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### **Effect of sevin on mating behavior of *Drosophila melanogaster*.**

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### **Abstract**

Behavioral toxicity of systematic pesticide Sevin has been tested in *Drosophila melanogaster* by larval feeding technique. Larvae of same age  $72 \pm 4$  h were grown in the wheat agar media mixed with different levels of Sevin, and the effect of the pesticide was analyzed by observing mating behavior of *Drosophila*. Key words: Sevin, Mating behavior, *Drosophila*.

### **Introduction**

Pesticides are deliberately added to the environment to combat target species. But nontarget species (Bhunja *et al.*,1994; Elliot *et al.*,1996; Green *et al.*,1994; Johan and Prakash.,1997; Krishnan and Ravi,1994; Marton,1974; Oleolzska and Sikorski,1991; Rao *et al.*,1994; Tanaka *et al.*,1996) including man also affected. Carbaryl causes behavioral and neurological problems (Dsi,1974; Anger and Setzer,1979; Branch and Jacqz,1986) in exposed animals. Hence, the present work was intended to understand the mating behavior of *Drosophila melanogaster* under the influence of Sevin.

### **Materials and Methods**

Commonly used systematic pesticide sevin (CAS No.63-25-2) was selected. Local *Drosophila melanogaster* has been tested at  $25 \pm 1^\circ\text{C}$  and RH 60%. The same aged larvae ( $72 \pm 4$  h)

were obtained after mating virgin females with males. 25 larvae per vial were exposed to different doses (1, 2, 3, 4 and 5 mg/100 ml) of Sevin to rest of their developmental time due to feeding is more in larval period (Vogel, 1976). One hundred larvae were allocated to each dose, and flies after emergence were collected immediately and isolated. Males and females were maintained separately on wheat agar media for three days, and their mating behavior (10 pairs per dose) was studied. The significance was tested with control group using simple 't' test.

Table 1. Mating behaviour of treated males with normal females.

↓Concentration (mg)	Behaviour				
	Orientation	Wing vibration	Waving	Licking	Circling
Control	26.92 ± 2.16	2.52 ± 1.02	11.81 ± 1.80	2.42 ± 1.13	0.41 ± 1.62
1	29.30 ± 1.33	2.63 ± 0.18	13.43 ± 2.10	2.80 ± 1.92	0.58 ± 1.28
2	28.32 ± 1.76	2.79 ± 1.90	12.58 ± 1.92	2.93 ± 1.27	0.63 ± 0.82
3	31.16 ± 1.24	3.23 ± 1.08	14.38 ± 0.62	2.62 ± 2.01	0.66 ± 1.34
4	39.21 ± 2.01*	3.01 ± 1.26	26.18 ± 2.18*	3.12 ± 1.88	2.38 ± 0.85*
5	47.36 ± 2.96*	5.31 ± 2.12*	25.70 ± 1.92*	3.68 ± 2.16*	3.24 ± 1.72*

\* p &gt; 0.05

Table 2. Mating behaviour of normal males with treated females.

↓Concentration (mg)	Behaviour				
	Orientation	Wing vibration	Waving	Licking	Circling
Control	26.92 ± 2.16	2.52 ± 1.02	11.81 ± 1.80	2.42 ± 1.13	0.41 ± 1.62
1*	24.30 ± 0.92	2.39 ± 1.80	12.10 ± 0.92	2.12 ± 1.82	0.30 ± 1.24
2*	24.23 ± 0.92	2.68 ± 1.36	13.82 ± 1.02	2.38 ± 1.24	0.38 ± 1.18
3*	25.48 ± 1.82	2.45 ± 1.68	14.60 ± 1.32	2.72 ± 0.90	0.47 ± 1.72
4*	26.72 ± 2.01	2.97 ± 0.28	14.28 ± 1.40	2.64 ± 1.54	0.52 ± 0.84
5*	28.34 ± 1.56	3.68 ± 1.44	14.76 ± 1.12	2.88 ± 1.14	0.49 ± 1.28

\* p &gt; 0.05

Table 3. Mating behaviour of treated males with treated females.

↓Concentration (mg)	Behaviour				
	Orientation	Wing vibration	Waving	Licking	Circling
Control	26.92 ± 2.16	2.52 ± 1.02	11.81 ± 1.80	2.42 ± 1.13	0.41 ± 1.62
1*	27.31 ± 1.82	2.83 ± 2.04	10.64 ± 0.34	2.68 ± 1.20	0.34 ± 1.14
2*	29.64 ± 1.64	2.14 ± 1.92	10.98 ± 1.62	2.56 ± 0.98	0.38 ± 0.84
3*	28.30 ± 0.98	2.36 ± 1.18	11.32 ± 1.14	2.74 ± 1.28	0.40 ± 1.26
4*	32.14 ± 2.14	2.58 ± 0.90	11.56 ± 1.18	2.66 ± 0.48	0.46 ± 1.76
5*	31.34 ± 1.96	2.94 ± 1.34	12.38 ± 0.62	2.88 ± 1.22	0.58 ± 1.32

\* p &gt; 0.05

Table 4. Mating behaviour of normal females with treated males.

↓Concentration (mg)	Behaviour	
	Decamping	Extrusion
Control	1.51 ± 0.32	1.68 ± 0.42
1	1.42 ± 0.18	1.64 ± 0.22
2	1.63 ± 0.24	1.72 ± 0.14
3	1.54 ± 0.82	1.60 ± 0.94
4	2.03 ± 0.28*	1.78 ± 0.14
5	3.94 ± 0.36*	2.94 ± 0.28*

\* p &lt; 0.05

Table 5. Mating behaviour of treated females with normal males.

