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Aedes aegypti: Origin of a "New" Chromosome from a Double Translocation Heterozygote

Abstract. *An apparent enhancement of crossing-over occurs in the region common to two translocated chromosomes derived from two different reciprocal translocations in the yellow-fever mosquito,* Aedes aegypti. *A "new" chromosome containing parts of all three linkage groups is created through crossing-over. In a reversible process, two haploid translocated sets in the double translocation heterozygote produce a new, normal chromosome set and vice versa in alternating generations.*

Genetic control mechanisms for insect pests are currently being developed for a number of important species. To date, the most successful genetic mechanism has utilized the production of dominant lethal mutations by gamma radiation in males of the screw worm fly (*1*). Although sterilized males effectively reduce population size in the generation after their release, they do not convey sterility to members of the next generation; thus, continued reintroductions are necessary to maintain control.

The use of the semisterility inherent in some chromosomal aberration systems provides one means for introducing this inherited sterility into natural populations. The use of a reciprocal translocation system for insect control was originally postulated by Serebrovskii in 1940 (*2*). More recently Curtis (*3*) has indicated the potential effectiveness of translocation systems in insect control programs. In addition to the inherited semisterility, the translocation systems are usually associated with an essentially unaffected genotype and phenotype and may not therefore affect the competitive ability of released insects. Such reduced competitive ability often becomes a limiting factor in mutagen-induced dominant lethality. As a result, over the past few years several investigators have been interested in the use of chromosomal aberrations for the control of insect pests (*4*). Wagoner *et al.* (*5*) and Laven (*6*) have reported inducing several translocations in the house fly, *Musca domestica*, and the mosquito, *Culex pipiens*, respectively.

In our laboratory, we have been interested in inducing reciprocal translocations in the yellow-fever mosquito, *Aedes aegypti* ($2n = 6$) and evaluating the same for population control purposes. We have recently completed genetic analysis on two sex-linked translocations. One, RT(1:2) involves linkage groups one and two (*7*), with the original break points 0.3 crossover unit from the gene for sex (*M*) on group one and 1.6 crossover units from the wild type allele of spot abdomen (s^+) on group two. The other translocation, RT(1:3) involves linkage groups one and three, with the original break points 1.0 crossover unit from the wild type allele of red eye color (re^+) on group one and 0.5 crossover unit from the wild type allele of black tarsus (blt^+) on group three (*8*). It may be mentioned that sex in this species is determined by a single pair of alleles, *M* and *m*; the genotype *Mm* producing maleness and *mm* femaleness (*9*).

In an effort to determine the effectiveness of individuals carrying more than one translocation in reducing population sizes, a program was undertaken to produce individuals that would be heterozygous for both RT(1:2) and RT (1:3). Cytological analysis of these individuals would also provide information enabling the assignment of linkage groups to the individual chromosomes.

