Abstract

To study hybrid sterility our laboratory uses mouse strains PWD/Ph (PWD), derived from *Mus musculus musculus* wild mice and the common laboratory strain C57BL/6J (B6) mostly of *Mus musculus domesticus* origin as a model. Crossing between PWD female and B6 male results in sterile male progeny. F1 hybrid males carry defects in the repair mechanisms of asymmetric double-strand DNA breaks (DSBs). Functional interplay of SPO11 and PRDM9 proteins in the meiotic prophase I is necessary for repairs. Its defect leads to incorrect synapse formation between homologous chromosomes, leading to halt in spermatogenesis and thus male sterility. The formation of DSBs and their subsequent repair is essential for first meiotic division. The working hypothesis stems from the findings in yeast model, where supposed antirecombinatorial mechanism of mismatch repair genes *Msh6* and *Msh2* prevents DSBs repairs during meiosis. Despite the functional mechanism of these two genes is not explicitly known, existence of similar repair system in mice is presumed.

Variety of methods was implemented in this thesis. The effects of *Msh6* deletion on meiotic prophase I and sperm maturation were performed by designing guide RNAs for CRISPR/Cas9 for creation of three knock-outs in B6 mice. The PCR was used to amplify regions adjacent to the deletion, run on agarose gel electrophoresis and, in the end followed by Sanger sequencing. The existence of the null mutant was verified by western blot. Immunoflorescent microscopy with specific protein markers served for monitoring of individual meiotic stages. As of now, homozygous knock-out *Msh6* lines have been successfully grown up to fifth backcross generation. The B6.*Msh6*$^{-/-}$males had significantly lower testes weight as well as number of mature sperm when compared to B6 wild-type control. Males carrying the deletion exhibited the symptoms of unrepaired DSBs at the pachytene stage. The effect of *Msh6* gene deletion in B6 males on the meiotic recombination rate has not been demonstrated.

Key words: mismatch repair, meiotic recombination, hybrid sterility, congenic strains