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On Recombination Among *In(2L)t*, *α-Gpdh* and *Adh* in *Drosophila melanogaster*

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ABSTRACT

The occurrence and patterns of linkage disequilibrium between an inversion and allozymic loci within it or nearby have been used in attempts to discriminate among different hypotheses for the maintenance of variability. The interpretation of the data on the best-documented case, that of the nonrandom association between *In(2L)t* and *α-Gpdh* or *Adh* in the second chromosome of *Drosophila melanogaster*, has been done on the basis that recombination between *α-Gpdh* and *Adh* is almost entirely due to the recombination between *In(2L)t* and the locus within it (*α-Gpdh*), the recombination between the inversion and the nearby locus (*Adh*) being negligible. In this paper, we show that the pattern of recombination is just the opposite.

OBSERVATIONS of linkage disequilibrium between an inversion and allozymic loci within it or nearby has variously been proposed to be proof of selection working upon the system (Prakash and Lewontin 1968), as a relic of an unique event (Nei 1975), and as the consequence of random drift with or without selection acting upon the inversion (Nei and Li 1980). These alternative explanations can be distinguished by collating the observed patterns of those associations with those expected under the different hypotheses. These expectations rely heavily on the recombination fraction (*rf*) between the markers that are considered.

The best-documented case is that of the nonrandom association between *In(2L)t* and *α-Gpdh* (*II*, 17.8) or *Adh* (*II*, 50.1) in the second chromosome of *Drosophila melanogaster*. In this paper, we report our results on the recombination among these markers.

MATERIALS AND METHODS

Experimental stocks: A near isogenic line for the second chromosome with *In(2L)t*, *α-GpdhF AdhS* phenotype extracted from the population Aspe (Malpica and Vassallo 1980) using *CyO* (Lindsley and Grell 1968) as balancer was used throughout this study. No instability was observed in the progeny of the crosses. Flies were reared at 25° on cornmeal-molasses medium.

Enzyme assays: Horizontal starch gel electrophoresis with a buffer system Tris-versene-borate (0.05 M, pH = 8, for the gel, 0.5 M in the tanks) was used. Staining recipes were those of Shaw and Prasad (1970).

Cytological analysis: Recombinant chromosomes were extracted as above. These near-isogenic stocks were crossed with *al b pr cn* stock carrying the standard arrangement. From these crosses, third-instar larvae were scored for inversions by temporary propionic-orceine-carmine squash preparations of salivary gland chromosomes (Becker 1962). In all cases the observed inversion had the same breaking points as *In(2L)t*, as reviewed by Ashburner and Lemeunier (1976).

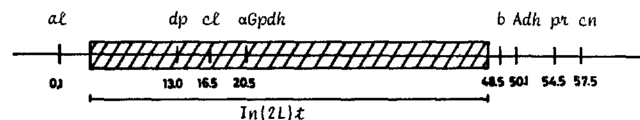

FIGURE 1.—Map position of the markers.

RESULTS AND DISCUSSION

Recombination fractions in this case were estimated by Mukai and Voelker (1977). They used the cross

$$\frac{\{In(2L)t\ \alpha\text{-}Gpdh^F\}\ Adh^S}{\{\ +\quad \alpha\text{-}Gpdh^S\}\ Adh^F}(♀) \times \frac{\{+\ \alpha\text{-}Gpdh^S\}\ Adh^F}{\{+\ \alpha\text{-}Gpdh^S\}\ Adh^F}(♂)$$

Electrophoresis of the resulting progeny indicated one *α-Gpdh-Adh* recombinant among 4377 gametes. They attributed this event to recombination due to a double crossover within the inversion, on the basis of the cytological location of the markers (Figure 1).

We have checked the hypothesis that crossing over does not occur in the region between the right end of *In(2L)t* and *Adh*, using the cross

$$\frac{\{In(2L)t\}\ +\ Adh^S\ +}{\{\ \ +\ \ \}\ b\ Adh^F\ pr}(♀) \times \frac{\{+\}\ b\ Adh^F\ pr}{\{+\}\ b\ Adh^F\ pr}(♂)$$

On a total of 5248 progeny, 18 were recombinants between *b* and *pr*. These animals are electrophoresed, and two were found to be recombinants between *b* and *Adh*, giving an estimated $rf = 3.8 \times 10^{-4}(4.2 \times 10^{-5} - 1.4 \times 10^{-3}, P = 0.95)$. Recombination in the region, therefore, does occur, and the *rf* estimated is in the range of the total *rf* between *Adh* and *α-Gpdh* given by Mukai and Voelker (2.3×10^{-4}).

We also studied the cross

$$\frac{+\ \{In(2L)t\ +\ +\ \alpha\text{-}Gpdh^F\}\ +\ Adh^S\ +\ +}{al\ \{\ \ +\ \ \ dp\ cl\ \alpha\text{-}Gpdh^S\}\ b\ Adh^F\ pr\ cn}\ (♀)\ \times$$

$$\frac{al\ \{\ +\ dp\ cl\ \alpha\text{-}Gpdh^S\}\ b\ Adh^F\ pr\ cn}{al\ \{\ +\ dp\ cl\ \alpha\text{-}Gpdh^S\}\ b\ Adh^F\ pr\ cn}\ (♂)$$

Among 3761 progenies, 82 recombinants were observed, all of them in the region *b-cn*. These were electrophoresed, and their cytological constitution assessed in their progeny. Two of them were recombinants between *b* and *Adh* ($rf = 5.3 \times 10^{-4}$, $6.0 \times 10^{-5} - 1.9 \times 10^{-3}$, $P = 0.95$). In the cytological examination, presence of *In(2L)t* was always linked with the wild-type allele of *b*.

These results indicate that double crossover within *In(2L)t* are irrelevant as a component of the recombination fraction between *α-Gpdh* and *Adh*, because no recombinants were observed between the *b* and a marker (*cl*, *II*, 16.5) very close to *α-Gpdh* (*II*, 17.8). The recombination fraction between *b* and *Adh* can, again, account for the total recombination observed. Small frequencies of double crossovers within large inversions, even in the range of gene conversion, are not uncommon (see ISHII and CHARLESWORTH 1977).

We therefore propose that the recombination fraction between *α-Gpdh* and *Adh* in heterozygotes for *In(2L)t* is almost entirely due to the recombination between the inversion and the *Adh* locus, the recombination between the inversion and *α-Gpdh* being negligible.

The interpretation of published data on linkage disequilibrium between these markers should be revised accordingly. Of particular interest is the suggestion that the present pattern of linkage disequilibrium for *α-Gpdh-In(2L)t* and for *Adh-In(2L)t* could be explained by the hypothesis of a unique occurrence of *In(2L)t* and a decay of linkage disequilibrium since then (MUKAI and VOELKER 1977; YAMAGUCHI *et al.* 1980). There are two arguments for this hypothesis, the congruences between (1) the strength of linkage disequilibria and the estimates of recombination fraction, and (2) the estimates of time since the introduction of the inversion in a population based on the nonrandom association *α-Gpdh-In(2L)t* and that based on the genetic variance analysis of sternopleural and abdominal bristle number. Both of these congruences depend on the assumption that the recombination fraction between *α-Gpdh* and *Adh* is mainly due to the recombination between *α-Gpdh* and *In(2L)t*, and they do not hold under our results.

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