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GENOTOXIC ACTIVITY OF NAPHTHYL CARBAMATE IN THE LARVAL STAGES OF *DROSOPHILA*

L. DEY, B. MAJHI, N. K. TRIPATHY and C. C. DAS

Department of Zoology, Berhampur University, Berhampur 760 007, India.

FOR an evaluation of the practical significance of mutagens, the value of the sex-linked recessive lethal (SLRL) test in *Drosophila melanogaster* is generally recognized[1]. Today this test is regarded as the most sensitive one available in the *Drosophila* system as it can detect point mutations and other genetic changes[2–5].

Naphthyl carbamate (Sevin) is shown to induce chromosomal breakages in plants[6]. Brzheskii[7] described his results in the SLRL tests as inconclusive. Sinha and Sinha[8] demonstrated the induction of sex-linked recessive lethals by Sevin in *Drosophila*. Because of conflicting reports on the mutagenicity of Sevin in different systems, the present study was undertaken to evaluate its mutagenic property through SLRL tests in the larval stages of *D. melanogaster*.

Three *Drosophila* stocks, 1. w^{co}/w^{co}, 2. $fs(1)\ K_{10}$ w/Y and 3. M-5/M-5 (for details of markers please see Lindsley and Grell[9]) have been used in the present studies. All these stocks are maintained on standard cornmeal–molasses–agar–yeast culture medium at 25 ± 2°C. The eggs were collected after mating the virgins of stock-1 with males of stock-2 on petri dishes containing normal food for duration of four hours. On hatching, different aged (72 hr, 48 hr and 24 hr) larvae were collected by floating them off in 50% glycerine and were fed in vials with Sevin supplemented food for a period of 48, 72 and 96 hr respectively. The LD_{50} (median tolerance limit) dose of different larval treatments was determined by counting the emerged adults from the vials. For each treatment hour, the LD_{50} dose (lethal) and 50% of this dose (sublethal) were taken into account (table 1) and the standard Muller-5 mating scheme was carried out[10,11]. The statistical methods are based on the tables of Kastenbaum and Bowman[12] and the χ^2 test of goodness of fit to the Poisson distribution.

The mutagenicity of naphthyl carbamate on the male germ line cells of *D. melanogaster* has been tested through the M-5 technique and the results are summarized in table 1. Figure 1 depicts the comparison of lethal induction between the control and the treated series. The frequency of induction of sex-linked recessive lethality increases over the control frequency in all the treated series at 1% level of significance. The dose-effect curves (figure 2) indicate the increase in lethality is related to both the concentration of Sevin as well as the treatment hours.

The 24 hr and 48 hr larvae, on being fed with the LD_{50} dose, yielded 2 individual males in the former and 1 from the latter-two lethal chromosomes each. These appear to be an indication of induction of clusters[13,14]. Such clusters are expected to have a common origin when the lethality is induced in premeiotic cells of the developing gonad. Clark[13]

Table 1 *Induction of sex-linked recessive lethals after naphthyl carbamate exposure*

Larval treatment	Conc. (%)	Males tested	Tested chromosomes (Mean ± SD)	Chromosomes			Lethal chromosomes per male		
				Total	Lethal	%	0	1	2
Control	0.0	254	18.42 ± 5.67	4678	4	0.08	250	4	0
48 hr	1.0×10^{-2}	68	14.8 ± 2.24	1007	8	0.79	60	8	0
	5.0×10^{-3}	70	15.0 ± 1.06	1050	6	0.57	64	6	0
72 hr	5.0×10^{-3}	69	13.04 ± 2.13	900	8	0.89	62	6	1
	2.5×10^{-3}	62	15.0 ± 2.23	930	7	0.75	55	7	0
96 hr	5.0×10^{-3}	69	16.0 ± 1.04	1104	11	0.99	60	7	2
	2.5×10^{-3}	65	14.0 ± 1.04	910	7	0.77	58	7	0

Significance test[12] in all cases showed a value at 1% level.

emphasized that clusters are expected when early larval stages are exposed to a chemical mutagen, since the testes at this stage contain spermatogonial and spermatocyte stages[15,16]. However, our data do not support this view of clusters. On performance of Poisson distribution followed by a χ^2 goodness of fit, it was found that the actual and expected numbers are very close ($P > 0.05$). We therefore conclude that the lethal mutations probably originated from independent events and do not have a common origin as in clusters.

Sevin induces chromosome breakage in plants[7] and chromosome loss in *Drosophila*[8]. Our results support the findings of Brzheskii, i.e., naphthyl carbamate does act as a chemical mutagen to induce sex-linked recessive lethals in *D. melanogaster* in higher frequencies possibly through deletion and/or point mutation.

