Abstract

Hybrid sterility is one of the reproductive isolation mechanisms restricting gene flow between the related species and leading to speciation. PR domain containing 9 (*Prdm9*), the only known vertebrate hybrid sterility gene, determines the sites of programmed DNA double-strand breaks (DSBs) and thus specifies hotspots of meiotic recombination but in hybrids between two mouse subspecies causes failure of meiotic chromosome synapsis and hybrid male sterility.

In the present study on sterile hybrids, the five smallest autosomes were more prone to asynapsis. To manipulate with the synapsis rate, random stretches of consubspecific homology were inserted into several autosomal pairs. Twenty seven or more megabases of consubspecific sequence fully restore synapsis in a given autosome. Further, at least two symetric DN double-strand breaks per chromosome were necessary for successful synapsis. Moreover, F_1 hybrids had sperm when synapsis was rescued in at least three of four segregating chromosomes.

To verify the assumption of a lack of symmetric DSBs in meiotic chromosomes of sterile males the chemotherapeutic drug cisplatin was used to induce exogenous DNA DSBs. Cells treated with 5 mg/kg and 10 mg/kg of cisplatin showed increased number of DSBs monitored by immunostaining of RPA and DMC1 sites and increased proportion of spermatocytes with fully synapsed homologs at pachytene.

The *Prdm9* gene and *Hstx2* locus are the known necessary components controlling the F_1 hybrid sterility. Hybrids with the part of genome derived from *Mus musculus castaneus* were used to verify the hypothesis that genotype $Prdm9^{PWD/B6}$ and $Hstx2^{PWD}$ were important for asynapsis and meiotic arrest in these hybrids. Hybrids with $Prdm9^{CAST/B6}$ allelic combination were fully fertile while the „sterile“ genotype $Prdm9^{PWD/B6}$ showed a range of phenotypes from full sterility with a high rate of asynapsis to fertility and quasi-normal meiotic chromosome synapsis. Thus, besides *Prdm9*, two or more CAST modifing genes could play a role in this cross. The $Hstx2^{CAST}$ and $Hstx2^{PWD}$ alleles had the same effect on fertility of hybrids, indicating that *Prdm9* gene acts as the only one major hybrid sterility gene in these hybrids derived from three subspecies *Mus musculus musculus, Mus musculus castaneus* and *Mus musculus domesticus*.