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ACETYLCHOLINESTERASE IN NORMAL AND DDT-INTOXICATED BEES

Adelina Derevic<sup>1</sup> and Victoria Nitu<sup>2</sup>

Bodgan P. Hajden 37, Bucarest, Romania

A B S T R A C T

The effect of DDT intoxication on AChE activity in bees was investigated. In bees collected in May the enzyme was found to be more active after exposure to residual contact with DDT, but showed reduced activity after oral ingestion. Those intoxicated by contact showed marked dehydration.

The use of pesticides in agriculture has become a necessity, however their harmful effects on beneficial insects and especially honey bees should be reduced. Metcalf (1955) found that acetylcholinesterase (AChE) of honey bees differs from that of house flies by being inhibited quite differently by certain organophosphorus (O-P) compounds. Such differences could open the way for finding insecticides acting selectively only on harmful insects and not on bees. This paper describes the level of AChE in DDT-intoxicated bees.

MATERIALS AND METHODS

The 3 500 bees used in the tests had not been previously exposed to insecticides. The following two groups of bees were examined: (a) collected in December, and (b) in May. DDT was applied either by feeding the bees with honey syrup containing DDT at a level of 10 µg of DDT/bee, or by exposing the bees to a DDT residual film of 200 mg DDT in a 200 ml BerceTius beaker. AChE was determined in the supernatant of bee's head and thorax which had been -triturated with sand in a buffered solution by the method of Hestrin (1949), modified by Huerta et al (1952). The enzymatic activity is expressed in this method in unit/ml, corresponding to the concentration of enzymes which hydrolyse 1 micromole of acetylcholine at 37° C during 60 minutes. The results were then computed per bee.

<sup>1</sup>Prof. Agregée

<sup>2</sup>Lecturer I. Med. Farm. Bucarest.

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## RESULTS

### A. Symptoms of intoxication

- (a) By ingestion. 1) an asymptomatic latency phase was first seen, which lasted from 12 to 14 hours; 2) then came excitation phase, followed by, 3) the onset of the critical phase with the appearance of shivering and falling down from the cage walls; 4) latent immovable phase and, 5) finally a 100% mortality after 6 days. The bees obtained in May showed only symptoms 1 and 2 and after 6 days their mortality was only 12%.
- (b) By contact (December bees). During the initial phase a marked agitation was observed, the first symptoms appearing about 30 minutes after the contact with the DDT crystals was made. Sixty minutes later the bees gathered at the upper part of the glass, under the gauze, where ventilation was better. Their movements became uncoordinated with shivering and continuous agitation; 100% death occurred 3 to 8 hours later.

Bees intoxicated by contact in May showed a longer asymptomatic phase with a progress of gradual agitation, accompanied by respiratory disorders;

The bees showed uncoordinated movements, but without shiverings. Mortality in this group was only 25%. Marked dehydration occurred especially in bees intoxicated by contact.

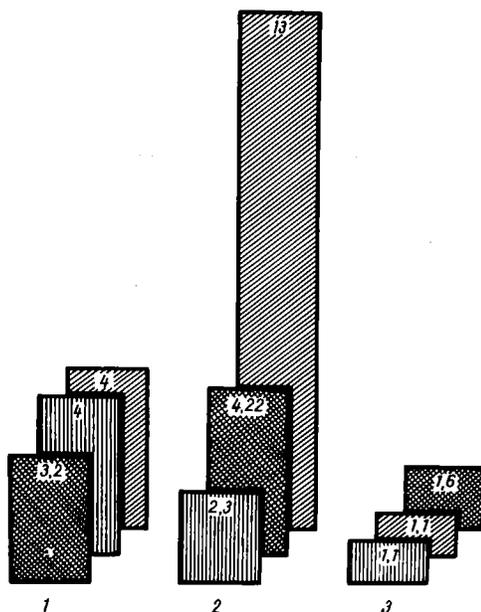
### B. AChE activity

In bees obtained in December the results were as follows: in the control bees 3.2 enzyme units per bee, in the intoxicated 4.0 units. The enzymatic activity of bees collected in May was only a little higher than of the previous group (4.22 units/bee). However the difference appeared in the bees intoxicated by contact (13 units), which showed a marked increase compared to the December group. In bees intoxicated per os, the activity of the enzyme was more reduced (2.3 units/bee). In the controls kept at 10°C during 60 minutes before the determination of the enzyme, the lowest values were found, 1.6 units versus 1.1 units/bee in the two other groups (fig. 1).

## DISCUSSION

Sternburg (1963) considers that during the DDT intoxication a substance resembling a toxin, the product of an excessive activity of the nervous tissue is eliminated. During DDT-intoxication different physiological systems are disturbed. Petuchov (1969) found a relationship between the intoxication symptoms with O-P compounds in bees and the reduced activity of AChE as follows: in the asymptomatic phase a reduction by 22%, in the excitation phase by 49%, in the paralytic one by 85% and during the agony phase 94%. Our results also show an unsteady state of the AChE system. This is illustrated by the different level of the enzyme, as compared to that of normal bees, as well as by the symptoms and mortality. While interpreting the results one must bear in mind that December bees were in a semi-hibernation state and older while those of May were young and very active.

Bees collected in May showed a lack of relationship in the activity of the enzyme between groups intoxicated through different ways. In those intoxicated by contact the activity increased by about 3 times, while those intoxicated per os showed a reduction by about 50% (Fig. 1). The almost identical values for the December bees might be attributed to their physiological state.



LEGEND OF THE FIGURE

Fig. 1. Activity of cholinesterase in normal and DDT-intoxicated bees (units per bee)

1 = dead bees (December);

2 = living bees (May)

3 = frozen bees (May)

net = normal bees

perpendicular lines = per os intoxicated bees

oblique lines = contact intoxicated bees

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