

## GENETIC EFFECTS OF THINNER, BENZENE AND TOLUENE IN *Drosophila melanogaster*

### 2. SEX LINKED RECESSIVE LETHAL MUTATIONS AND TRANSLOCATIONS II-III

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#### ABSTRACT

The effects of thinner, benzene and toluene on the induction of sex-linked recessive lethal mutations and translocations II-III in *Drosophila melanogaster* were investigated by means of a genetic scheme designed by Oster and a modified z for statistical analysis. Thinner induced sex-linked recessive lethals while benzene and toluene did not. Translocations II-III were produced only by benzene.

#### RESUMEN

Se investigaron los efectos del tñner, del benceno y del tolueno sobre la inducción de mutaciones letales recesivas ligadas al sexo y sobre la frecuencia de translocaciones de los cromosomas II y III en *Drosophila melanogaster*. Se utilizó el sistema de cruas Oster y la prueba de diferencia de proporciones (z modificada) para valorar estadísticamente los resultados. El tñner indujo mutaciones letales recesivas ligadas al sexo, mientras que el benceno y el tolueno no las produjeron. Las translocaciones II-III fueron producidas solamente por el benceno.

#### INTRODUCTION

*Drosophila* has been employed to test the whole spectrum of genetic damage induced by chemicals such as dominant lethals, chromosome loss, non-disjunction and translocations (Sobels 1974; Zimmering 1975; Vogel and Sobels 1976). The

genetic events are scored as segregations which are clearly due to gene mutations (Kilbey *et al.* 1981). *Drosophila* microsomes have the ability to biotransform compounds that require metabolic activation (Baars *et al.* 1980).

This paper reports the results obtained with thinner, benzene and toluene on the induction of sex-linked recessive lethal mutations and translocation II-III in *Drosophila melanogaster*.

## MATERIALS AND METHODS

The genetic scheme designed by Oster (1958) was employed. For sex-linked recessive lethals the following crosses were done.

$$\begin{array}{lcl}
 \text{P "Oster" females} & \times & \text{"Oster" males} \\
 y \text{ sc}^{S1} \text{ Inv 49 sc}^8; \text{ bw; st, pp} & & \text{X}^{C2} yB/\text{sc}^8 Y \\
 \quad \quad \quad (y \text{ white}) & & \quad \quad \quad (y B) \\
 \text{F}_1 \text{ Females: } y \text{ sc}^{S1} \text{ Inv 49 sc}^8/\text{X}^{C2} y B (y B) & & \\
 & \times & \\
 \text{F}_1 \text{ Males: } y \text{ sc}^{S1} \text{ Inv 49 sc}^8/\text{sc}^8 Y (wild) & & \\
 \text{F}_2 \text{ Females: } y \text{ sc}^{S1} \text{ Inv 49 sc}^8/y \text{ sc}^{S1} \text{ In 49 sc}^8 (y) & & \\
 \quad \quad \quad y \text{ sc}^{S1} \text{ Inv 49 sc}^8/\text{x}^{C2} y B (y B) & & \\
 \text{F}_2 \text{ Males: } y \text{ sc}^{S1} \text{ In 49 sc}^8/\text{sc}^8 Y (wild) & & \\
 \quad \quad \quad \text{X}^{C2} y B/\text{sc}^8 Y (B/+) & & 
 \end{array}$$

The induction of sex-linked recessive lethals was scored when the phenotype B + in males was not present, being the sex ratio 2:1 instead of 2:2.

For translocation II-III the following crosses were done:

$$\begin{array}{lcl}
 \text{P "Oster" females} & \times & \text{"Oster" males} \\
 \text{bw/bw; st p}^D/\text{st p}^D & & +/+; +/+; +/+ \\
 (white eyes) & & (wild eyes) \\
 \text{F}_1 \text{ Females and Males: bw/+; st p}^D/+ + & & \\
 \quad \quad \quad (wild eyes) & & \\
 \text{P}_2 \text{ Females bw/bw; st p}^D/\text{st p}^D & \times & \text{Males (F) bw/+; p}^D/+ + \\
 (white eyes) & & (wild eyes) \\
 \text{F}_1 \text{ bw/+; st P}^D/+ +, \text{ bw/+; st p}^D/\text{st p}^D & & \\
 (wild) & & (scarlet pink peach) \\
 \text{bw/bw; st p}^D/+ +, \text{ bw/bw; st p}^D/\text{st p}^D & & \\
 (brown) & & (white)
 \end{array}$$

P males were treated orally with solvents at different concentrations. Males and females were allowed to mate for three days after treatment. Fifteen days later, F<sub>1</sub> was scored and F<sub>1</sub> males were mated with "oster" females of the constitution bw/bw; st p<sup>D</sup>/st p<sup>D</sup>. Females were put into fresh vials (one per vial) and fifteen days later F<sub>2</sub> flies were scored.

