whenever enzyme-treated DNA is present; and (3) an additional ability of mutant cells to attain a positive selective value as soon as they are present in sufficient proportions, an effect which, as indicated by the "initial mixture" and "transfer" experiments, may be quite independent of the presence of the DNA "derivative." The nature of the inhibitory factor(s) produced by *S* cells in the presence of DNA + DNase is unknown, but may involve a metabolite since the effectiveness of the DNA + DNase effect depends not only upon the strain but also upon the type of medium employed. Finally, the ability of DNase alone to enhance population changes slightly (table 1) may indicate a normal accumulation of DNA in the culture medium which is subsequently converted into "active" DNA by the presence of DNase.

It remains to be determined whether the described non-specific effects of a DNA-like substance upon bacterial population changes might play a secondary role in the more specific phenomenon of bacterial transformations caused by highly polymerized DNA. A more comprehensive report of these studies will be published elsewhere.

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*THE PRODUCTION OF MUTATIONS IN DROSOPHILA MELANOGASTER BY THE FAST NEUTRON RADIATION OF AN ATOMIC EXPLOSION**

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Recently the United States Atomic Energy Commission gave us the unusual opportunity of making a small scale study of the effects of the high intensity fast neutrons produced by an atomic explosion on the mutation rate in the mature sperm of *Drosophila melanogaster*. The males were shielded during the explosion so as to avoid temperature extremes, blast, and nearly all of the other radiation produced by the explosion. Dosimeters were included with the flies, and the estimated dosage received by the flies was calculated and given to us in terms of Roentgens Equivalent Physical (REP).

In the laboratory within a week after exposure the radiated males were mated individually in part to Muller-5 females and in part to *res* females, *res* being a combination of eight recessive third chromosome phenotypic mutations used in a similar test of x-rays by Alexander.[1] The offspring of the Muller-5 cross were used for sex-linked lethal tests by Levine. The offspring of the *res* cross were classed by Ives for four groups of visible mutations: *res*-like (resembling one or another of the eight mutants of that stock), Minute (thin bristles), other bilateral mutants, and unilateral mutants of all kinds. The recovered sex-linked lethals were tested by

Ives and Levine for crossing-over with *y ct*^{6} *ras*^{2}*f*, a group of four sex-linked visibles well spread out in the *X*-chromosome. This showed how many of the lethal chromosomes gave normal crossing-over and how many gave abnormal crossing-over of a type generally associated with a chromosome inversion or translocation. It also proved that in the present instance all lethals derived from a given male were in each case independent mutations.

Altogether this made an unusually comprehensive test of the genetic effects of the radiation received. While the data are not large in comparison with many Drosophila experiments, it seems best to report them in their present form, particularly since it is very unlikely that both intensity and dosage, to say nothing of other conditions, can be even approximately duplicated in another atomic explosion in the near future.

The stock of normal flies used was a strain known as Oregon-R, which had been inbred by single pairs for 100 generations and then kept in mass cultures for about 10 generations before the exposure to atomic radiations. During those 10 generations a similar analysis had been made of soft x-rays by Ives and Yost, using the *res* stock only. In the previous year[2] an earlier derivative of the same inbred line had been tested for sex-linked mutation rate with soft x-rays. The data of the x-ray *res* tests and some of the results of the x-ray sex-linked lethal tests are included

TABLE 1

SEX-LINKED LETHAL MUTATION RATES

DOSAGE	TESTS	LETHALS	PER CENT
315 REP	1453	45	3.1
760 REP	1000	61	6.1
1400 REP	425	48	11.3
2500 r x-rays	2403	165	6.9

here for comparison with the effects of high-intensity fast neutrons. Conditions and criteria for the sex-linked lethal determinations in the present study were similar to those in the earlier study.[2] Radiation in the x-ray study of visibles using *res* was also from the same source as in the previous work (120 KV, 10 ma) at a rate of 200 r per minute.

Table 1 shows the sex-linked lethal mutation rates observed in this study. Included for comparison is the mutation rate of 2500 r soft x-rays found by Young and us and reported earlier.[2] The table shows that 760 REP of high-intensity fast neutrons produced by an atomic bomb are comparable in effect to 2500 r of low intensity soft x-rays. On the basis of this dosimetry the fast neutrons were $3^1/_2$ times as effective as the x-rays.

