Positional cloning of the maternally-acting, selfish gene, *Medea*[1], in *Tribolium castaneum*.

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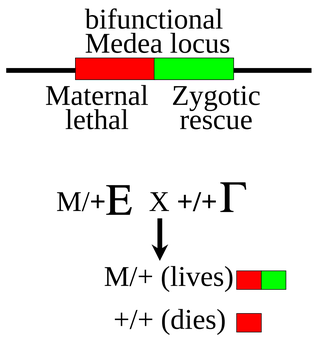
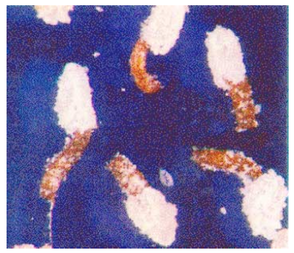
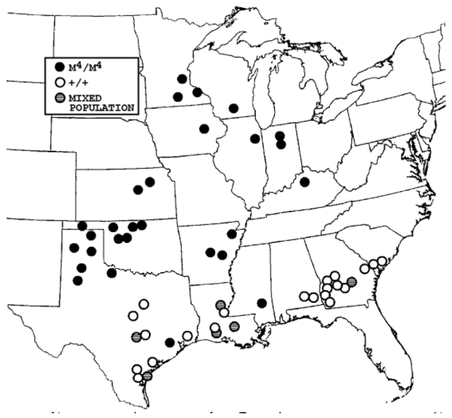


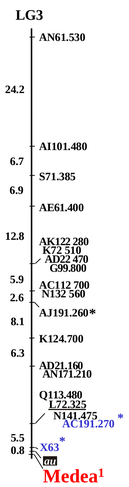


Medea-killed hatchlings

(Larvicidal action is by conferred by maternal lethal effect. Selfish behavior is conferred by zygotic rescue from the maternal lethal effect)



Medea (M^4) distribution in the U.S.A. N. latitude 33° separates northern (Medea) populations from southern (non-Medea) populations.

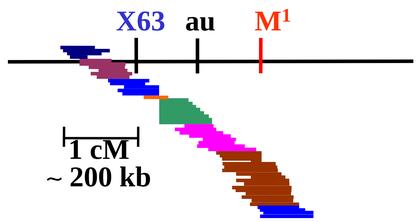


Medea and H genes are incompatible:

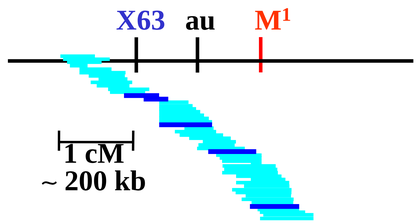
M/M X H/H → all progeny die
H/H X M/M

Medea[1] maps to the far end of the third linkage group, near the RAPD marker **X63**. This marker was used as starting point for a chromosome walk.

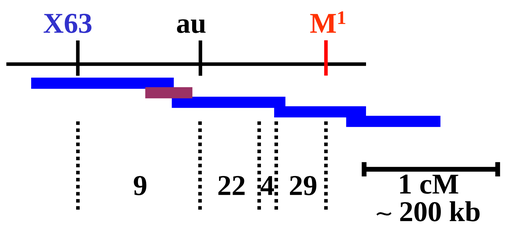
For each step of the walk, either of two BAC libraries was hybridized with 36-mer "overgo" probes from end-sequences of BACs identified in the previous step. BACs were ordered and contigged by PCR based on end-sequences. The walk was oriented by high-resolution recombination mapping internal to the contig, left-of-Medea.



chromosome walk from X63 to Medea, in two BAC libraries

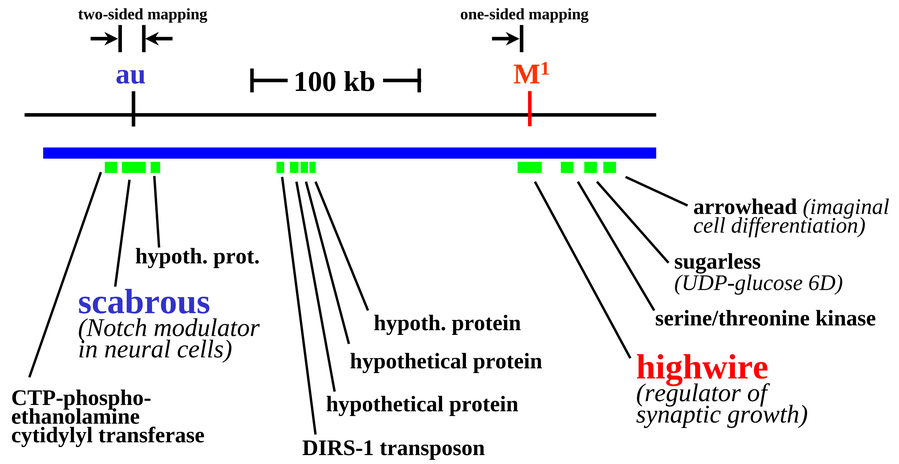


BACs indicated in dark blue were shotgun-sequenced



High-resolution recombinational mapping within sequenced BAC contig. Total numbers of recoms analyzed per interval are indicated (ca. 7000 chromosomes screened).

Gene map of *au*-to-*Medea* region



The *aureate* and *Medea* loci were positioned on the molecular sequence map using very high-resolution recombinational mapping (average recombinant spacing of 3 kb). The *aureate* and *Medea* loci map to within approximately 3 kb of *scabrous* and *highwire*, respectively. The localization of *Medea* relied on one-sided mapping, and therefore assumes the absence of a recombination coldspot in the immediate vicinity of that locus.

Summary and conclusions: **We have demonstrated the feasibility of positional cloning in *Tribolium* by chromosome walking in a BAC library. Two genes, *aureate* and the unique, maternal selfish gene *Medea*, defined only by phenotypic effect, were cloned and mapped to the *scabrous* and *highwire* regions, respectively, using very high-resolution recombinational mapping. Confirmation will include molecular mapping of seven *Medea* revertant (knockout) lesions induced by radiation, mapping of one spontaneous and one radiation-induced mutant lesion in *aureate*, and expression analysis of the candidate genes in mutant beetles.**