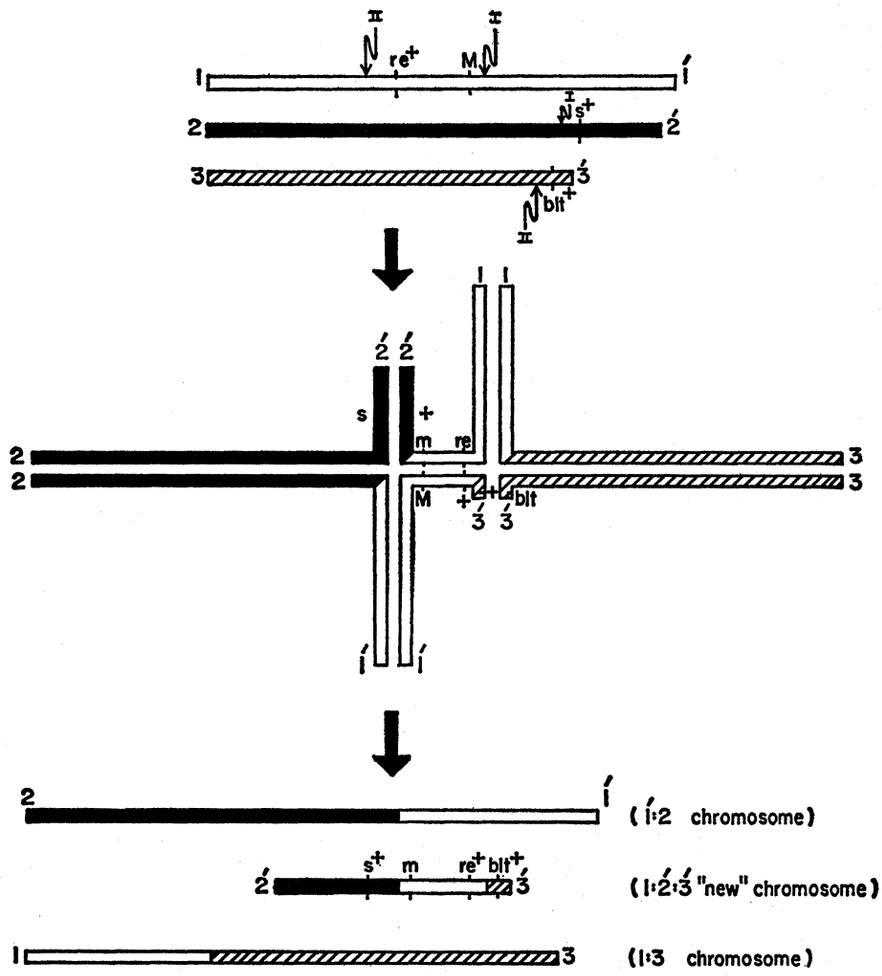

Fig. 1. Derivation of a "new" chromosome from a double translocation heterozygote. The three linkage groups (1-1́, 2-2́, 3-3́) are drawn to scale; the approximate location of the genetic markers used and the original radiation-induced breaks I and II producing RT(1 : 2) and RT(1 : 3), respectively, are indicated (top portion of the figure).

Table 1. Progeny from crosses involving *RED* stock females [homozygous for the genes for red eyes (*re*), spot abdomen (*s*), and black tarsi (*blt*)] and males heterozygous for both translocations and the markers (group A) or new karyotype males (RT1 : 2 : 3) heterozygous for the markers (group B).

Genotype of male parent	Progeny recovered	
	Non-crossovers	Crossovers
	Group A males	
$\frac{+\ M\ re\ blt}{s\ m\ +\ +}$ (Double heterozygote)	11 re$^+$ blt$^+$ s ♀♀ (RT1 : 3) 13 re blt s$^+$ ♂♂ (RT1 : 2) 4 re blt s ♂♂ (Normal or RT1 : 2) 1 re$^+$ blt$^+$ s$^+$ ♀♀ (RT1 : 3 or RT1 : 2 : 3)	12 re$^+$ blt$^+$ s$^+$ ♂♂ (RT1 : 2 : 3) 14 re blt s ♀♀ (Normal)
$\frac{+\ m\ re\ blt}{s\ M\ +\ +}$ (Double heterozygote)	17 re$^+$ blt$^+$ s ♂♂ (RT1 : 3) 8 re blt s$^+$ ♀♀ (RT1 : 2) 2 re blt s ♀♀ (Normal or RT1 : 2) 1 re$^+$ blt$^+$ s$^+$ ♂♂ (RT1 : 3 or RT1 : 2 : 3)	8 re$^+$ blt$^+$ s$^+$ ♀♀ (RT1 : 2 : 3) 11 re blt s ♂♂ (Normal)
	Group B males	
$\frac{+\ M\ +\ +}{s\ m\ re\ blt}$ (RT1 : 2 : 3)	7 re$^+$ blt$^+$ s$^+$ ♂♂ (RT1 : 2 : 3) 9 re blt s ♀♀ (Normal) 2 re blt s$^+$ ♀♀ (Normal or RT1 : 2)	9 re$^+$ blt$^+$ s ♀♀ (RT1 : 3) 7 re blt s$^+$ ♂♂ (RT1 : 2)

