



Association analysis of thirty-one single nucleotide polymorphisms with antler weight in sika deer

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Sika deer (*Cervus nippon*) were once widely distributed in eastern Asian countries and comprise one of the most commercially valuable species for the production of velvet antler by the males for medicinal use.¹ Large herds of sika deer are kept mainly for the production of velvet antlers. As the profitability of farming depends on the antler weight, the identification of molecular markers associated

with antler weight offers potential for selective breeding of sika deer.

In this study, we carried out an association analysis using SNPs detected by a previous GWAS of antler weight² (PRJNA541418), and transcriptome sequencing for males with different antler weights³ (PRJNA470791). We first selected 100 annotated SNPs corresponding to GWAS peaks and located in or near genes with expression levels positively correlated with antler weight (Table S1). Population genetic indices including expected heterozygosity (H_e), observed heterozygosity (H_o), HWE and MAF were calculated using HAPLOVIEW 4.1 software, and the results are shown in Table S1.

From the 100 SNPs, we selected 31 SNPs on the basis of SNP detection rate and MAF (SNP detection rate >80% and MAF >0.1; from SNPs within the same gene, the SNP with the highest MAF value was selected). These SNPs were used for genotyping 341 farmed sika deer from northern China with heavy and light antlers (Fig. 1a,b) as recorded at an age of 5–6 years during the rapid antler growth stage after about 75 days of growth (Table S2).

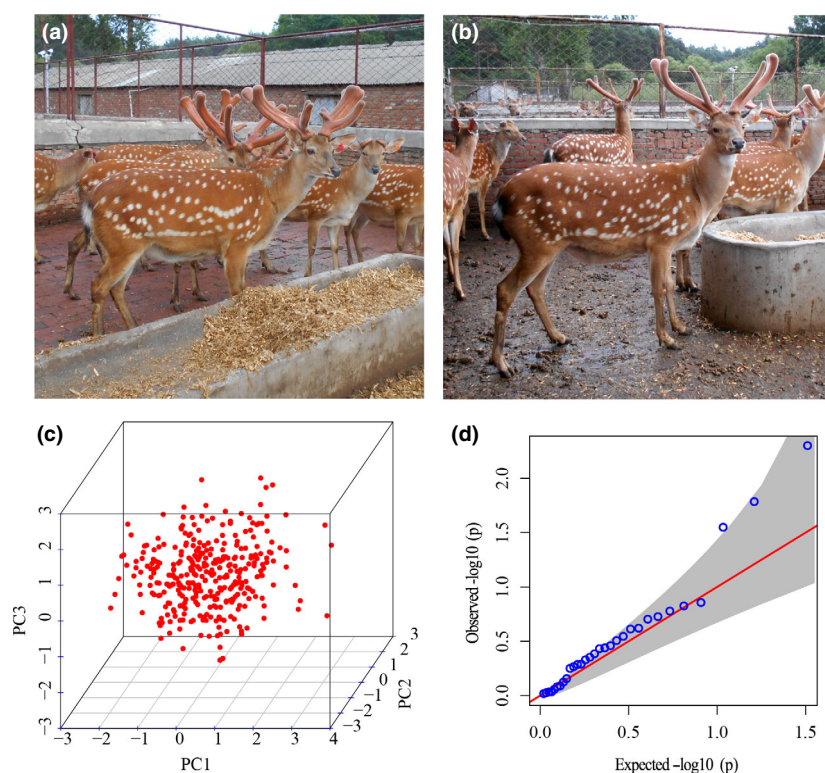


Figure 1 Sika deer with (a) heavy and (b) light antlers. (c) PCA analysis of 341 farmed sika deer using 31 SNPs. (d) Q-Q Plot analysis of 31 SNPs

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Genotyping was done using the MassARRAY SNP detection technology and primers listed in Table S3. Association analysis was performed using the linear regression model in SPSS STATISTICS 22 software. PCA, Q-Q plots and allele effects were calculated using the R-GAPIT package.

The results revealed that the effect of population stratification has been avoided (Fig. 1c,d), and that 16 SNPs were significantly associated with antler weight (Tables S4 and S5).

Several candidate genes related to antler weight have been obtained in previous studies,^{3–5} but these did not contain SNPs that can be used for marker-assisted selection. Testing of the *OAS2* and *ALYREF/THOC4* mutations² showed that *OAS2* is not expressed in the antler and that the expression level of *ALYREF/THOC4* is not positively correlated with antler weight, so they were excluded in this study. The rapid growth of antler is a complex process controlled by multiple genes, and nutritional and environmental factors also contribute to antler weight, so further study with a large sample size is needed to confirm the genetic effects of these SNPs.

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Conflict of interest

The authors declare that they have no competing interests.

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Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1. The 100 SNPs associated with antler weight

Table S2. The 31 SNPs genotyped in 341 farmed sika deer with antler weight recorded at rapid antler growth stage

Table S3. Primer information for 31 SNPs

Table S4. Association analysis results of 31 SNPs with antler weight

Table S5. Genotype frequency and antler weight differences between genotypes of candidate SNPs