

Pachciarz, J.A.* and W.M. Luce. University of Illinois, Urbana, Illinois. The effect of caffeine on axenically grown *D. melanogaster*.

During an investigation of caffeine mutagenesis in *D. melanogaster*, two effects of caffeine were noted: 1) a marked increase in the length of the developmental period; 2) a corresponding decrease in the percent of larvae surviving to adulthood.

Previous studies on caffeine mutagenesis (Andrews, 1959; Yanders and Seaton, 1962) have utilized standard media. This study used nucleic acid free, chemically defined medium and the strain Livingston red, kindly provided by R. Rayle, which was axenically grown. The effect of several concentrations of caffeine in this medium was tested by the Muller-5 technique for sex-linked recessive lethals.

Livingston red eggs less than twenty-four hours old were collected, sterilized by a modification of Geer's method (Geer, 1963), and aseptically transferred to sterilized vials containing 5 ml of Geer's medium, omitting RNA and substituting sucrose for fructose. Each vial contained an average of four ± .5 larvae. Caffeine concentrations in the medium ranged from .0025% to .1%. Larvae were scored every three days for viability, and male adult survivors were mated to Muller-5 virgins. F₁ females were individually mated to test for sex-linked recessive lethals.

% Caffeine	% Larvae Reaching Adulthood	% Pupae From Larvae	No. of Broods	Length of Developmental Period	Matings LR♂ x M5♀	No. of chromosomes Tested	No. of lethals	No. Non-lethals	Mutation Rate
0	23	44	4	20d ± 4	10	551	3	390	.77%
.0025	14.4	18.4	2	24d ± 2	9	551	2	419	.48%
.005	2	5	3	34d ± 7	0	-	-	-	-
.01	-	-	3	-	1	18	0	14	0%
.1	0	0	1	death at 7-9 d	0	-	-	-	-

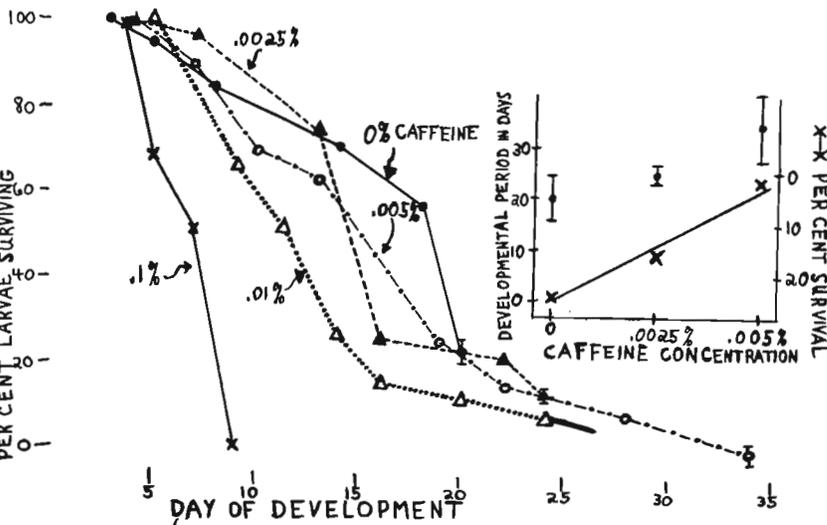
Vials found to be contaminated are not included in the data. Periodic checks were made for the presence of aerobic bacteria and molds by plating on bacteriological media, but the presence of anaerobic bacteria, Mycoplasma, or viruses could not be excluded.

Forty-seven male flies were obtained and twenty successful matings were made, with 1120 chromosomes being tested. Results are summarized in the table.

Of the three lethal control mutations, two were cluster events with the flies phenotypically all Muller-5, one a block of two vials and the other a block of twenty-eight vials. The third control mutation and the two lethals with .0025% caffeine were single events. These lethal mutations were verified by F₂ crosses.

The graph shows the effect of caffeine concentration on the length of the developmental period and on the percent of larvae surviving to adulthood.

While no statistically valid conclusion can be drawn from the sex-linked recessive lethal mutation rates, caffeine is shown to prolong the developmental period with a corresponding decrease in survival of larvae, and this effect seems to be proportional within the limits of the caffeine concentrations used.



That these low concentrations of caffeine demonstrate such an effect may be potentiated by the axenic nature of the experiment.

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References: 1. Andrews, L.E. 1959, *Am. Naturalist* 93: 135. 2. Yanders, A.F. and Seaton, R.K. 1962, *Am. Naturalist* 96: 277. 3. Geer, B.W. 1963, *J. Exp. Zool.* 154: 353.

* Now in the Department of Microbiology, St. Louis University, St. Louis, Missouri.