MEASUREMENT OF THE RATIO OF THE NUMBER OF X CHROMOSOMES TO SETS OF AUTOSOMES IN *DROSOPHILA MELANOGASTER*

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ABSTRACT

In *Drosophila melanogaster* both sex determination and dosage compensation are governed by the ratio of the number of X chromosomes to sets of autosomes. We propose a mechanism by which this ratio can be measured at the cellular level. The mechanism helps in understanding the effects of, and interactions among, mutants affecting the processes of sex determination and dosage compensation.

IN the fruitfly, *Drosophila melanogaster*, the rate of transcription of the X chromosome in the male is about twice that of each of the two X chromosomes in the female. As a result, the amount of gene pro-

duced per cell in a male, which normally has one X chromosome, and that per cell in a female, which has two X chromosomes, is nearly the same. This method of compensating for the difference between the two sexes in the number of X chromosomes has been called *dosage compensation*[1-3].

It has been known for a number of years that sex in *Drosophila* is determined by the ratio of the number of X chromosomes (X) to sets of autosomes (A)[4].

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It has recently become recognised that the mechanism of dosage compensation in *Drosophila* also involves the measurement of the X/A ratio[2]. We outline here a mechanism which explains how the X/A ratio can be measured at the intracellular level. Since both sex determination and dosage compensation require the monitoring of the X/A ratio, the proposed mechanism has a direct bearing on these two general problems.

Mutants affecting either sex determination or dosage compensation would be expected to yield important information on mechanisms underlying the measurement of X/A ratios. We have therefore examined the properties of such mutants, especially those described by Cline[5] who has studied the characteristics of, and interactions among, a set of remarkable sex-specific lethals. The essential features of these mutations are as follows: (i) Daughterless (*da*) is an autosomal recessive. Only male progeny of *da/da* females survive. Female embryos of *da/da* mothers can also be made to develop normally by injection of cytoplasm from wild-type eggs[6], lending support to the view that the *da+* gene codes for a diffusible factor is essential for female development. (ii) Sex lethal, female specific (Sxl^{F1}) is on the X chromosome. It is normally recessive, and lethal in females but not in males. (iii) Sex lethal, male specific (Sxl^{M1}) is very closely linked to Sxl^{F1}. It is lethal to males, leaves females unaffected and, surprisingly, is a dominant suppressor of daughterless. Cline[6] has shown that the phenotypes of all these mutations can be accounted for on the following assumptions: Sxl^{F1} is a structural gene which synthesizes the Sxl^+ product; Sxl^{M1} is the control region which regulates the rate of transcription of the Sxl^{F1} gene; the *da+* locus produces a factor in the mother which somehow measures the X/A ratio in the fertilized egg and appropriately stimulates the Sxl gene to turn it *on* if the ratio corresponds to that of a female and to turn it *off* if the ratio corresponds to that of a male; Sxl^+ product is essential for females and lethal for males. Because Sxl appears to be a dosage-sensitive locus, Cline has speculated that Sxl^+ product might itself be involved in dosage compensation[6] and sex determination[7]. The manner in which the *da+* factor measures the X/A ratio is, however, *not* specified by Cline.

A simple way by which the *da+* factor could measure the X/A ratio and signal this information to the X-linked Sxl locus is by directing the synthesis of limiting concentrations of an autosomal repressor. If each Sxl locus (comprising both the Sxl^{M1} and Sxl^{F1} regions) competes for this autosomal repressor, the amount of repressor bound per Sxl locus would depend on the X/A ratio. This is because the amount of repressor available would be proportional to the number of sets of autosomes while the number of Sxl loci would be

proportional to the number of X chromosomes. However it is not sufficient if the rate of transcription is proportional to the amount of repressor bound per Sxl locus. It turns out that the mutants described by Cline are best explained when the level of Sxl^+ product first increases and then decreases as a function of repressor concentration. Our reasoning is as follows: (i) Daughters of *da/da* mothers are inviable because they would have little or no repressor and synthesize little or no Sxl^+ product. (ii) Wild type females would have moderate quantities of repressor bound per Sxl locus because they have two sets of autosomes synthesizing repressor and two Sxl loci per cell to bind it. Males on the other hand would have higher levels of repressor bound per Sxl locus because, while they also have two sets of autosomes synthesizing repressor, they have only one Sxl locus to bind it. (iii) Since the Sxl^+ product is assumed to be essential for females and lethal for males[5], females would be expected to have higher levels of Sxl^+ product than males. As a result of these three conditions the level of Sxl^+ product sythesized as a function of the amount of repressor bound per Sxl locus should first increase and then decrease as shown in Fig. 1.

In a separate publication we propose a molecular mechanism which yields such a bell-shaped curve and show that the amount of Sxl^+ product synthesized under such conditions is proportional to the X/A ratio over a wide range of chromosome constitutions. If the Sxl^+ product is an inhibitor of X chromosome activity (as postulated by Cline[6]), this proportionality between the amount of Sxl^+ product and the X/A ratio results in an increase in the activity of the single X chromosome in the male relative to that of either of the two X chromosomes in the female: in other words, dosage compensation occurs.

