Abstract

The diploid amphibian Xenopus tropicalis represents a significant model organism for studies of early development, genes function and evolution. Such techniques as gynogenesis, injection of morpholino antisense oligonucleotide into fertilized eggs or transgenesis were established. In the recent ten years, many efforts have been made to complete the sequence information. X. tropicalis genome has been sequenced but the completion of its assembly only on the basis of sequence data has been impossible. Therefore, our first work was focused on one of approaches for a genome completing- genetic mapping. First of all, the genetic map of Xenopus tropicalis was established pursuant linkage and physical positions of markers. Since the map contained gaps, we developed a new method for genetic mapping based on the next generation sequencing of laser microdissected arm. Using Illumina next generation sequencing of fifteen copies of a short arm of chromosome 7, we obtained new insights into its genome by localizing previously unmapped genes and scaffolds as well as recognizing mislocalized portions of the genome assembly. This was the first time laser microdissection and sequencing of specific chromosomal regions has been used for the purpose of genome mapping. These data were also used in the evolution study of the sex determining area placed on the q arm of chromosome 7, which showed that Xenopus tropicalis sex chromosomes contain large pseaudoautosomal areas. Moreover, we made Zoo-FISH analysis using X. tropicalis microdissected chromosomes as probes for labeling Xenopus laevis chromosomes, which revealed similarity of meiotic quartets even after 65 million years of separate evolution. Our novel approach for next generation sequencing of microdissected chromosomal area is also applicable to species without sequenced genomes or for clinical applications in medical cytogenetics and oncology where tissue availability may be limiting. This method is likely to be of widespread use in species where individual chromosomes are distinguishable by cytological methods.