

Construction of an Indian Muntjac BAC Library and Production of the Most Highly Dense FISH Map of the Species

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(Accepted October 19, 2007)

Chyi-Chyang Lin, Pei-Ching Hsu, Tzai-Shiuan Li, Shu-Ju Liao, Ya-Ming Cheng, Lie-Jiau Hsieh, and Yueh-Chun Li (2008) Construction of an Indian muntjac BAC library and production of the most highly dense FISH map of the species. *Zoological Studies* 47(3): 282-292. Following completion of the genome sequences of some mammalian species, comparative genomic studies in mammals have been actively conducted to assess gene changes or to identify syntenic conservation during evolution. The Indian muntjac (*Muntiacus muntjak vaginalis*) ($2n = 6$ in the female and 7 in the male) may have evolved from an ancient deer species with a karyotype $2n = 70$ through extensive chromosome rearrangements creating the lowest chromosome number of a mammalian species. Therefore, the species has become a good resource for studying syntenic conservation among deer species. An Indian muntjac bacterial artificial chromosome (BAC) library that contains 126,336 individual BAC clones with an average insert size of 80 kilobases was obtained in this study. The frequency of clones with inserts was 88%, and thus this library corresponds to approximately 4x coverage of the Indian muntjac genome. Individual chromosomal locations of 1619 BAC clones on the Indian muntjac metaphase chromosomes were identified by fluorescence *in situ* hybridization (FISH). Among these clones, 1517 BAC clones were mapped onto specific loci, and 102 BAC clones were mapped onto the centromeric region. This provides the most highly dense FISH BAC clone map for the species. This densely ordered map can be used as a blueprint for comparative FISH mapping studies of other deer species in order to investigate the mechanism of genomic rearrangement and karyotypic evolution. Moreover, centromeric BAC clones will provide an excellent resource for studying the structure and function of mammalian centromeres.
<http://zoostud.sinica.edu.tw/Journals/47.3/282.pdf>

Key words: BAC library, FISH mapping.

With the completion of the sequencing of the human, rat, and mouse genomes (Lander et al. 2001, Venter et al. 2001, Waterston et al. 2002, Gibbs et al. 2004) more-accurate and higher-resolution comparative mapping studies have been achieved among 8 phylogenetically distinct species (human, horse, mouse, rat, cat, dog, pig, and cattle). Those studies found that nearly 20% of chromosome breakpoint regions were reused during mammalian evolution. In addition, it was shown that the reused breakpoint regions were significantly associated with centromeric sequences (Murphy et al. 2005). An early study

by Frönicke and Scherthan (1997) revealed that centromeric satellite DNAs cluster at the margins of conserved syntenic segments based on human-Indian muntjac Zoo-fluorescent *in situ* hybridization (FISH) experiments. Other FISH analyses also shown that centromeric satellite DNA and telomeric DNA are located at interstitial chromosomal sites on Indian muntjac (*Muntiacus muntjak vaginalis*) chromosomes, representing a remnant of chromosome break and fusion sites during restructuring of the Indian muntjac karyotype (Lin et al. 1991, Lee et al. 1993, Scherthan 1995, Li et al. 2000). Thus it is possible that evolutionary

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