LETTERS TO THE EDITORS

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Genetic Effects of X-Rays in Relation to Dose-rate in *Drosophila*

VARIATION in dose-rate has generally been observed to be ineffective in determining the incidence of mutations and structural changes produced by X-rays in *Drosophila* spermatozoa[1]. However, since it is now clear that a considerable fraction of the genetic damage brought about by radiation results from an indirect path of action, intensity effects might be expected to operate under certain conditions. Chemical mutagens induced by radiation must have a finite life-span before they are removed or rendered ineffective by one or another metabolic pathway. Therefore very high dose-rates, leading to an accumulation of mutagens, should effect a greater amount of damage as a result of diffusion of the active substances to other regions within the nucleus. Sobels's observations[2] that pretreatment with cyanide or azide increases the yield of sex-linked lethals obtained from irradiated *Drosophila* males suggests that the accumulation of chemical mutagens can be facilitated by the concurrent use of suitable enzyme inhibitors.

Haas *et al.*[3] have indeed reported recently that they observed an intensity effect while studying the production of translocations in *Drosophila* sperm. Using dose-rates of 100 r./min. and 2,000 r./min., we have been able to confirm that, for a given dose, a greater amount of genetic damage is produced if the radiation is delivered at high intensity. With a total dose of 2,000 r., the high dose-rate gives an increase of up to 25 per cent in the yield of recessive sex-linked lethals and of translocations. If, just prior to irradiation, the flies are injected with 0·005 *M* sodium azide in saline, the intensity effect is enhanced. The figures in Table 1 illustrate the effects of such an injection on the production of sex-linked recessive lethals by 2,000 r. of X-irradiation delivered to spermatozoa within the spermathecæ of inseminated females.

An especially interesting feature of high-intensity treatment is the production of numerous F_1 females that prove to be mosaics for a sex-linked lethal. By subculturing a single female from those F_2 cultures which fail to yield a lethal, a further crop of mutations may be obtained in the F_3 generation at a frequency of about one-tenth that observed in the preceding generation. This delayed appearance of lethals recalls the similar effects produced by chemical mutagens such as mustard gas[4]. Delayed mutations may also be obtained by mating males that have received high-intensity radiation to attached-*X* females and then crossing the F_1 males to Muller-5 females in the usual way. It should be emphasized that such delayed effects are quite rare if the same dose of radiation is administered at an intensity less than 1,000 r./min. Further study is needed before it can be decided whether the lethals recovered in the F_3 generation are really instances of delayed mutation or whether they are the result of partial breakage of a multi-strand chromosome, followed by sorting out of a mixture of broken and unbroken chromonemata.

Table 1. SEX-LINKED LETHAL MUTATION-RATE

Dose-rate	Inseminated females received 2,000 r. preceded by an injection of 0·2 μl. of:	
	0·7 per cent NaCl	0·005 *M* NaN_3 in 0·7 per cent NaCl
100 r./min.	6·3% ± 0·8	7·6% ± 1·0
2,000 r./min.	8·4% ± 0·6	13·1% ± 1·2

Table 2. PRODUCTION OF HYPERPLOID FEMALES

Treatment	Total No. females	Hyperploids	Standard error		P
A	1,875	1·07%	0·24		
B	1,718	1·16%	0·26	= 0·3	
C	1,772	1·52%	0·29		< 0·01
D	2,316	0·52%	0·15		

A, undivided dose; *B*, four equal fractions, 1 min. between each; *C*, four equal fractions, 5 min. between each; *D*, four equal fractions, with a 30-min. interval between each. Temperature, 16° C. Total dose, 4,000 r.

In an attempt to estimate the duration of life of the active substances produced by the high-intensity irradiation, fractionation experiments were carried out with varying time-intervals between successive treatments. The experiments were scored by recording the proportion of hyperploid females obtained from a single 24-hr. mating of attached-*X* females (homozygous for the recessive gene, yellow) to Canton *S* males that had received a total dose of 4,000 r. delivered at high intensity (Table 2). At a temperature of 16° C., division of the dose into four equal fractions does not reduce the incidence of hyperploids obtained unless the time-interval between successive doses of radiation is more than five minutes. The critical time-interval would, of course, be expected to be related to the magnitude of each fractional dose, but clearly the effects may persist long enough for further experimental analysis to be practicable.

Full details of this work will be published elsewhere.

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[2] Sobels, F. H., *Z. indukt. Abstammung u. Vererbl.*, **86**, 399 (1955).
[3] Haas, F. L., Dudgeon, Edna, Clayton, F. E., and Stone, Wilson S., *Genetics*, **39**, 453 (1954).
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A Ventral Ectodermal Ridge of the Tail in Mouse Embryos

THE limb buds of mammalian and avian embryos show a thickening of the ectoderm on the margin of the foot plates, the so-called apical ectodermal ridge, and it is known that this ridge plays an important part in the growth and differentiation of the limbs[1,2]. It has recently been discovered that a similar thickening of the ectoderm is present on the ventral side of the tail in 10- and 11-day old mouse embryos. In 10-day old embryos (crown-rump length 3·4–4·2 mm.; lens invaginations still widely open), there occurs a very conspicuous zone of columnar epithelium on the ventral aspect which extends from the very tip of the tail in a proximal direction for roughly 250 micra; the exact length of the zone is difficult to determine as it gradually disappears