It follows from Fig. 1 that males, which require low levels of Sxl^+ product, will be viable at the two ends of the bell-shaped curve; that is, at both very low and very high levels of repressor concentration. Females on the other hand need high levels of Sxl^+ product and will therefore be viable only at intermediate levels of repressor concentration. One consequence of this is that a partial reduction in repressor concentration should lead to increased levels of Sxl^+ product (see Fig. 1) and therefore decreased viability of males. It should also lead to a decreased rate of X-chromosome transcription in the male. An autosomal mutation *mle* has precisely these properties[8].

We postulate that increasing levels of Sxl^+ product induce a female phenotype whereas decreasing concentrations induce a male phenotype. Departures from the wild type values in the level of Sxl^+ product would be expected to affect both viability and the sexual phenotype of an individual. When a mutation affects the level of Sxl^+ product, but does not signi-
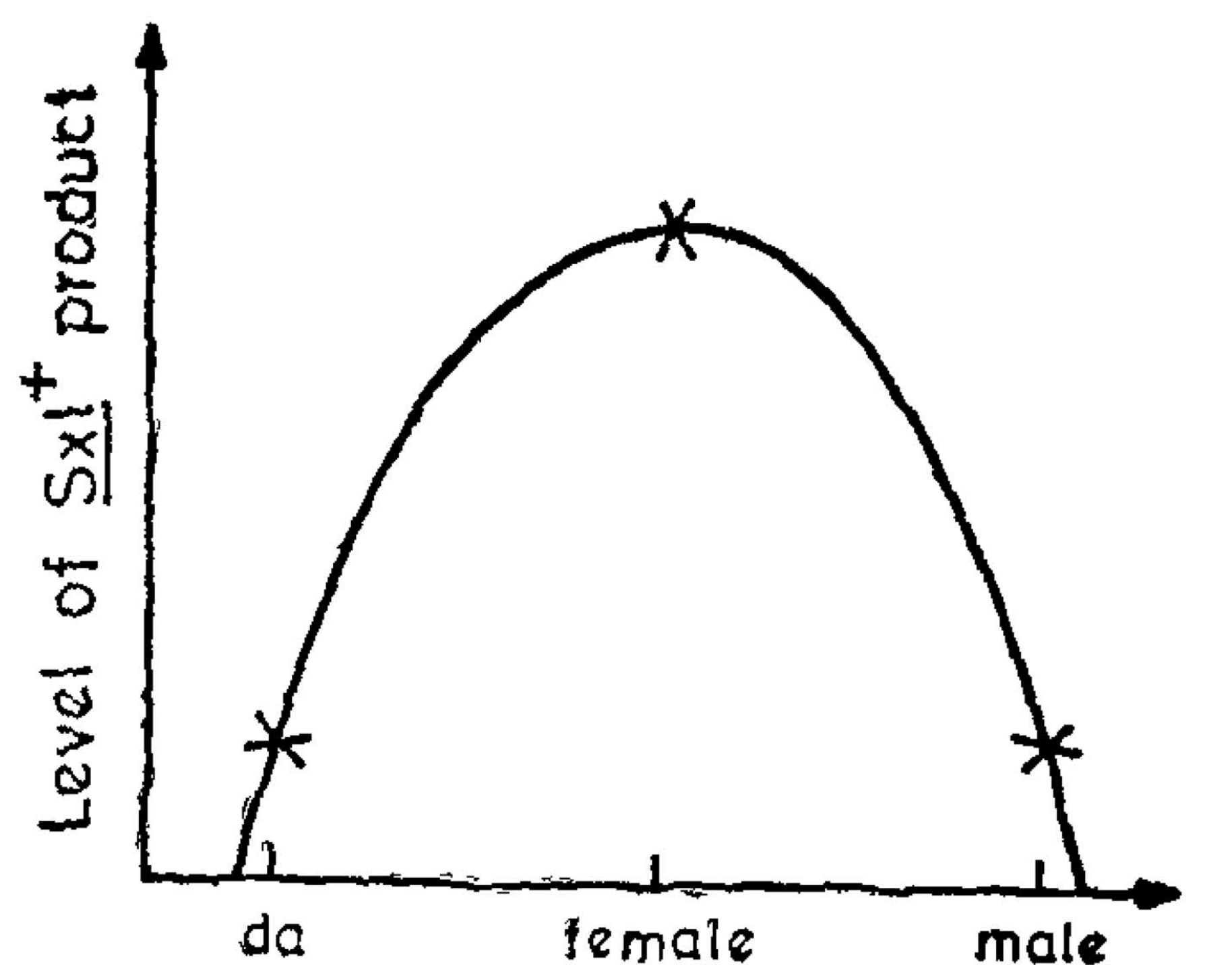
Fig. 1. Synthesis of Sxl^+ product expressed as a function of the amount of repressor bound per Sxl locus. *da* on the abscissa refers to daughters of *da/da* mothers.

ficantly affect viability, it is possible to predict its effects on the sexual phenotype. For example, a single copy of Sxl^{F_1}, a mutation in the structural region, would reduce the level of Sxl^+ product in a female to about half the normal level. The mutation *mle/mle*, which is expected to lead to a partial reduction in repressor concentration, would also lower the level of Sxl^+ product. If a female of the genotype $Sxl^{F_1}/$ Sxl^+; *mle/mle* survives, it would obviously be expected to have rather low levels of Sxl^+ product and therefore show male-like characters. Such flies have recently

been examined by Skripsky and Lucchesi[9] and the females were indeed found to develop sex combs, a male secondary sexual character.

A detailed description of the model, including the quantitative results and predictions about the effects of various mutations, is being published separately.

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