Our results in *Drosophila* SLRL tests were inconclusive by the adult feeding method. But our present data after larval feeding indicate an increase of induction of sex-linked recessive lethals. Vogel[17] pointed out that food consumption is very high during larval life and feeding the larvae might well become a useful application procedure for mutagen testing, since it would tend to increase exposure to the mutagen and he was able to show that larval stages are more sensitive than adults to the

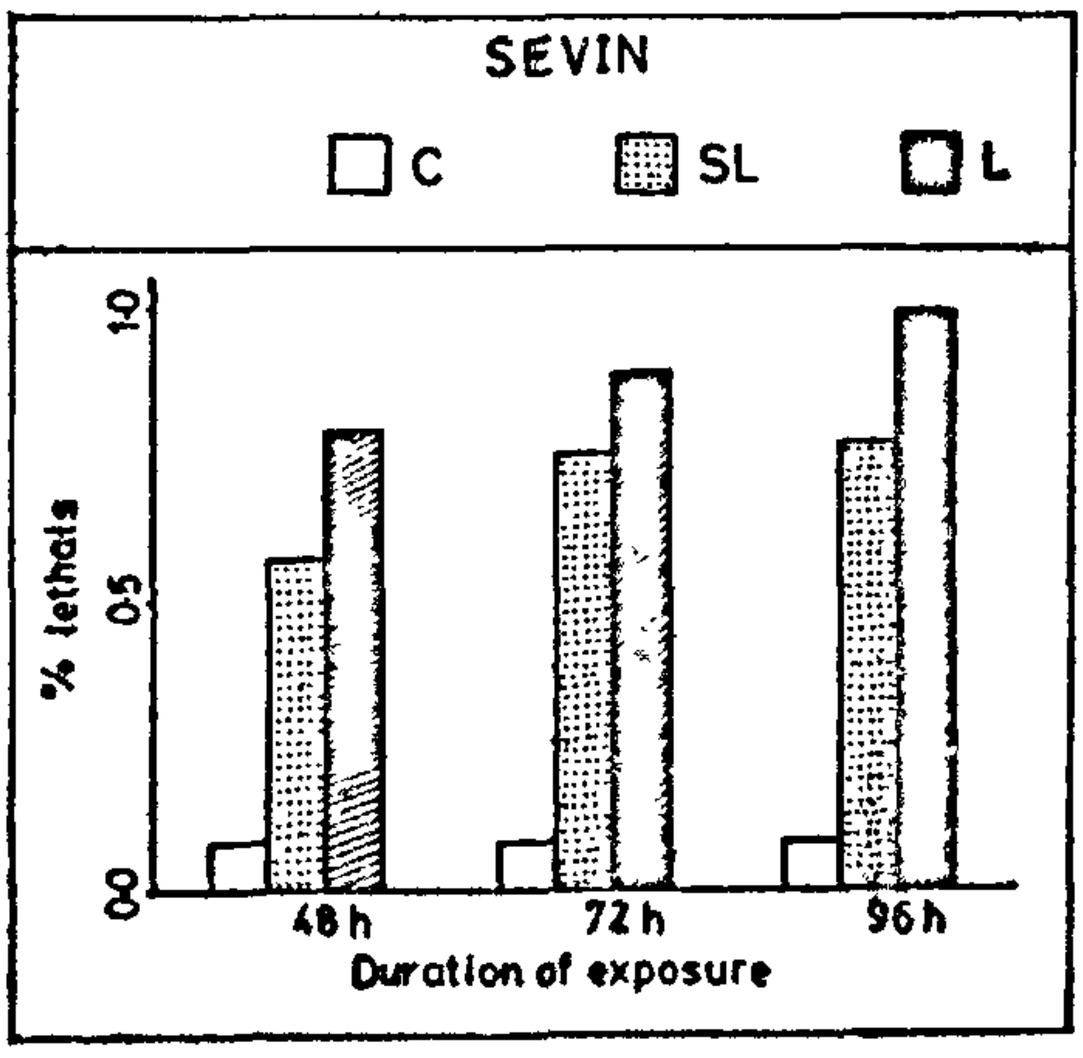

Figure 1. Frequency of induction of sex-linked recessive lethals on exposure of larvae to the lethal and sublethal doses of Sevin for 48 hr, 72 hr and 96 hr. [C = control; SL = sublethal; L = lethal.]