Table 2 shows the results of analyzing the lethals by means of *y ct*^{6} *ras*^{2}*f*. Most of the lethals of the fast neutron series were successfully analyzed by this method. But only a small random sample of the soft x-ray lethals were similarly analyzed in the earlier study. Also included for comparison is a series of lethals appearing in a study with a naturally occurring mutator and published previously.[3] Any lethal chromosome showing either allelism to one of the marker genes in the F_1 of this test or abnormal crossing-over in both the males and females of the F_2 were classed as complex. The others were classed as simple. Crossing-over was con-

sidered as abnormal when in a count of not less than 100 flies it was reduced to less than half the map distance in one or more regions of the chromosome. Only a few cases of allelism were found and most of them were also associated with reduced crossing-over. No cytological study of radiation lethals has been attempted, but an earlier study[4] showed that the complex lethals in the mutator series were cases of gross chromosomal rearrangement, chiefly inversions.

The data of table 2 show that a large portion of the lethals in the fast neutron series were complex mutations. In practically all cases these showed marked reduction in crossing-over in one or more regions of the chromosome. The proportion did not change significantly from one dosage to another and averaged 42 per cent in the three dosages together. Data on the distribution of these chromosome aberrations in terms of the cross-over regions involved are not included in this report. It may be said, however, that in general the distribution appears to be random. A large portion of them involved the *ras-f* region, as should be the case, since the

TABLE 2

FREQUENCY OF COMPLEX LETHALS

DOSAGE	LETHALS	SIMPLE	COMPLEX	COMPLEX, %
315 REP	39	21	18	46
760 REP	57	35	22	39
1400 REP	43	24	19	44
TOTAL	139	80	59	42.4
2500 r	40	34	6	15.0
Mutator	351	332	19	5.4

TABLE 3

PROPORTIONS OF SIMPLE AND COMPLEX LETHALS

DOSAGE	LETHALS, %	SIMPLE	COMPLEX
315 REP	3.1	1.7	1.4
760 REP	6.1	3.7	2.4
1400 REP	11.3	6.4	4.9
2500 r	6.9	5.8	1.1

large heterochromatic region of the chromosome is entirely beyond *f*. In some cases, however, recovered cross-overs were of normal proportion only in the *ras-f* region, indicating that the neutron bombardment also caused breaks in the euchromatic region of the chromosome.

The fast neutron series had far more complex lethals than appeared in either of the other two series, three times as many as the x-ray series and eight times as many as the mutator series. The number of lethals tested in the x-ray series was not large enough to make the comparison a completely satisfactory one. In both comparisons, however, the chi-square *P* value (with Yates correction) is less than one per cent, indicating a statistically significant difference between the fast neutron series and each of the other two.

Using the lethal rates in table 1 and the percentage of complex lethals in table 2, it is possible to divide the total portion of lethals in each radiated series into two parts, simple lethals and complex lethals as in table 3. The comparison is again a rough one. It suggests that 1400 REP of fast neutrons was approximately equiva-

lent to 2500 r of x-rays in producing simple lethals, while 315 REP equaled 2500 r in producing complex lethals. In the one case, neutrons were twice as effective as x-rays; in the other they were eight times as effective.

TABLE 4

VISIBLE MUTATIONS HIGH-INTENSITY IN FAST NEUTRON SERIES

		DOSAGE IN REP: 315	760	1400	TOTAL
CLASS	TYPE	FLIES: 2250	1445	103	3798
1	*res*-like				
	No.	10	6	0	16
	%	0.44	0.42	0	0.42
2	Minutes				
	No.	11	12	5	28
	%	0.49	0.83	4.9	0.73
3	Other bilat.				
	No.	14	16	4	34
	%	0.62	1.11	3.9	0.89
4	Unilat.				
	No.	15	12	4	31
	%	0.67	0.83	3.9	0.81
	Total				
	No.	50	46	13	109
	%	2.2	3.2	12.6	2.85

TABLE 5

VISIBLE MUTATIONS IN SOFT X-RAY SERIES

		DOSAGE IN r: 3000	5000	7500	10,000	TOTAL
CLASS	TYPE	FLIES: 13,301	5,130	672	393	19,496
1	*res*-like					
	No.	68	32	12	3	115
	%	0.51	0.62	1.79	0.76	0.59
2	Minutes					
	No.	135	87	17	16	255
	%	1.01	1.70	2.53	4.07	1.31
3	Other bilat.					
	No.	62	44	13	11	130
	%	0.47	0.86	1.94	2.80	0.67
4	Unilat.					
	No.	146	78	21	18	263
	%	1.10	1.52	3.13	4.58	1.35
	Total					
	No.	411	241	63	48	763
	%	3.1	4.7	9.4	12.2	3.91