Two interesting, albeit unusual, results have emerged from this work. First, a new chromosome with new linkage relationships has been established. As a result of a crossover between the sex and the red-eye genes in the double translocation heterozygous individual, a chromosome containing parts of linkage groups one, two, and three has been created (Fig. 1). This chromosome, together with two of the translocated chromosomes, forms a haploid set that is capable of functioning in the presence of a normal haploid set in a way that allows normal development to the adult stage. The known genetic lengths of linkage groups one, two, and three are 38, 37, and 30 crossover units, respectively (*10*). The original radiation-induced breaks producing RT(1:2) and RT(1:3) and a genetic exchange in the double heterozygote in the region between sex and red-eyed genes will be expected to change the standard genetic lengths to 18, 43, and 44 crossover units. Thus, a very small "new" chromosome has been established.

The second unusual aspect of this double heterozygote system is an apparent enhanced crossing-over that occurs in the regions between the genes for sex and red eyes as well as between genes for sex and spot abdomen. This has the effect of producing a higher than expected frequency of individuals with the new karyotype from double translocation heterozygotes and conversely, a higher than expected frequency of individuals heterozygous for a translocation from individuals of the new karyotype.

In crosses between *RED* stock females (which are homozygous for the recessive markers red eyes (*re*), spot abdomen (*s*), and black tarsi (*blt*) on linkage groups one, two, and three, respectively) and males heterozygous for both translocations and all these markers, 45 of the 102 progeny resulted from crossing-over between the genes for sex and red eyes, giving an apparent map distance of 44.2 crossover units instead of 6.8 crossover units for RT(1:3). Of the 102 progeny, 8 were crossovers between sex and spot for a distance of 7.8 compared with 1.9 crossover units for RT(1:2) (Table 1). When males with the new karyotype obtained from the above crosses were crossed with *RED* stock females, the rate of crossover was (16 out of 34) 47.1 percent between sex and red, and (2 out of 34) 5.9 percent between sex and spot (Table 1). Because of this apparent enhanced recombination, data in Table 1 approximate a 1:1:1:1 segregation of RT(1:2), RT (1:3), new translocation karyotype RT (1:2:3), and normal individuals.

The occurrence of exchange in the region between the sex and the red-eyed loci in this system may facilitate the orientation of the interchange complex on the metaphase plate so as to produce the functional haploid sets different from the parental haploid sets. This would work in both cases: the double heterozygote parent has two RT heterozygote haploid sets and produces the new karyotype set plus a normal karyotype set; and the new karyotype individual has one new and one normal haploid set and produces the two translocation heterozygous haploid sets. The recombinant progeny included in Table 1 leave no doubt that this is indeed what happens.

In the normal stock the distance between sex and red-eyed gene is approximately 7 crossover units. Thus it may be expected that 7 percent of the gametes will result from a crossover in this region. In the *RED* stock where fertility is ordinarily 65 percent, crossovers could account for 7 percent of 65 percent fertility or 4.6 percent fertility. In crosses of *RED* with double heterozygote and of *RED* with new karyotype, the fertility was usually between 7 percent and 10 percent, and it may be possible for the 44.2 to 47.1 percent recombinants to arise from preferential recovery of the crossovers without any increase in frequency of the actual physical exchanges in this region.

The advantages of the use of the double translocation heterozygote for population control cannot be overemphasized. Whereas in crosses with *RED* genotype the fertility of each translocation heterozygote [RT(1:2) and RT (1:3)] is around 30 percent, the fertility of double heterozygotes ranges from 7 to 10 percent. Furthermore, they generate into a breeding population RT (1:2), RT(1:3), the new translocated karyotype RT(1:2:3), and the normal (standard homozygotes) individuals. Thus the rate of population suppression after releases of such males should be much more rapid than that after releases of single translocation stocks.

The production of a new chromosome with changed morphology originating from a genetic exchange in a double translocation heterozygote may also provide an evolutionary means for the origin of new karyotypes in nature.

P. T. McDonald
K. S. Rai
Department of Biology, University of Notre Dame, Indiana 46556

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