Khan, A. H. and T. Alderson. University of Cambridge, England. An attempt to sensitize Drosophila chromosomes to X-irradiation after 5-bromodeoxyuridine "incorporation" into DNA.

Substitution of 5-bromodeoxyuridine (BdU) for thymine in DNA has been shown to enhance the sensitivity of mammalian cells and micro-organisms to X-irradiation.

Using an aseptic and chemically-defined culture medium, the radiosensitivity (for sex-linked recessive lethal mutations) of Drosophila larval spermatogonia has been compared in the presence and in the absence of BdU. (Drosophila does not utilize pyrimidine bases unless they are supplied as the nucleoside). In order to increase the chance of BdU incorporation, larvae were cultured in the presence of the folic acid analogue, aminopterin, which inhibits thymine synthesis; this procedure limits the culturing time to 48 hours, since longer periods impose a folic acid deficiency (by aminopterin) which is not corrected in time for emergence of adult flies. (Folic acid is required for adult emergence).

Oregon-K eggs were collected and sterilized by Sang's method, and spread evenly over a sterile 3 percent agar surface. On emergence the larvae were transferred under aseptic conditions to the media (minus folic acid and RNA) at a density of 100 larvae/25 ml of medium for 48 hours, transferred onto sterile 3 percent agar for X-irradiation, and then onto a normal chemically-defined medium (plus 0.4% RNA) supplemented with additional folic acid (0.02 percent) until emergence of the adult flies. Irradiation was by a Maximar General Electric Machine at a dose of 810 r (delivered at 180 r per minute). Virgin males and females were examined for sex-linked recessive lethals by the Muller-5 (basc) method using a single brood by mating individual males to two virgin females for 3 days, and, in the case of females, by mating individual females to two males for 3 days.

The types of treatment for both male and female larvae are listed in Tables 1 and 2, where larvae are cultured in the presence of either dU (deoxyuridine) or BdU for 48 hours (with and without aminopterin), and followed by irradiation, or without irradiation. None of the sex-linked recessive lethal frequencies differ significantly from one another; there is no evidence for BdU-induced mutagenesis; and only a trivial increase in mutation following X-irradiation of males. There is no evidence for an increase in mutational radiosensitization of the X-chromosome in the presence of BdU, nor is there evidence for BdU incorporation, although the BdU + aminopterin cultures do slow down larval development compared with the dU + aminopterin cultures.

Table 1.--Sex-linked recessive lethal frequencies in Drosophila males induced by X-irradiation (810 r) after larval feeding treatments in the presence of 5-bromodeoxyuridine (BdU), or deoxyuridine (dU).

	dU	BdU	BdU + X-irradiation	dU + X-irradiation	dU + X-irradiation	BdU + X-irradiation
Concentration of dU and BdU (%)	0.02	0.02	0.02	0.02	0.02	0.02
Concentration of Aminopterin (%)	-	-	0.002	-	0.002	0.002
Hatchability (%)	78.3	73.8	51.1	65.5	56.3	61.6
No. males examined	74	99	47	94	45	90
Average no. chromosomes examined/male	7.8	6.4	7.9	7.3	8.2	9.5
No. chromosomes examined	5577	636	371	692	371	855

Table 1.--continued.

	dU	BdU	BdU + X-irradiation	dU + X-irradiation	dU + X-irradiation	BdU + X-irradiation
No. lethal chromosomes	1	2	1	4	2	4
Lethals (%)	0.17	0.31	0.26	0.57	0.53	0.46

Table 2.--Sex-linked recessive frequencies in Drosophila females induced by X-irradiation (810 r) after larval feeding treatments in the presence of 5-bromodeoxyuridine (BdU) or deoxyuridine (dU).

Treatment	dU	BdU	BdU + X-irradiation	dU + X-irradiation	dU + X-irradiation	BdU + X-irradiation
Concentration of dU and BdU (%)	0.02	0.02	0.02	0.02	0.02	0.02
Concentration of Aminopterin (%)	-	-	0.002	-	0.002	0.002
Hatchability (%)	78.3	73.8	51.1	65.5	56.3	61.6
No. females examined	40	47	55	55	50	45
Average no. chromosomes examined/female	6.8	6.4	4	5.8	7.3	8.6
No. chromosomes examined	273	301	220	320	365	389
No. lethal chromosomes	1	1	0	0	0	1
Lethals (%)	0.36	0.33	0.0	0.0	0.0	0.25

Oster, I. I., J. Duffy and R. Binnard.
The Institute for Cancer Research. Observations on a piece of tail.

During the course of counting the number of spermatozoa utilized in successive matings by males of Drosophila melanogaster in connection with experiments on radiation sensitivity, we found that two structural elements could be recognized in the spermatozoon's tail following fixation. Hitherto, observations by others (Cooper, K. W., 1950, Biology of Drosophila; Yanders, A. F., and J. P. Perras, 1960, DIS; Kaplan, W. D., et al., 1962, DIS; Lefevre, G. Jr. and U. B. Jonsson, 1962, Genetics) had revealed that Drosophila has the type of sperm usually described for insects--that is, a filiform head, no separately discernible mid-piece, and a tail. In fruit flies, the only unique feature which had been noticed until now was the unusual length of the tail (0.2 mm to 6.6 mm, depending on the species, although the diameter is of the order of 0.2μ). Our experiments involved the removal with watch-maker's forceps of the vagina and uterus from a female immediately after copulation to a slide containing a drop of Drosophila Ringer's solution, teasing