Genetics and Molecular Research

Isolation and characterization of novel microsatellite markers from the sika deer (*Cervus nippon*) genome

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Genet. Mol. Res. 14 (3): 11524-11534 (2015)
Received December 3, 2014
Accepted June 18, 2015
Published September 28, 2015
DOI http://dx.doi.org/10.4238/2015.September.28.4

ABSTRACT. Microsatellite markers are widely and evenly distributed, and are highly polymorphic. Rapid and convenient detection through automated analysis means that microsatellite markers are widely used in the construction of plant and animal genetic maps, in quantitative trait loci localization, marker-assisted selection, identification of genetic relationships, and genetic diversity and phylogenetic tree construction. However, few microsatellite markers remain to be isolated. We used streptavidin magnetic beads to affinity-capture and construct a $(CA)_n$ microsatellite DNA-enriched library from sika deer. We selected sequences containing more than six repeats to design primers. Clear bands were selected, which were amplified using non-specific primers following PCR amplification to screen polymorphisms in a group of 65 unrelated sika deer. The positive clone rate reached 82.9% by constructing the enriched library, and we then selected positive clones for sequencing. There were 395 sequences with CA repeats, and the CA repeat number was 4-105. We selected sequences containing more than six repeats to design primers, of which 297 pairs were designed. We next selected clear bands and used

non-specific primers to amplify following PCR amplification. In total, 245 pairs of primers were screened. We then selected 50 pairs of primers to randomly screen for polymorphisms. We detected 47 polymorphic and 3 monomorphic loci in 65 unrelated sika deer. These newly isolated and characterized microsatellite loci can be used to construct genetic maps and for lineage testing in deer. In addition, they can be used for comparative genomics between Cervidae species.

Key words: Sika deer; Microsatellite markers; Enriched library; Polymorphism