↓Concentration (mg)	Behaviour	
	Decamping	Extrusion
Control	1.51 ± 0.32	1.68 ± 0.42
1*	1.52 ± 0.33	1.72 ± 0.44
2*	1.48 ± 0.22	1.70 ± 0.82
3*	1.61 ± 0.82	1.65 ± 0.22
4*	1.72 ± 0.92	1.68 ± 0.34
5*	1.96 ± 0.12	1.62 ± 0.48

\* p &lt; 0.05

Table 6. Mating behaviour of treated females with treated males.

↓Concentration (mg)	Behaviour	
	Decamping	Extrusion
Control	1.51 ± 0.32	1.68 ± 0.42
1*	1.56 ± 0.24	1.82 ± 0.56
2*	1.58 ± 0.88	1.73 ± 0.42
3*	1.49 ± 0.28	1.73 ± 0.82
4*	1.44 ± 0.33	1.64 ± 0.20
5*	1.52 ± 0.44	1.78 ± 0.44

\* p &lt; 0.05

## Discussion

From the results it is confirmed that when *Drosophila* flies (both male and female) treated with 4 and 5 mg of Sevin showed significant changes in their mating behavior. In *Drosophila* mating behavior is always started by males (Speith, 1952; Patterson, 1980). Significant changes of orientation, vibration, waving, licking, and circling by males indicate that there is an effect of Sevin. These prolonged activities are only to attract opposite sex to mount. Similarly females showed significant decamping and extrusion behavior towards males developed on 4 and 5 mg Sevin media, indicating non-readiness to accept and mate. The normal males, however, were accepted by the treated females of all doses. When mating between treated males and treated females is evaluated, no significant variation occurred in *Drosophila* mating behavior is a prerequisite for copulation (Speiss, 1970) and is a fitness character deciding the success of the organism (Fulker, 1966). Any changes in these behaviors have direct effects on the copulation and propagation of life.

## Results

The results of mating behavior of treated males with normal females is shown in Table 1. The orientation ( $39.21 \pm 2.01$ ,  $47.36 \pm 2.96$ ), waving ( $26.18 \pm 2.18$ ,  $25.70 \pm 1.92$ ) and circling ( $2.38 \pm 0.85$ ,  $3.24 \pm 1.72$ ) of males treated with 4 and 5 mg of Sevin are significantly more compared to control. The vibration ( $5.31 \pm 2.12$ ) and licking ( $3.68 \pm 2.16$ ) behavior of males from 5 mg Sevin treated group is significant to their control. The data of mating behavior of normal males with treated females is represented in Table 2, and there is no significant difference to the control group. The mating behavior values of treated males with treated females are shown in Table 3. All the values are insignificant to control values.

Similarly, the values for female mating behavior, that is, decamping and extrusion, are depicted in Tables 4, 5, and 6. Decamping of female is significant ( $2.08 \pm 0.28$ ), ( $3.94 \pm 0.36$ ), and extrusion is significant ( $2.94 \pm 0.28$ ) when allowed to mate with 4 and 5 mg Sevin treated males. The values obtained after mating of treated females with normal males and treated females with treated males are, however, insignificant to their control groups.

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### Expression of the *cbut-RB* isoform during embryonic development in *Drosophila melanogaster*.

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## Introduction

*cbut* (*cbt*) is a *Drosophila* gene encoding a transcription factor, which is involved in several developmental processes such as embryonic dorsal closure (Muñoz-Descalzo *et al.*, 2005) and epithelial regeneration (Blanco *et al.*, 2010), ecdysone response (Beckstead *et al.*, 2005), neuroendocrine cell remodeling (Zhao *et al.*, 2008), circadian rhythm (Kadener *et al.*, 2007), axon guidance and synaptogenesis (Kraut *et al.*, 2001; Mindorff *et al.*, 2005), pole cell formation (Yatsu *et al.*, 2008), cell growth (Guertin *et al.*, 2006), autophagic cell death (Gorski *et al.*, 2003), and cell cycle (A.J. Katzaroff and B.A. Edgar, personal communication). The *cbt* gene contains two exons separated by an intron, and two different mRNA products have been associated to this gene, *cbt-RA* and *cbt-RB* (Figure 1A). *cbt-RA* and *cbt-RB* mRNAs encode two Cbt protein isoforms, Cbt-RA (428 amino acids) and Cbt-RB (347 amino acids), respectively, both containing a serine-rich region at the amino terminus and three classical zinc finger domains C<sub>2</sub>H<sub>2</sub>-type at the carboxy terminal region. The Cbt-RB isoform, however, lacks 81 residues in the amino terminal region. It has been recently shown that *cbt* mRNAs are maternally contributed, since they are present in unfertilized and early embryos (Yatsu *et al.*, 2008; Belacortu *et al.*, 2010). During germ band retraction and dorsal closure, this gene is mainly expressed in epidermal cells, yolk nuclei, amnioserosa, hindgut, and anal pads (Muñoz-Descalzo *et al.*, 2005; Belacortu *et al.*, 2010). Although previous analyses suggested that the *cbt-RB* isoform is probably an artifact, expression analyses in microarrays (Arbeitman *et al.*, 2002) as well as the presence of new expressed sequence tags (EST) in *Drosophila* cDNA libraries from S2 cells and embryos indicate that *cbt-RB* mRNAs are probably expressed. To determine whether *cbt-RB* transcripts are indeed present in *Drosophila* embryos, different experiments have