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Effects of Irradiation on the Pupae and Their Offsprings of *Drosophila virilis*^{1), 2)}

By

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(With 2 Tables)

A number of investigators have reported the genetic effects of irradiation on germ cells in different stages of spermatogenesis in *Drosophila*. These investigations have shown that the effects, such as, mutation rate, dominant lethals and translocations, vary with the developing stages of germ cells (Lüning 1952, Auerbach 1954, Stone 1954, Alexander and Stone 1955, Alexander 1956). Subsequently, Clayton (1957) has reported the detailed observation concerning the differentiation of germ cells in pupal stages of *Drosophila virilis* from the morphological standpoint.

This brief note deals with observations on the differential response to irradiation in *Drosophila* pupae and their germ cells at various developmental stages.

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Material and Methods: The Sapporo strain of *Drosophila virilis* was used as the standard stock for testing radiation effects on the pupae and their developing germ cells. The stock was maintained on the standard cornmeal-molasses-agar food at a temperature of 21°C. In order to facilitate counting eggs, the food was darkened by the addition of carbon powder. Radiation treatments were made from 30 to 100 hours after pupation at 10 hours intervals. These irradiated pupae were allowed to emerge on filter papers, and the emergence-rate was examined. Each of the mature males, which were isolated for five days after emergence, was mated to a non-irradiated virgin female every two days for three successive mating periods. After the male was removed, the number of eggs laid by the normal females was counted. Subsequently, the females were dissected and examined

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motile sperm in the seminal receptacles and spermathecae.

Most of the pupae received 2,000r at 467r per minute from an X-ray machine operating at 200Kv, 25 mA with filters of 0.5 mm aluminium and 0.3 mm copper. The others were irradiated by gamma-ray from cobalt-60 source. The dose was 2,000r for 57 minutes, the distance of the material to the source being 75 cm.

Results with Remarks

Pupae of 1,063 individuals at various stages were irradiated with X-rays and those of 315 specimens with gamma-rays for the test of emergence-rate. These results are given in Table 1. Ninety-one percent of 666 pupae emerged in the control. The pupae irradiated at late stages showed high emergence-rates. However, most of the pupae irradiated at young stages, especially at 30 hours after pupation, died before the emergence.

Table 1. The percentage of adults which emerged from irradiated pupae of *D. virilis*.

	Age of pupae at irradiation	Number of pupae irradiated	Number of emerged adults	Percentage of emergence
X-ray	100	222	184	82.9
	90	195	179	91.8
	80	173	162	93.8
	70	129	114	88.4
	60	209	152	72.7
	50	154	114	74.0
	40	129	14	10.9
	30	106	0	0.0
Gamma-ray	100	28	27	96.5
	90	32	32	100.0
	80	51	47	92.2
	70	53	50	94.3
	60	35	28	80.0
	50	40	28	70.0
	40	39	7	17.9
	30	37	3	8.1
Control		Number of pupae 666	Number of emerged adults 607	Percentage of emergence 91.1

The eggs laid by the females mated with irradiated males for the first time generally showed considerably low survival. The lowest survival of eggs was shown by those from the females mated with the males irradiated at 60 and 70 hours after pupation. In the control for egg-survival, 68.6 percent of 774 eggs developed

Table 2. Radiation damage to the developing germ cells.

Age at irradiation	Mating period	Number of eggs laid	Number of pupae	Hatching-rate	Number of adults
100	1	533	26	4.9	8
	2	350	79	22.6	44
	3	325	164	50.5	103
90	1	515	83	16.1	37
	2	249	101	40.7	80
	3	126	63	50.0	58
80	1	356	8	2.2	7
	2	81	10	12.3	5
	3	25	15	60.0	4
70	1	499	1	0.2	0
	2	272	12	4.4	8
	3	146	2	1.4	2
60	1	246	2	0.8	2
	2	176	6	3.4	4
	3	253	53	20.9	35
50	1	49	8	16.3	4
	2	92	41	44.7	21
	3	33	0	0.0	0
40	1	301	92	30.6	20
	2	45	7	15.6	4
	3	33	20	60.6	8
Control		774	531	68.6	476

into pupae. The data are given in Table 2.

The eggs laid by the females mated for the second time showed higher survival than those by the females for the first time, excepting those from the females mated with the males irradiated at 40 hours after pupation. The lowest pupation-rate was also observed in the eggs from the females mated with the males irradiated at 60 and 70 hours after pupation.

The eggs laid by the females mated with irradiated males for the third time showed the highest survival among the experiment series, with the exception of those originated in the males irradiated at 40, 50 and 60 hours after pupation as shown in Table 2.

Clayton (1957) analyzed the normal meiotic cycle in *Drosophila virilis*. The developmental stages in germ cells in the pupal males observed in this work have agreed with those in Clayton's report. As the latest stages of differentiation, the 40-hour pupae had primary spermatocytes at late prophase, the 50-hour pupae contained primary spermatocytes at telophase, the 60-hour pupae and the older ones included germ cells at postmeiotic spermiogenesis, the 70-hour pupae showed a few sperm bundles, the 100-hour pupae had compact sperm bundles in the lumen

of the posterior part of the testis.

In the present study, most of the pupae irradiated at 30 hours after pupation died before the emergence, so there is no datum which shows immediate connection with the sensitivity of spermatogonia. However, high survival shown in the eggs laid by the females mated for the second or third time may indicate that spermatogonia have great resistance to radiation damage.

High susceptibility to radiation was observed in the eggs laid by the females mated for the first time with the males irradiated at 60 and 70 hours after pupation. These eggs were supposed to have received the spermatozoa irradiated at early spermatids. The eggs laid by the females mated for the first time with males irradiated at 40, 50 and 90 hours after pupation showed relatively high survival. This may show the high resistance of the primary spermatocytes and late spermatids.

Clayton (1962) concluded from her detailed investigation that spermatids are highly susceptible to radiation, and meiotic cells and mature spermatozoa are also radiation sensitive. The results of the present study are in close agreement with those reported by Clayton.

Summary

The differential response of *Drosophila* pupae and male germ cells at various developmental stages to irradiation was studied. 2,000r of X-rays or gamma-rays were irradiated on the pupae in various developing stages. The greatest radiation damage was found in the pupae irradiated at young stages. Most of them died before emergence. On the male germ cells, the highest sensitivity was observed in the early spermatids. The meiotic cells and late spermatids showed relatively high susceptibility.

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