

Publication

A genetic system to detect mitotic recombination between repeated chromosomal sequences in Drosophila Schneider line 2 cells

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In order to study mitotic homologous recombination in somatic Drosophila melanogaster cells in vitro and to learn more on the question how recombination is influenced by mutagens, a genetic system was developed where spontaneous and drug-induced recombination could be monitored. Two recombination reporter substrates were stably introduced in multiple copies into the genome of established D. melanogaster Schneider line 2 cells: one plasmid (pSB310) contained the 5' and 3' deleted neomycin phosphoribosyltransferase alleles neoL and neoR as direct repeats; the other (pSB485) contained similar deletions (lacZL and lacZR) of the beta-galactosidase gene (lacZ). Restoration of a functional neo gene upon mitotic recombination between homologous sequences allowed direct selection for the event, whereas recombination in single cells harbouring the integrated lacZ-based reporter plasmid was detected by histochemical staining or flow cytometric analysis (FACS). The neo-based construct in the clonal transgenic cell line 44CD4 showed a spontaneous recombination frequency of 2.9×10^{-4} , whereas the 485AD1 cell line harbouring the lacZ-based construct exhibited a frequency of 2.8×10^{-4} . The alkylating agents EMS and MMS and the clastogen mitomycin C were able to induce recombination in the 485AD1 cell line in a dose-dependent manner. The results obtained from these studies suggest that the transgenic cell lines are potentially useful tools for identifying agents which stimulate direct repeat recombination in somatic Drosophila cells.

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