The assayed solvents were: Thinner \* at concentrations of 0.5, 1.0, 1.5, 2.0 2.5 and 3.0%; benzene (Balker) at 0.10, 0.25, 0.50, 0.75, 1.00 and 1.25%; and toluene at 0.10, 0.25, 0.50, 0.75, 1.00, 1.25 and 1.50%. LD<sub>50</sub> was the highest concentration used for all substances. The solvents were administered orally with food. For all experiments and concentrations, parallel controls were run. All experiments were carried out at 25°C ± 1. Statistical significance tests were done on the basis of a modified z (Spiegel 1961).

## RESULTS AND DISCUSSION

Tables I to III summarize the results obtained. The spontaneous frequency of sex-linked recessive lethals was 0.70%, while translocations varied from 0.19 to 0.45% in the present report. Thinner (Table I) induced sex-linked recessive lethals, while benzene (Table II) and toluene (Table III) did not prove to be mutagenic.

Benzene did not induce sex-linked recessive lethals. These results were in agreement with those reported by Kale and Baum (1983) in *Drosophila melanogaster*. Also the results obtained with toluene in the present report were in agreement with those of Donner *et al.* (1981) and Norppa *et al.* (1981) in the same species. Translocations II-III were induced only by benzene (Table II).

It has been shown that some chemicals induce sex-linked recessive lethal mutations at high exposure levels, while the doses needed to induce translocations should be even higher (Ahon and Lee, 1978). In the present report thinner induced sex-linked recessive lethals. This could be due to the presence of other solvents like ethanol, isopropanol, ethyl acetate, n-hexane and n-heptane in the mixture.

*Drosophila* has demonstrated the existence of mutagens that are efficient in

TABLE I. INDUCTION OF SEX-LINKED RECESSIVE LETHALS AND TRANSLOCATIONS IN *Drosophila melanogaster* MALES TREATED WITH DIFFERENT CONCENTRATIONS OF THINNER

Concentration %	Chromosomes with sex-linked recessive lethals	Frequency %	Chromosomes with translocations II-III
Control	0/249		0/281
0.5	6/254	2.36*	0/301
1.0	7/275	2.54*	0/323
1.5	7/244	2.86*	0/335
2.0	8/241	3.32*	0/299
2.5	9/270	3.33*	0/302
3.0	7/212	3.31*	0/289

\*  $p < 0.05$ .

\* Gas chromatogram run in the Centro Mexicano de Salud Mental showed that thinner constituents are: toluene 52.0% n-hexane, 25.5%, ethano 12.5%, ethyl acetate 6.0%, isopropanol 2.0%, benzene 1.0% and n-heptane 1.0%.

TABLE II. INDUCTION OF SEX-LINKED RECESSIVE LETHALS AND TRANSLOCATIONS IN *Drosophila melanogaster* MALES TREATED WITH DIFFERENT CONCENTRATIONS OF BENZENE

Concentration %	Chromosomes with sex-linked recessive lethals	Frequency %	Chromosomes with translocations II-III	Frequency %
Control	3/450	0.70	2/435	0.45
0.10	3/262	1.15	3/270	1.11*
0.25	3/260	1.15	3/273	1.09*
0.50	2/383	0.52	3/263	1.14*
0.75	3/410	0.73	3/256	1.17*
1.00	3/335	0.89	2/265	1.13*
1.25	2/264	0.75	3/209	1.43*

\*  $p < 0.5$ .

TABLE III. SEX-LINKED RECESSIVE LETHALS AND TRANSLOCATIONS IN *Drosophila melanogaster* MALES TREATED WITH DIFFERENT CONCENTRATIONS OF TOLUENE

Concentrations %	Chromosomes with sex- linked recessive lethals	Chromosomes with translocations II-III	% Frequency
Control	0/480	1/519	0.19
0.10	0/432	0/351	
0.25	0/428	0/353	
0.50	0/376	0/356	
0.75	0/381	0/336	
1.00	0/326	0/316	
1.25	0/231	0/268	
1.50	0/212	0/262	

the induction of gene mutations, but fail to induce chromosome breakage or cause chromosomal aberration except at toxic levels of exposure (Vogel and Leigh 1975).

Auerbach (1976) suggests that the induction of sex-linked recessive lethal mutations is a test so sensitive that it can be used as a biological dosimeter. Nevertheless, some objections have been made, because sex-linked recessive lethals include a heterogenous group of genetic changes ranging from deletions to several types of mutations, and the rate of such events varies among concentrations and mutagens (Vogel and Natarajan 1979).

The fact that the induction of structural aberrations (translocations) required

higher doses (Abrahamson *et al.* 1969) than those employed to point mutations, supports the hypothesis that the probability of interactions of induced events may be different, a concept already expressed by Muller (1941) early in the research with chemical mutagens.

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