MORTALITY, STERILITY, FECUNDITY AND NON-DISJUNCTION INDUCED BY CHLOROETHYL METHANESULPHONATE (CEMS) AND CYCLOHEXANE (CH) IN *DROSOPHILA MELANOGASTER*

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CHLOROETHYL methanesulphonate (CEMS) has been shown to induce recessive lethal, dominant lethal, visible mutations, crossing over, translocation, non-disjunction, chromosomal aberrations and deletions in *Drosophila melanogaster*[1-9]. Cyclohexane (CH) has been shown to induce mutations in yeast and bacteria[10,11], crossing over in *D. melanogaster*[12]

There is paucity of information on the combined mutagenic effects of CEMS and CH. This note is the first report on the mutagenicity of CEMS and CH when administered independently and in combination in *D. melanogaster*.

Freshly-prepared Drosophila standard media were mixed with the chemicals at 50–60° C. About 300 pairs of flies were allowed to lay eggs in quarter pint bottles containing Drosophila culture media. The flies were moved from the bottles after 4–5 hr. After 24 hr, about 200 first instar larvae were transferred to the bottles containing food with different concentrations of the chemicals. The treated larvae were allowed to develop at 20 ± 2° C. Flies from these bottles were collected to calculate the mortality rate and sex ratios. Flies obtained from LD_{50} were further used for mating, to the required strains to assess the mutagenicity. Control experiment was simultaneously conducted.

The results obtained for different series have been presented in table 2. CEMS (0.02%) and CH (0.3%) when administered independently, the mortality was 47% and 48% respectively. The combined treatment of CEMS and CH (0.03 + 0.2%) resulted in the death of 63% of the population as against 9% of control value (table 1). The emergence of adults completed between the 9th and 14th day in series A, 12th and 22nd in series B, 12th and 21st in series C and 14th and 26th day in series D (figure 1). Comparatively, in series D, the required period for the development of larvae to adult

TABLE 1

Percentage of mortality in D. melanogaster treated with different concentrations of CEMS, CH and CEMS + CH.

Concentration %	No. of larvae treated	No. of larval and pupal death	No. of adults emerged	Mortality % Mean ± SD values
CEMS				
0.01	1200	430	770	35.83 ± 2.14
*0.02	1200	566	634	47.16 ± 1.19
0.03	1200	716	384	59.66 ± 2.04
0.05	1200	905	295	75.51 ± 2.31
Control	1200	106	1094	8.83 ± 0.69
CH				
0.1	1200	363	837	30.25 ± 1.18
0.2	1200	451	749	37.58 ± 1.32
*0.3	1200	576	624	48.00 ± 1.46
0.4	1200	722	488	60.00 ± 2.54
0.5	1200	835	365	69.00 ± 2.07
Control	1200	102	1098	8.50 ± 0.14
CEMS + CH				
0.01 + 0.05	1400	363	1036	26.00 ± 1.03
0.02 + 0.1	1400	447	953	31.92 ± 0.91
0.02 + 0.2	1400	622	778	44.42 ± 0.83
*0.03 + 0.2	1400	753	647	53.78 ± 0.48
0.04 + 0.3	1400	801	519	62.92 ± 1.15
Control	1400	127	1273	9.00 ± 0.60

* Individuals survived at this concentration (LD_{50}) were used for genetic variations induced.

