Spindle-Fiber Attachment End of the X-Chromosome of *Drosophila virilis* *

By

M. Demerec and G. A. Lebedeff

Department of Genetics, Carnegie Institution of Washington
Cold Spring Harbor, N. Y.

Received March 16, 1934

For a comparison of chromosomal homology of various *Drosophila* species, it is important to know the spindle-fibre attachment points of comparable chromosomes. Since there is a simple method for the determination of this point for the X-chromosomes, experiments were planned to determine the location of the spindle-fiber attachment of the chromosomes of *D. virilis*. The results of these experiments are reported here.

Method.—It has been shown by Anderson and Bridges (Anderson 1925; Bridges and Anderson 1925) that if two maternal X-chromosomes heterozygous for various characters are retained in the daughters, then factors which were heterozygous in the parent frequently become homozygous in the offspring (equational exceptions). These authors showed also that the frequency of the equationals increases with the distance of the locus from the spindle-fiber attachment. In order to determine the spindle-fiber end of a chromosome, therefore, it is necessary to determine the frequency of the equational exceptions for the different regions of the chromosome.

In these experiments X-ray radiation was used to increase the frequency of non-disjunctions. Females heterozygous for a number of sex-linked genes were treated with a dosage of 1500 to 2500 r-units and their offspring was examined for female primary non-disjunctions. The genetic constitution of every exceptional female was determined by breeding. Only fertile females, therefore, are considered here.

The following sex-linked loci will be mentioned in connection with this experiment: scute (*sc*-3.0), crossveinless (*c*-23.5), miniature (*mt*-68.5), small-bristles (*sb*-109.7), apricot (*ap*-114.7), ragged (*rg*-132.0) and

* The preliminary work on this problem has been aided by a grant from the National Research Council, Committee on the Effects of Radiation upon Living Organisms.

bobbed (*bb*-146.0). Characters apricot and bodded were received from Professor T. Komai of Kyoto Imperial University.

Results.—A summary of the results pertaining to the homozygosis of both dominant and recessive alleles of the various loci is given in table 1. The detailed data on crossing-over in the non-disjunctional females are reserved for another study in which the question of crossing-over in the X-chromosome will be discussed.

Table 1. Frequency of homozygosis among primary non-disjunctional females (h=homozygous; t=total).

Constitution of X-rayed females	Non-disjunctions															
	<i>sc</i>		<i>c-v</i>		<i>mt</i>		<i>s</i>		<i>sb</i>		<i>ap</i>		<i>rg</i>		<i>bb</i>	
	h	t	h	t	h	t	h	t	h	t	h	t	h	t	h	t
<i>sc c v/c mt sb</i>	1	7	2	7	5	7	—	—	2	7	—	—	—	—	—	—
<i>sc c v/c mt sb ap rg</i>	2	14	5	14	4	14	—	—	4	14	3	14	2	14	—	—
<i>c v mt sb rg/sc v ap</i>	5	15	4	15	6	15	—	—	5	15	5	15	3	15	—	—
<i>c v mt sb rg/sc v s ap</i>	4	9	3	9	2	9	2	9	2	9	2	9	2	9	—	—
<i>ap/ap rg bb</i>	—	—	—	—	—	—	—	—	—	—	—	—	4	24	1	24
Total	12	45	14	45	17	45	2	9	13	45	10	38	11	62	1	24
Per-cent homozygosis	26.7± 4.4		31.1± 4.7		37.8± 4.9		22.2± 9.4		28.9± 4.6		26.3± 4.8		17.7± 3.3		4.2± 2.8	

Discussion.—As shown by work of Bridges and Anderson, equational non-disjunctions in *D. melanogaster* appear as a result of cross-overs occurring when chromosomes are already split, viz., when they are in a four strand stage. The appearance of similar non-disjunctions in *D. virilis* indicates that in this species also crossing-over occurs in the four strand stage.

The data in table 1 show that the frequency of homozygosis for *sb*, *ap*, *rg* and *bb* loci decreased rapidly with the proximity of the locus to the right hand end of the chromosome. This indicates that the location of the spindle-fiber is at the same end. The presence of the locus bobbed is another confirmatory evidence for this assumption since this character is similar to the one located in the same region of the melanogaster chromosome. Both bobbed characters affect the size of bristles and, moreover, in both species the mutant allele is located in the Y-chromosome and the wild-type allele in the X-chromosome.

