Genet. Res., Camb. (1968), **11**, *pp.* 115–118
Printed in Great Britain

Dosage compensation as a developmental phenomenon in *Drosophila*

By GEOFFREY L. LEE
University of California at Davis, California, U.S.A.

(*Received* 18 *August* 1967)

There are three theories of dosage compensation in *Drosophila*. These are:

(1) The *X*-activation theory. This theory has developed as a parallel to the inactive *X* hypothesis which is considered to explain dosage compensation in mammals (see Lyon, 1961, 1962). Briefly the idea is that the single male *X* in *Drosophila* is much more active than either one of the two female *X*'s. Cytological observation by Offermann (1936) and Dobzhansky (1957), among others, support this theory.

(2) The second theory is due mainly to Muller (1950). It is based on the idea that each sex-linked gene has a set of dosage modifiers (called compensators and anti-compensators by Muller) which are also sex linked and these act to equalize male and female expression for a particular gene. Some genes are thought to have more than one set of compensators although the compensator genes themselves are not excluded from having other effects (Muller & Kaplan, 1966).

(3) A third theory has been advocated by Goldschmidt (1955). He maintains that the sex-determining genes themselves may act as dosage modifiers. These genes in determining sex also set the developmental pattern for each individual and this in turn affects gene expression. Muller anticipated this alternative explanation but was unable to adequately challenge it. His objection (Muller, 1950) that transformed females (i.e. females homozygous for the gene *transformer* that causes them to be phenotypically similar to males) show female dosage effects is not tenable because such individuals are really extreme female intersexes and developmentally resemble females more than males (Goldschmidt, 1955).

Muller & Kaplan (1966) have critically examined the first two of these theories. They conclude that, although the male *X* chromosome does differ cytologically from a single female *X*, this difference does not seem large enough to account for dosage compensation. Furthermore their experiments show no evidence for a centre of activation which would be analogous to the inactivation centre located on the *X* chromosome of the mouse. It is worth noting, however, that whole chromosome control has been demonstrated in other insects, notably Sciara, while in the water strider *Gerris lateralis*, Geitler (1937) has demonstrated that the *X* chromosomes form chromocentres in interphase somatic cells. Frizzi (1948) working with *Bombyx mori* and Smith (1945) with the spruce bud worm (*Archips fumifera*) have demonstrated a similar phenomenon. In addition recent evidence from several mammals indicates that *X* inactivation may not be complete and so in Drosophila we are not yet in a position to reject partial *X* activation.

In the second portion of their paper Muller & Kaplan discuss the theory that sex-linked modifying genes are responsible for dosage compensation.

The evidence for this theory comes from experiments in which small fragments of the *X* chromosome were added to a *Drosophila* genome and their effect on various characters noted. Some characters showed an increase in expression while others showed a decrease. The validity of the conclusions reached from these experiments is at best dubious. The genome is a highly integrated unit in which the chromosomes themselves tend to act as

functional units (see Linsley, 1964; Brosseau, Nicoetti, Grell & Linsley, 1961). The inserted piece of chromosome may disturb the normal activity of the genome in a fashion completely unrelated to its normal function as a part of a complete chromosome. Furthermore, in order to demonstrate the existence of compensators it would be necessary to locate genes whose sole effect would be the sex-limited enhancement or suppression of a specific sex-linked gene. Such genes have not been found, even in selection experiments whose design specifically favoured their detection; for example, the experiment of Harrison (1953), Merrell & Underhill (1956) and our own experiment described below.

Muller and Kaplan do not discuss Goldschmidt's theory that dosage compensation has its basis in the different developmental rates. In *Drosophila* there are a number of autosomal mutants that show sex dimorphism and also the sexes often are observed to respond differentially to selection and to temperature stress. Such differences are not included in Muller's theory but are both predicted and explained by Goldschmidt's hypothesis.

Table 1

		Emergence time (days)					Total no. of flies scored
		14	15	16	18	20	
Cumulative % of females	Selected line	48·9	50·2	49·6	49·1	48·4*	9583
Average over 50 single pair families	Control line	56·0*	53·4*	52·6*	50·6	50·2	1775

* χ^2 shows a significant deviation from a 1:1 sex ratio at the 5% level.

Our results support Goldschmidt's hypothesis. If the developmental rate of *Drosophila* is disturbed by subjecting them to high temperature (30° C) throughout life, the gene *white honey* which at normal temperatures shows full dosage compensation, becomes sexually dimorphic (Lee, unpublished). This would seem to link dosage compensation with developmental rate.

More extensive evidence comes in *Drosophila* from a selection experiment for complete dosage compensation of scutellar bristle number in the presence of the gene *scute* using family selection. The difference between male and female bristle number has been decreased from 0·9 of a bristle to 0·3 of a bristle. An investigation of the selected population at generation 17 showed that the relative developmental rates of males and females had been noticeably altered (see Table 1). Males in the selected line tended to develop faster than females and the overall sex ratio had been reversed.

Table 1 shows that in the control line there was a significant excess of females for the first 3 days of emergence after which the sex ratio approximated 1:1. On the other hand the selected line showed a non-significant excess of males for the first 5 days of emergence and by the seventh day there was a significant excess of males. This indicates that selection for complete dosage compensation of the scute gene has speeded up the developmental rate of males with respect to females, suggesting that a close relationship exists between developmental rate and dosage compensation.

In addition to the developmental changes a consistent but low proportion of two phenocopies were found in the selection line. The first of these resembled the mutant *vestigial* with a large proportion of flies having a weak vestigial phenotype and a smaller proportion having the severe *vestigial-hemithorax* phenotype. The significance of this is that this mutant is known to be associated with a disturbance of the developmental rate (see Bridges & Brehme, 1944; Harnly & Harnly, 1935). The second phenodeviant is one which we call '*sexless*'. It is characterized by an absence of external genitalia in the male.

