Browning, L. S. University of St. Thomas, Houston, Texas. Recessive lethals produced during oogenesis in D. melanogaster by ethyl methanesulfonate.

Females aged seven days or more were fed ethyl methanesulfonate in sucrose solution for 24 hours according to the method of Lewis (DIS 43: 193). This method of treatment should have insured the presence of one stage 14 oocyte in each ovariole. Two strains of females were used, one having approximately thirty ovarioles per ovary (Oregon R 60) and the other about twelve (Canton S). After treatment, ten bottles containing fifteen females each were mated to Basc males every other day for eight broods, and their F_1 virgin daughters mated individually to Basc males in order to detect recessive lethals in the X chromosome. Controls for the Canton S females showed a low spontaneous rate of 0.1% (4/4,407). The spontaneous recessive lethal frequency for the Oregon R 60 stock has not yet been measured. At the same time, Canton S males were treated and their daughters tested for recessive lethals by the Basc technique to confirm the mutagenicity of the chemical. The results are shown in the table below.

Broods (2-day)	Females: OR R 60 No.	OR R 60 L	OR R 60 %	Females: C S No.	C S L	C S %	C S Males No.	C S Males L	C S Males %	C S Controls No.	C S Controls L	C S Controls %
1	60	3	5.0	44	3	6.8	30	11	36.6	1,069	1	0.1
2	400	22	5.5	377	13	3.5	148	67	45.1	921	1	0.1
3	412	14	3.4	380	13	3.4	181	70	38.6	522	0	0.0
4	436	20	4.6	418	4	1.0	130	56	43.0	759	1	0.1
5	72	11	15.3	380	12	3.2	208	75	36.1	534	0	0.0
1-5	1,380	70	5.1	1,599	45	2.8	697	279	40.0	3,805	3	0.1
6	319	14	4.4	439	12	2.7	130	12	9.2	602	1	0.1
7	391	19	4.9	217	3	1.4	182	16	8.8			
8	202	8	4.0	-	-	-	64	1	0.1			
6-8	912	41	4.5	656	15	2.3	376	29	7.7	4,407	4	0.1

The continued appearance of recessive lethals through the sixteenth day after treatment shows that the mutagen is remarkably effective at all stages of oogenesis, including the germarium or oogonial cells. Since it takes approximately three days for an egg to pass from stage 1 to stage 14 at a maximum rate of egg-laying (R. C. King, 1957, Growth XXI, 95-102), eggs that were laid in broods 1 or 2 would probably have been in various stages of maturation at the time of treatment, and later broods would have been derived from cells that were oogonia at the time of treatment, although no egg counts were taken from individual females. Also, because of the mass mating of females, clusters or the presence of pre-existing lethals could not be detected. However, a record was kept of the lethals recovered from each bottle, and in both the Oregon R 60 and Canton S females the distribution of mutations and their frequencies were roughly similar. Even though the possibility exists that certain females retained their eggs much longer than others and so might have laid eggs that were in varying stages of sensitivity in broods subsequent to brood 2, it seems very unlikely that this would have persisted after brood 5, when they would have been 10 days post-treatment. It has been shown that the recessive lethal frequency in the second pair of autosomes is as low for eggs laid 10-15 days after acute irradiation with 4000r X-rays as for those laid later than 15 days after irradiation (Muller and Meyer, 1961, Genetics 46: 882). As will be seen from the table, a total of 56 lethals were recovered in 1,568 tests from the two types of females combined between the tenth and sixteenth days after treatment. Our average rate of 4.5% found in the Oregon R 60 females in these broods is double the $2.1\pm0.2\%$ found after acute X-ray irradiation of 4000r in very large-scale tests made by other workers (Muller, Oster, and Zimmering, 1963, Repair from Genetic Radiation Damage, Sobels, Ed., pp. 275-304). It might be pointed out, however, that this chemical produced from broods 1 through 7 far fewer mutations than are produced in the male germ line, in contrast to our finding that chloro-ethyl-methanesulfonate produces more mutations in the female than in the male germ line (Browning and Altenburg, 1965, Genetics 52: 431).

Parker has reported (1963, Repair from Genetic Radiation Damage, Sobels, Ed., pp. 11-19) that after X-irradiation, stage 7 oocytes are about one-half as sensitive as stage 14

oocytes when recessive lethals are measured but are one-tenth to one-twentieth as sensitive when hatchability is measured. He postulates that the increase in sensitivity of stage 7 oocytes with regard to recessive lethals may be due to an increased production of chromosomal aberrations, perhaps small deficiencies. It would be of interest to see if EMS behaves similarly when the same treatment techniques are applied.

Although an increase in recessive lethal frequencies is not shown clearly for each brood when Oregon R 60 females are compared with Canton S females, the table does indicate that a higher overall frequency for the Oregon R females probably exists, presumably due to the larger number of ovarioles. The use of this stock might then make the study of mutations arising in the female germ line less laborious.

Rai Chaudhuri, A. and A.S. Mukherjee. University of Calcutta, India. Developmental changes in puffing pattern in the mutant "ft" in D. melanogaster.

The mutant "fat" (ft;2:12.0) of D. melanogaster shows certain "vacuolar lipo-protein bodies" in the larval salivary gland cells (Slizynski, 1964; Rai Chaudhuri, 1968). This effect is accompanied by an initiation and increase in puffing activities in various sites of their chromosomes. Analysis of the sequential changes in the puffing pattern of these sites in ft during the late third instar to prepupae has been made and summarily presented below.

Table 1. Comparison of puffing activity in the wild type and ft third instar larvae and prepupae.

Group	puffing Sites	Oregon R+		ft	
		Larvae	Prepupae	Larvae	Prepupae
A	15CD	++	±	-	+
	18B	+	++	-	+
	53DE	+	++	-	+
	66D	+	±	-	+
	83EF	±	++	-	+
	85CD	±	±	-	±
	85EF	+++	++	-	++
B	42B	++	++	+++	-
	100EF	++	±	+++	±
C	7B	-	-	-	+
D	1A	-	-	+	+
E	2B	++	+	+++	++
	21B	++	+	+++	++
	61A	+	-	++	+
	74EF	+	-	+++	-
	75AB	+	-	+++	-
	47A	++	+	++	++
	50CD	++	+	+++	++
	72BC	+	+	++	+

Altogether 77 sites have been found to show activity during one or the other stages (from late third instar to prepupa). Among them, 42 were active during the late third instar, and the remaining 35 sites were active only during the prepupa; 23 puffs were active during both stages.

A comparative analysis of puffing patterns in ft larvae and prepupae with those in Oregon R+ shows (Table 1) that 7 puffs which are present either during the late third instar or prepupa in the wild type are absent in the ft larvae (Group A). Two puffs present in Oregon R+ larvae and prepupae are super-activated in ft larvae only (Group B). A single puff one each in Groups C and D is present either in prepupae (Group C) or in both stages of ft (Group D). Five puffs in ft larvae and three puffs in ft prepupae become more activated as compared to those in Oregon R+ (Group E). Three other puffs which are present in both stages of Oregon R+ and ft show a reduced activity in wild type strain as compared to ft larvae and prepupae (Group F).

Table 1. Legend:
+++ : Activity index 2 or more
++ : Activity index >1.6<2
+ : Activity index ~1.5
± : Activity index ~1.2 to 1.3
- : Activity index 1.0

References: Rai Chaudhuri, A., 1969, DIS 44: 118. Slizynski, B.M., 1964, Cytologia 29: 330-336.