The data on visible mutations with *res* also suggest relatively more chromosomal effect by the fast neutron treatment than by x-rays. These data are presented in tables 4, 5, and 6. Table 4 shows the frequency of the four classes of visibles in the fast neutron series. Table 5 shows the effects of several different dosages of soft x-rays. A comparison of the relative frequencies of the different types of visible mutations in the two kinds of radiated flies is given in table 6. The only class of visible mutations to show a statistically significant difference in relative frequency

is the class called "other bilaterals." They are relatively more frequent in the fast neutron series. This is shown in the summary in table 6 and can be seen by inspec-

TABLE 6

RELATIVE PERCENTAGES OF DIFFERENT CLASSES OF VISIBLE MUTATIONS IN TOTALS OF TABLES 4 AND 5

TREATMENT	MUTANTS	CLASS 1	2	3	4
Neutrons	109	15	26	31	28
X-rays	763	15	33	17	35
	Chi-square	0.00	1.60	9.23	1.11

tion to be true in individual series in tables 4 and 5. This class of mutations consists mostly of dominant visible mutations which resemble well-known dominants found often in radiated material. Studied cytologically many of the previously found radiation-produced dominant visible mutations have proved to be associated

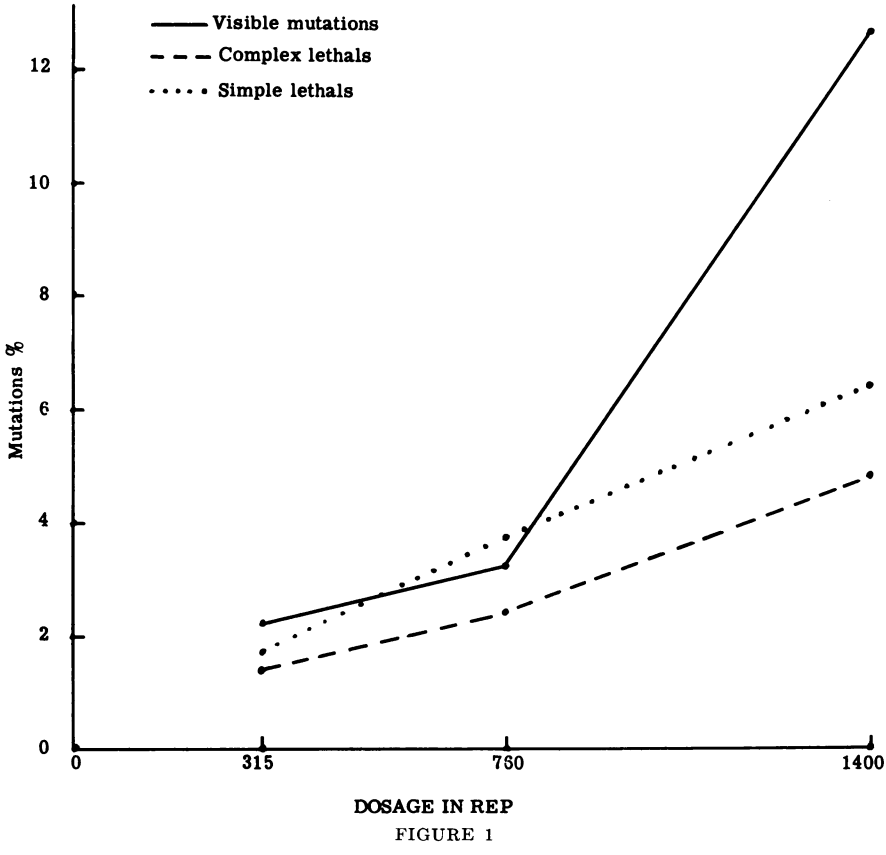

FIGURE 1

Dosage-mutation rate curves.

with easily detectable chromosome rearrangements which frequently give reduced crossing-over. It might be expected, therefore, that if fast neutrons produce relatively more gross chromosomal aberrations, this class of visibles would be increased relative to the other classes as in the present instance.

The data on autosomal visible mutation rates also indicate an over-all great effectiveness of fast neutrons. The total of 3.2 per cent visibles of all classes at 760 REP in table 4 is similar to the total of 3.1 per cent at 3000 r in table 5. The approximately four times greater effectiveness of the fast neutrons in producing autosomal visibles parallels the three and one-half times greater effectiveness in producing sex-linked lethal mutations.

Thus, it is evident that the data from autosomal visible mutations are in close agreement with those from sex-linked lethal mutations, both in the increased total mutation rate and in the increased portion of gross chromosomal aberrations in the fast neutron series.

