Bug Transposons

Nancy L. Craig, Howard Hughes Medical Institute and Department of Molecular Biology & Genetics, Baltimore MD 21295 USA

We study the transposition of 2 cut & paste transposons, Tn7 from E. coli and Hermes from the house fly, both hosts being bugs although of different sorts. Tn7 is a member of the widespread Retroviral Integrase Superfamily and Hermes of the hAT family that includes McClintock's Ac element.

The Hermes system is relatively simple: the transposase is comprised of a single polypeptide. We have established that recombination proceeds via double strand breaks that excise the element from the donor site, followed by joining to the target DNA. Interestingly, the breaks occur as in V(D)J recombination with hairpins being formed on the flanking donor DNA. The exposed 3'OH ends of the transposon then join to the target DNA. We have also established a Hermes transposition system in Saccharomyces cerevisiae and have used a genetic screen to isolate transposase mutants that promote high frequencies of transposition. We have also used site-directed mutagenesis to probe structure-function relationships of the transposase

The Tn7 transposition system is far more elaborate. Tn7 insertion into its preferred chromosomal site attTn7 requires 4 Tn7-encoded proteins TnsABC+D and an essential ATP cofactor. We have found that after the DNA breakage and joining events that result in transposition have occurred, the transposition product is still included in an elaborate nucleoprotein complex. Notably, the gaps the flank the ends of the newly inserted transposon are inaccessible to a repair polymerase in this complex. We are using a variety of methods to probe this complex.