Kroeger (1960) describes a similar phenotype in a *facet-notch* stock of *Drosophila* and interprets the phenomenon as being due to a disturbance of development induced by the mutant *facet-notch*. Our production of this phenotype in the absence of the *facet-notch* mutant suggests that we have created a similar developmental disturbance.

Both phenodeviants usually occurred as 1–2% of the offspring of any single-pair family. Both were first seen at about the thirteenth generation of selection and despite considerable fluctuation both tended to increase in frequency, reaching a maximum at the seventeenth generation, where '*sexless*' was observed in 10% of all families and *vestigial hemithorax* in 50%. The occurrence of phenodeviants in the selection line is probably due to the type of selection applied because the same stock carrying the scute gene has been subjected to mass selection for both high and low scutellar bristle number as well as having been extensively inbred in the course of other experiments (Fraser, 1966) and none of these phenomena were observed.

Studies in *Habrobracon* by Clark & Mitchell (1951) and Van Pelt (1966) also support Goldschmidt's theory of dosage compensation in insects. *Habrobracon* is particularly amendable to such studies because it is possible to have both haploid and diploid males as well as triploid females.

It is feasible to suggest that in mammals where the developmental rate of males and females is virtually identical, evolution has favoured a special method of dosage compensation, whereas in insects such a system would be superfluous because males and females represent different genetic systems each with its own pattern of gene action. Such a mechanism allows us to explain both dosage compensation and sex dimorphism without postulating a complex of compensator and anti-compensator genes.

SUMMARY

The three theories of dosage compensation in *Drosophila* are examined. Data are presented supporting a developmental interpretation. The reason why such a mechanism is applicable in insects but not in mammals is discussed.

This work was financed by NIH Grant no. GM 11778–02 to Dr A. S. Fraser.

Thanks are due to Dr Fraser for his invaluable help and guidance during the course of this work.

REFERENCES

Braun, W. (1942). The effect of changes in time of development on the phenotype of mutants of *Drosophila melanogaster*. *Univ. Calif. Publs Zool.* **49**, 61–84.

Bridges, C. B. & Brehme, K. S. (1944). The mutants of *Drosophila melanogaster*. *Publs Carnegie Instn*, no. 552.

Brosseau, G. E., Nicoetti, B., Grell, E. H., & Linsley, D. L. (1961). Production of altered *Y* chromosomes bearing specific sections of the *X* chromosome in Drosophila. *Genetics* **46**, 339–346.

Clarke, A. M. & Mitchell, C. J. (1951). Gene dosage relationships for haploids and diploids of *Habrobracon*. *Genetics* **36**, 185–198.

Dobzhansky, T. L. (1957). The *X* chromosome in the larval salivary glands of hybrids *Drosophila insularis* × *Drosophila tropicalis*. *Chromosoma* **8**, 691–698.

Fraser, A. S. (1966). Variation of scutellar bristles in Drosophila. XII. Selection in scute lines. *A.J.B.S.* **19**, 147–154.

Frizzi, G. (1948). L'eteropicnosi come indice di riconoscimento dei sessi in *Bombyx mori* L. *Ric. Sci.* **18**, 1–7.

Geitler, L. (1937). Die Analyse des Kernbaus und der Kernteilung der Wasserläufer *Gerris lateralis* und *Gerris lacustria* (Hemiptera, Heteroptera) und die Somadifferenzierung. *Z. Zellforsch mikrosk. Anat.* **26**, 641.

GOLDSCHMIDT, R. B. (1955). *Theoretical Genetics*, pp. 356–358. Berkeley: University of California Press.

HARNLY, M. H. & HARNLY, M. L. (1935). The effect of temperature on the wings and eyes of the dimorphos vestigial combinations in *D. melanogaster*. *J. exp. Zool.* **72**, 75–99.

HARRISON, B. J. (1953). Reversal of a secondary sex character by selection. *Heredity* **7**, 153–164.

KROEGER, H. (1960). Hypo- and hyperdevelopment of the male genital apparatus and the Bd-M combination in *Drosophila melanogaster*. *J. Morph.* **107**, 227–232.

LINSLEY, D. L. (1964). Chromosomal function at the supragenic level. *Natn. Cancer Inst. Monogr.* **18**, 275–290.

LYON, M. F. (1961). Gene action in the *X* chromosome of the mouse (*Mus musculus* L.). *Nature, Lond.* **190**, 372–373.

LYON, M. (1962). Sex chromatin and gene action in the mammalian *X* chromosome. *Am. J. Human Genet.* **14**, 135–148.

MERRELL, D. J. & UNDERHILL, J. C. (1956). Competition between mutants in experimental populations of *Drosophila melanogaster*. *Genetics* **41**, 469–477.

MULLER, H. J. (1950). Evidence for the precision of genetic adaptation. In *The Harvey Lect.* ser. XLIII, 1947–48, pp. 165–299. Springfield, Ill.: Charles C. Thomas.

MULLER, H. J. & KAPLAN, W. D. (1966). The dosage compensation of *Drosophila* and mammals as showing the accuracy of the normal type. *Genet. Res.* **8**, 41–59.

OFFERMANN, C. A. (1936). Branched chromosomes as symmetrical duplications. *J. Genet.* **32**, 103–116.

SMITH, S. G. (1945). Heteropycnosis as a means of diagnosing sex. *J. Hered.* **37**, 195–199.

VAN PELT, G. S. (1966). The problem of gene dosage in *Habrobracon*. (Abstract only.) *Genetics* **54**, 367.