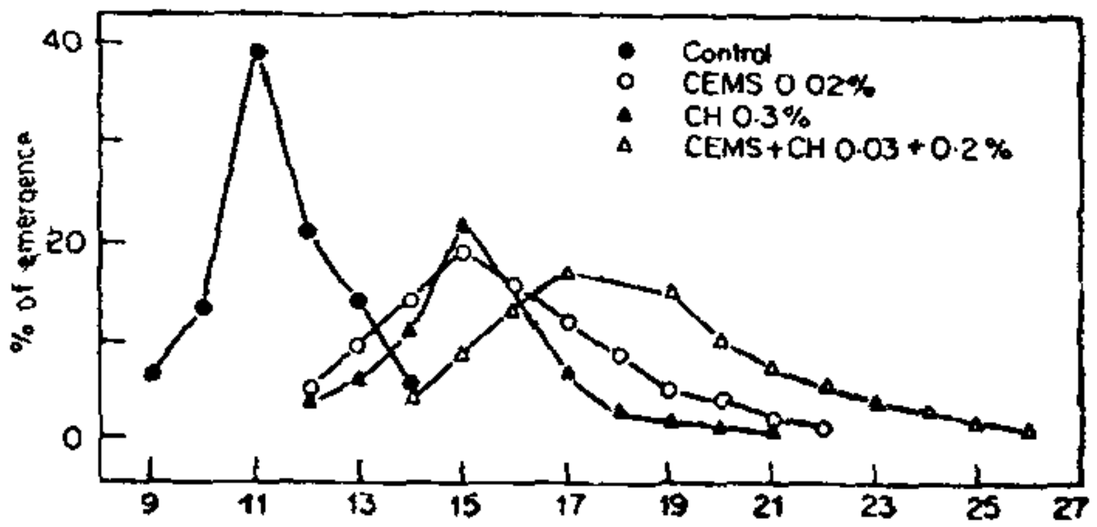

Figure 1. Pattern of emergence of *D. melanogaster* after treatment with different chemicals.

is more and the rate of emergence is very slow than in the other series. This is attributed to the cytotoxic effects of CEMS and CH, in combination (figure 1 and table 1).

Virgin flies (2–3 days old) collected from the series A, B, C and D were further tested for induced genetic endpoints as estimated by the disturbance in the segregation pattern of the sex pairs of chromosomes, by mating to required strain. The percentage of sterility in the series A is very low exceptionally high in series D and it is normal in series B and C (table 2). The high frequency of sterility has been attributed to the induced genetic effects of CEMS + CH.

The rate of fecundity in the series A is 42 and 48; 26 and 20 in series B; 36 and 37 in series C, while it is only 18 and 19 in series D among males and females respectively. The rate of fecundity is comparatively low in series B and C and it is very low in series D (table 2).

The frequency of non-disjunction is 0.066% and 0.073% in series A, 0.404 and 0.533% in series B and 0.353 and 0.399% in series C and 0.92 and 1.18% in series D, in males and females respectively (table 2). Non-disjunction has also been reported in *D. melanogaster* after treatment with CEMS[9].

The frequency of genetic endpoints as determined by the frequency of loss and/or gain of the sex chromosomes in males and females is very high in the group treated with CEMS and CH, as against those groups which were exposed to the chemicals independently. The high rate of mortality and sterility is attributed to the cytotoxicity and inhibitory action of CEMS + CH on the dividing somatic cells. Further, CEMS and CH behave as potential mutagenic in the dividing germ cells of *D. melanogaster* and their action may be synergistic, additive or both.

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TABLE 2

Percentage of sterility, non-disjunction and rate of fecundity induced by CEMS and CH independently and in combination in D. melangaster.

Treatment %	Sex	No. tested	% of sterility*	Total progeny	Rate of fecundity	% of non-disjunction**
Control	Male	180	1.60	7504	42.39	0.066
(Series A)	Female	170	1.76	8125	48.65	0.073
CEMS (0.02)	Male	194	3.09	4946	26.30	0.404
(Series B)	Female	171	7.01	3181	20.00	0.533
CH (0.3)	Male	187	8.02	6224	36.18	0.353
(Series C)	Female	182	15.93	6103	39.88	0.399
CEMS + CH						
(0.03 + 0.02)	Male	190	12.10	3145	18.83	0.920
(Series D)	Female	195	24.61	2845	19.36	1.180

* Percentages of sterility of males and females in control (Series A) are not significant.
** There is no difference between males and females as far as the percentage of non-disjunction is concerned in both control and treated series.
Percentage of non-disjunction in case of CEMS (Series B) is not significantly different from CH (Series C).
All other comparisons have been highly significant ($P < 0.01$).