In figure 1, the data of table 1 are presented graphically by using as ordinate the genetic map of the X-chromosome. The locus *s* has been left out of the figure since the number of observations for that locus is too small to be considered significant. This figure indicates that the homozygosis curve for *virilis* is for about three quarters of its length a horizontal line and that in its last quarter it rapidly approaches zero.

Figure 1. Homozygosis curves for *D. virilis* and *D. melanogaster* (Anderson 1931 data). Map distances for melanogaster loci are entered on the virilis map using locus bobbed (146.0) as the starting point.

If non-disjunction occurred only at the reductional division, then the spindle-fiber attachment points would never remain together in the same gamete. Homozygosis of a locus in that case would be brought about by crossing over occurring between this locus and the spindle-fiber attachment end, and the incidence of homozygosis for loci close to the spindle-fiber would be equivalent to their map distance from the spindle-fiber point. For loci distant from the spindle-fiber point, the frequency would be lowered by multiple crossovers until the value for random assortment is reached, which is 33.3 per cent. Non-disjunctions occurring at the equational division would produce an effect just opposite from that produced by non-disjunctions occurring at the reductional divisions, viz., they would cause complete homozygosis of spindle-fiber point and decrease it for loci toward the left hand end of the chromosome.

Homozygosis curve indicates that in *virilis* few of the non-disjunctions occur at the equational division. The curve reaches its zezo point near the bobbed locus and the random assortment level about 40 units from bobbed. One homozygous bobbed female found among 24 tested may be due either to a crossover between bobbed and the spindle-fiber attachment or it may be due to an equational non-disjunction. The

results of the experiments with melanogaster (Bridges and Anderson 1925; Rhoades 1931) indicate that crossing-over does not occur in that species between bobbed and the spindle-fiber. If the same is true for virilis then the homozygous bobbed female observed in our experiment was produced by a non-disjunction occurring at the equational division which in turn would indicate that, in our case, about four per cent of non-disjunctions occurred at that division.

Figure 1 gives also the homozygosis curve for melanogaster prepared from data of Anderson (1925). In preparing this curve bobbed locus was used as the starting point and the melanogaster map distances from bobbed were entered on the virilis map scale of ordinate. The virilis map is about twice as long as the melanogaster map. Except for the lower level of the melanogaster curve the two curves coincide well. The beginning of the slope comes in both of them at about the same point. As pointed out by Anderson, the high frequency of non-disjunctions occurring at equational division is probably responsible for the lower level of the melanogaster curve. Since the physical lengths of melanogaster and virilis X-chromosomes are approximately the same (Metz 1918) this indicates that the shape of the homozygosis curve is entirely dependent on the genetic map, viz., on the distances of the loci from the spindle-fiber attachment.

Summary.—Virgin females heterozygous for several sex-linked genes were subjected to X-ray treatment to increase the occurrence of primary non-disjunctional females. The percentage of homozygosis among these females for loci tested was as follows: *sc* (3.0) 26.7, *c* (23.5) 31.1, *mt* (68.5) 37.8, *s* (90.5) 22.2, *sb* (109.7) 28.9, *ap* (114.7) 26.3, *rg* (132.0) 17.7, *bb* (146) 4.2. The decrease of homozygosis toward the bobbed end of the chromosome indicates that the spindle-fiber attachment is located at that end.

Literature cited

- Anderson, E. G., 1925 Crossing over in a case of attached X-chromosomes in *Drosophila melanogaster*. *Genetics* 10: 403-417.
 ———. 1931 The constitution of primary exceptions obtained after X-ray treatment of *Drosophila*. *Genetics* 16: 386-396.
 Bridges, C. B., Anderson, E. G., 1925 Crossing over in the X-chromosome of triploid females of *Drosophila melanogaster*. *Genetics* 10: 418-441.
 Metz, C. W., 1918 The linkage of eight sex-linked characters in *Drosophila virilis*. *Genetics* 3: 107-134.
 Rhoades, M. M., 1931 The frequencies of homozygosis of factors in the attached-X females of *Drosophila melanogaster*. *Genetics* 16: 375-385.