Finally, it is possible to construct dosage curves for the three major classes of mutations observed in the fast neutron series: the simple lethals, the complex lethals, and the visible mutations of all kinds. This is done in figure 1. The slope of the curves for the two kinds of lethals is similar and the points approximate a straight line. The percentage of visible mutations, however, rose higher than expected in the 1400 REP series on such an interpretation. The data for this dosage are small (13 visibles in 103 test flies) and cannot be said to offer strong evidence against a straight line dosage curve, even though there is a P value of 1 per cent for the difference between 12.6 per cent and the average of the other two mutation rates at that dosage. Nevertheless, they do suggest that the curve may not be linear for visible mutations as a group.

The data on visibles are not large enough to warrant further breakdown into dosage curves for the several kinds of visibles. Further studies in this direction are in progress here involving soft x-rays, the gamma radiation of a 500 Curie Co-60 slug which is being installed at this laboratory, and both thermal and lower intensity fast neutrons now available elsewhere.

The finding of so much gross chromosome damage in lethals from the fast neutron radiation produced by an atomic explosion and the fact that the proportionate amount did not change as the frequency of the lethals rose with increasing dosage suggests that probably a single hit was able to produce two or more chromosome breaks. This appears to be characteristic of fast neutrons generally.[5, 6] It is not the case in the effects of x-rays where a single hit appears to be incapable generally of producing the two breaks necessary for a gross chromosome rearrangement.[7]

While the sex-linked lethal mutation rate in our x-ray series is comparable to that found generally with a similar dosage, the rates in our fast neutron series are comparatively higher than those found by others. Dempster,[5] Giles,[6] and others more recently have reported fast neutrons to be less effective than x-rays in producing sex-linked lethals in Drosophila. It seems probable that the reason for the opposite finding in our case is due to impoved methods now in use for estimating the REP dosage of fast neutrons. It needs to be emphasized therefore that fast neutrons, at least those produced by an atomic explosion, are much more effective than soft x-rays in producing sex-linked lethal mutations in *D. melanogaster*.

Summary.—The fast neutrons of an atomic explosion were three to four times as effective as x-rays in producing sex-linked lethal and autosomal visible mutations in the mature sperm of *D. melanogaster*. About 42 per cent of the sex-linked lethal chromosomes gave evidence of containing gross chromosomal aberrations, eight times as many as appeared in a group of mutator-caused lethals. The increase in

mutation rate with increasing dosage of fast neutrons appeared to be linear for sex-linked lethals, both with and without gross chromosomal aberrations; but the rate may not have been linear in the case of the autosomal visibles.

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*PROTECTIVE PROPERTIES OF CYSTEINE, SODIUM HYPOSULFITE, AND SODIUM CYANIDE AGAINST RADIATION INDUCED CHROMOSOME ABERRATIONS**

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Introduction.—Several investigators have shown the importance of the presence of oxygen to the process of chromosome breakage during exposures to x-rays. By lowering the oxygen tension below 2 per cent or by exposing tissue to x-rays in the presence of other gases such as nitrogen and helium, a considerable decrease in the frequencies of chromosome aberrations was obtained. This was shown by Thoday and Read[1] in the root tips of *Vicia faba*, by Hayden and Smith[2] in barley seeds, and by Giles and Riley[3, 4] in microspores of *Tradescantia paludosa*. By removal of most of the oxygen in the tissue the frequency of x-ray induced aberrations was reduced to nearly one-third of that obtained when the tissue was exposed in air or in higher oxygen tensions. Apparently, it is also possible to obtain a similar reduction in aberration frequencies with chemicals. Mikaelsen[5] has shown that reduced glutathione added to the nutrient solution of *Tradescantia paludosa* reduced the frequency of chromosome fragments by about 50 per cent after a 48-hour exposure to chronic γ-radiation at a dose rate of 25 r/day. The results obtained with glutathione suggested testing of other chemicals for protection against radiation induced chromosome aberrations. In the present paper the effect of cysteine, sodium hyposulfite, and sodium cyanide are reported.

Material and Methods.—Cuttings of *Tradescantia paludosa* (clone B2-2) were used as experimental material. Root development was initiated by placing the cuttings in tap water with continual aeration at ordinary greenhouse temperature (65–70°F.).[5] When the cuttings showed good development of primary roots, they were transferred to specially designed lucite vessels which contained Hoagland and Snyder's nutrient solution.[6] The vessels were 30 × 2 × 12 cm. in size. The side of the vessel facing the source was 3 mm. thick.