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COMBINING ABILITY FOR RESISTANCE TO BLAST DISEASE IN RICE

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THE concept of combining ability in disease resistance is a novel idea to bring out the best combiners for breeding programme. Studies are available on the combining ability of rice to bacterial blight resistance[1], wheat to brown rust resistance[2], maize to *Helminthosporium mavdis*[3] and maize to brown stripe mildew[4]. Though Zenith, Tetep and Tadukan are utilized by breeders and pathologists in most of the rice-growing countries of world, as blast donors in resistance study, the efficiency of these rice varieties in combining ability for blast resistance has not yet been investigated. In the present study attention has been paid to construct superior genotypes with the aid of additive genetic variance in the self-fertilized crop, the rice for selecting the best combiners in blast resistance.

Seven rice varieties were chosen from the germplasm stock of the Central Rice Research Institute assuming a high degree of homozygosity due to inbreeding of more than 8 generations. The varieties *viz.* Zenith, Tetep and Tadukan were resistant. Jaya was moderately resistant and Ratna, Karuna and Co. 13 were susceptible to the isolate C_2 used in the present study. Seeds of parents, crosses and reciprocals (from the complete diallel set) were grown in zinc trays (80 × 80 × 10 cm) by complete random design (49 microblocks/tray) with 3 replications. Trays were filled with field soil amended with farm manure. Seeds were germinated by blotter method and 5 germinated seeds were kept in a microblock in each tray at a distance of 5 cm apart from seed to seed. Seeds of Karuna were sown surrounding the varieties close to the tray wall for detection of artificial infection. Ten g of nitrogen were applied to each tray with 15-day old seedlings.

Artificial inoculation was carried out according to Padmanabhan *et al*[5]. Seedlings (21-day old) in inoculation chambers were sprayed uniformly by an atomizer with a spore suspension of *Pyricularia oryzae*, containing approximately 10^5 spores/ml suspension. Spraying took place late in the evening. Relative humidity was maintained at above 90% by keeping the thick gunny hessian surrounding the inoculation chamber wet. Disease was rated on the 10th day after inoculation according to Padmanabhan and Gangully[6] on the 2nd leaf from the top[7] and averaged over 5 seedlings. The combining ability was statistically analysed according to model I method I of Griffing[8]. The experiment was conducted during 1981 *rabi*.

Significant differences were observed among the genotypes for disease reaction. Table 1 shows that variation due to general combining ability (*gca*) and specific combining ability (*sca*) was highly significant for disease reaction. However, variance due to reciprocals was non-significant. The variance of *gca* was much higher than those due to *sca* suggesting that additive genetic variance was more important than non-additive variance. Effects of *gca* were estimated

TABEL 1

Analysis of variance for combining ability

Source	Df	Mss	F
Gca	6	499.9338	80.7178[a]
Sca	21	70.3594	11.3600[a]
Reciprocals	21	10.2984	1.6627
Error	96	6.1936	

[a]Significant at 1% probability

for the seven varieties for disease reaction (table 2). The lowest *gca* values were recorded in Zenith, Tetep and Tadukan *i.e.* -5.754, -5.809 and -5.782 respectively while the highest value was recorded in Co. 13 (8.140). All the *gca* values were highly significant except Jaya which was significant at 5% level. Analysis of C. D. indicates that the resistant varieties were differentiated from the susceptible ones and had almost equal *gca* values. This depicts that Zenith, Tetep and Tadukan were the best combiners to evolve blast-resistant and high-yielding rice varieties.

Sca data revealed that F_1 hybrids (table 2) showing maximum negative (maximum resistance) effects were in Tetep × Co. 13 (-8.240), Tadukan × Co. 13 (-8.217) and Zenith × Co. 13 (-8.145). Individual *sca* values in Zenith × Jaya, Tetep × Jaya, Tadukan × Karuna, Jaya × Ratna, Jaya × Karuna were insignifi-