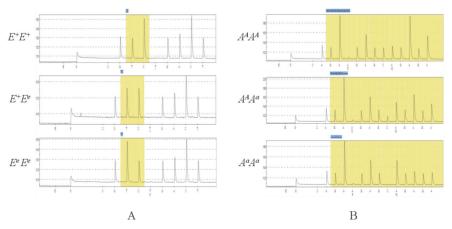


Fig. 13-4. Pyrograms generated with the PyroMark Q96 ID showing the pyrosequencing pattern and genotypes for MC1R (A) and ASIP (B).



National Institute of Animal Science

Table 13-1. Distribution of genotypes and frequencies of MC1R and ASIP among different Jeju horse coat colors.

Genetic variation	Genotype	No. of horse (Frequency)	Black	Bay	Chestnut
MC1R g.901C⟩T	E+E+	10 (0.093)	5 (0.208)	5 (0.131)	-
	E+Ee	52 (0,481)	19 (0.791)	33 (0.868)	-
	EeEe	46 (0.426)	-	-	46 (1.000)
ASIP exon 2 11-bp deletion	A^AA^A	23 (0,213)	-	13 (0.342)	10 (0.217)
	AAa	51 (0.472)	-	25 (0.657)	26 (0.565)
	A ^a A ^a	34 (0,315)	24 (1.000)	-	10 (0.217)

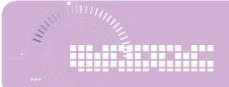
Table 13-2. Relationship between the basic coat color patterns and the combined genotypes of MC1R and ASIP in Jeju horse population tested in this study

Basic coat color	Genotype of MC1R/ASIP	of MC1R/ASIP No. of horse	
Black	E+E+/A ^a A ^a	5	0.046
	E+E ^e /A ^a A ^a	19	0.176
Bay	E+E+/A ^A A ^A	1	0.009
	E+E+/A ^A A ^a	4	0.037
	E+E ^e /A ^A A ^A	12	0.111
	E+E ^e /A ^A A ^a	21	0.194
Chestnut	E ^e E ^e /A ^A A ^A	10	0.093
	E ^e E ^e /A ^A A ^a	26	0.241
	E ^e E ^e /A ^a A ^a	10	0.093

(4) The study on cattle manure compost applications for organic grazing pasture in southern region of Korea

The dry matter yield of orchard grass pasture depending on the application of cattle manurecompost were found that cattle manure compost 100% and 200% were similar tendencies of dry matter yield to chemical fertilizer group (on the basis of nitrogen 200 kg/ha), but cattle manure compost 50% was $(p \langle 0.05)$ somewhat low dry matter yield.





2010 Annual Research Report

Even in bermuda grass pasture, moreover, it was found that cattle manure compost 100% and 200% were not significant difference in the dry matter yield from chemical fertilizer, but cattle manure compost 50% was (p $\langle 0.05\rangle$) low productivity of roughage. The dry matter yield of bermuda grass pasture depending on the application level of fermented cattle manure compost was found that the larger amount of cattle manure compost application was associated with the higher dry matter yield of bermuda grass, but there was no significant differences. Based on the findings of this study, it is found that organic roughage pasture can be produced on the pasture of both orchard grass and bermuda grass with the application of fermented cattle manure compost (nitrogen 200 kg/ha; based on application of chemical fertilizer) across warm regions including Jeju Island. And it is expected that the specification of livestock will be properly managed through graze breeding without concentration, ultimately contributing significantly to management of farmhouses in livestock industry.

Table 13-3. The dry matter yield of orchard grass pasture depending on the application of cattle manure compost

Treatments	2008	2009	Mean	
chemical fertilizer (N-200kg/ha)	19,808 ^{ab}	18,524	19,166 ^{ab} (100)	
cattle manure compost 50%	17,377 ^b	15,299	16,338 ^b (86)	
cattle manure compost 100%	18,982 ^a	17,757	18,369 ^{ab} (96)	
cattle manure compost 200%	22,578 ^a	17,073	19,825 ^a (104)	

 $^{^{}a-b}$ Means in the same column with different superscripts are significantly different (p $\langle 0.05\rangle$



National Institute of Animal Science

Table 13-4. The dry matter yield of bermudagrass pasture depending on the application of cattle manure compost

Treatments	1-time cut (Jun _. 3)	2-time cut (Jul. 20)	3-time cut (Aug. 20)	4-time cut (Sept _. 9)	Total
chemical fertilizer (N-200kg/ha)	3,098	3,308 ^a	3,687 ^a	3,497 ^b	13,590 ^a
cattle manure compost 50%	2,878	2,839 ^b	1,730 ^b	3,522 ^b	10,969 ^b
cattle manure compost 100%	3,246	3,118 ^{ab}	1,606 ^b	3,110 ^b	11,080 ^{ab}
cattle manure compost 200%	2,780	3,434 ^a	1,866 ^b	4,341 ^a	12,420 ^{ab}

 $[\]overline{a^{-b}}$ Means in the same column with different superscripts are significantly different (p $\langle 0.05\rangle$