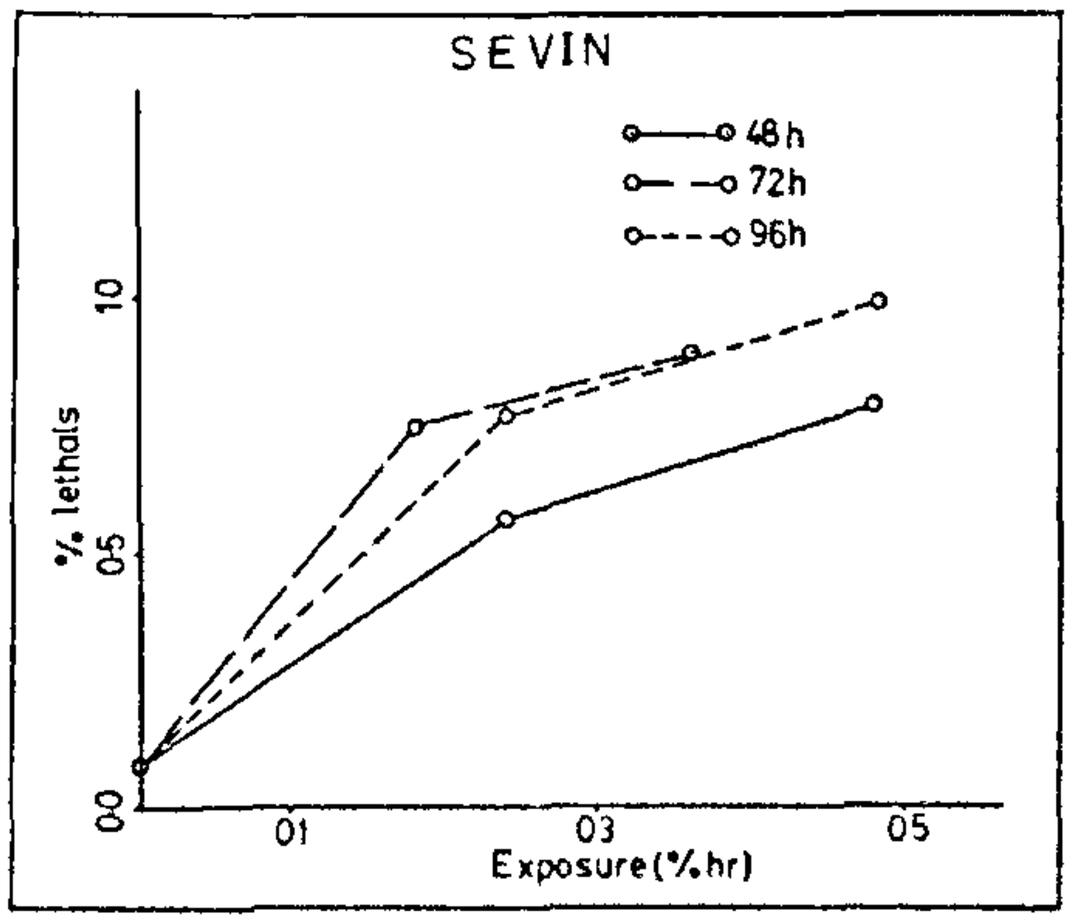

Figure 2. Dose-effect curves for the induction of sex-linked recessive lethals on exposure of larvae to the lethal and sublethal doses of Sevin for 48 hr, 72 hr and 96 hr. Data not corrected for spontaneous induction of sex-linked recessive lethals.

mutagenicity of diethylnitrosamine. Angus *et al*[18] reported that certain triarylmethane dyes used as food colouring agents, and which are mutagenic in the *Salmonella* test, show much higher levels of mutagenicity when fed to *Drosophila* larvae than when fed or injected into adult males. In general, it can be concluded that larval stages are often more sensitive than adults for screening purposes.

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ACCUMULATION OF B-CHROMOSOMES IN *DROSOPHILA NASUTA ALBOMICANA*

N. B. RAMACHANDRA and
H. A. RANGANATH
Department of Zoology, University of Mysore, Manasagangotri, Mysore 570 006, India.

Drosophila is a potent eukaryotic system to explore many facets of population cytogenetics of chromosomal variability. Despite extensive studies on the chromosomes of *Drosophila*, B-chromosomes in *Drosophila* have been reported only recently[1,2]. The present note reports preliminary studies on B-chromosomes of *Drosophila nasuta albomicana*.

D. n. albomicana is most advanced in the *nasuta* subgroup of the *immigrans* species group of *Drosophila*[3–10]. The different dimensions of population cytogenetics of the B^+ and B^- strains of *D. n. albomicana* concerning their competitive fitness[11], resource utilization divergence[12], population fitness at different temperatures[13] and the characterization of heterochromatin (paper preparation) have been studied. During these studies, the frequency distribution of B-chromosomes of *D. n. albomicana* has been analysed over a period of three years and the results are reported.

The relative frequency of B-chromosome distribution was analysed during February 1983, August 1985 and February 1986. For each one of these assessments, 100 larvae were chosen at random to reveal the frequency distribution of B-chromosomes employing the temporary squash method[9].

Table 1 gives the number of individuals with B-chromosomes and the mean number of B-chromosomes per individual along with the standard error during three years of study. The mean number of B-chromosomes per individual during February 1983, August 1985 and February 1986 was 1.03, 1.88 and 2.47 respectively. Table 2 provides information about the number of individuals with different number of B-chromosomes analysed during the three years of study. During February 1983, 67 out of 100 individuals had B-chromosomes and the maximum number of B-chromosomes recorded in an individual was 3. After