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TOXIC EFFECTS OF DIFLUBENZURON ON PUPAL-ADULT DEVELOPMENT OF *Corcyra cephalonica* STAINTON

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Diflubenzuran and its related compounds have been found to affect moulting of insects by interfering with deposition of new cuticle. The topical application of the ED_{50} was 0.265 µg/prepupa of susceptible 'NAIDH' housefly. Method of testing and solvent used also influences the toxicity of a compound.

In the toxicity studies of diflubenzuron, different solvents like tetrahydrofuran (Cerf and Georghiou, 1974), acetone (Ascher and Nemny, 1976) and dimethylsulfoxide (Chang and Borkovec, 1980) were utilised. Many research workers did not give enough emphasis for finding out the accurate toxicity of diflubenzuron by injecting into the insect. So experiments were conducted to find out (i) the accurate toxicity of diflubenzuron by injecting into the pupae of *C. cephalonica* simultaneously identifying a safe solvent of diflubenzuron with little interference to the toxicity and (ii) effects of diflubenzuron on pupae of *C. cephalonica*.

MATERIALS AND METHODS

The pupae of *C. cephalonica* (Pyralidae: Lepidoptera) were obtained from a laboratory colony maintained on *Sorghum bicolor* (L) Moench, grains. Technical grade diflubenzuron was a gift from Dr. A. B. Borkovec, Insect chemosterilants laboratory, USDA, Maryland, USA. Pupae were injected with the help of an alcohol sterilized 27 gauge needle attached to 1 ml tuberculin syringe using an electrically operated micro-applicator. The syringe was calibrated by using clean mercury. The amount of mercury delivered through the needle for different time intervals viz., 0.5, 1.0, 1.5, 2.0 and 2.5 seconds was collected in pre-weighed aluminium foil boats by operating the micro-applicator for different time of periods. The weight of mercury delivered per unit time was noted. This opertion was repeated six times and the mean weight of mercury was taken to calculate the delivery in volume. The volume was calculated by using the formula: volume $= \frac{\text{Mass}}{\text{Density}}$. The syringe delivered 0.9µ1 in one second.

The material was injected to pupae below the wing pads, keeping the needle parallel to the body line. To avoid any damage to these small pupae while handling, the pupae were held in the grooves of the small corrugated cardboard piece. The fluid was injected and the pupae allowed to stay as such for 15-20 seconds so that the injected material got thoroughly mixed with haemolymph. The pupae were removed from the needle with the help of a forceps. Preliminary experiments with

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Table 1 : Percentage mortality of *C. cephalonica* pupae at different doses of Diflubenzuron

S. No.	Dose μ g/pupa	Per cent mortality
1	0.1	86.7
2	0.075	73.3
3	0.05	56.7
4	0.025	43.3
5	0.01	26.7
6	Contr	16.7

saline indicated that the amount of fluid that could be safely injected to each pupa without injury or loss of fluid was found to be 0.25, 1/pupa.

To find out a safe solvent of diflubenzuron with least toxicity to pupae of *C. cephalonica* different solvents viz., acetone, dioxan, dimethylformamide (DMFA) and dimethylsulfoxide were injected to 0-day pupae and they were allowed to develop as adults. Among the solvents tested, DMFA recorded high adult emergence of 43.3 % followed by dioxan (27.0%) dimethylsulfoxide (23.3%) and acetone (0.0%) DMFA which had maximum adult emergence was further tested by diluting it with ethyl alcohol or methyl alcohol at 1:1 or 2:3 (v/v) ratio. Dilution of DMFA with methyl alcohol at 2:3 ratio further enhanced the adult emergence to a level of 80 percent indicating the least self-toxicity to the insect. DMFA with methyl alcohol at 2:3 (v/v) ratio was selected as a safe solvent.

To find out the toxicity, different doses viz., 0.01, 0.025, 0.05, 0.075 and 0.1 μ g of diflubenzuron per pupa were injected to 0-day pupae. DMFA : methyl alcohol at 2:3 ratio was injected to control pupae. The experiment was carried out with 10 pupae in each treatment replicated thrice. Mortality counts were recorded after emergence of adults in control. ED_{50} value was computed by Probit Analysis (Finney, 1981) after making correction for natural mortality (Abbott, 1925)

RESULTS AND DISCUSSION

There was no pronounced effect of diflubenzuron during pupal growth of *C. cephalonica*. The toxic effects manifested only after 6 days ie., at the time of pupal - adult transformation (Fig. 1).

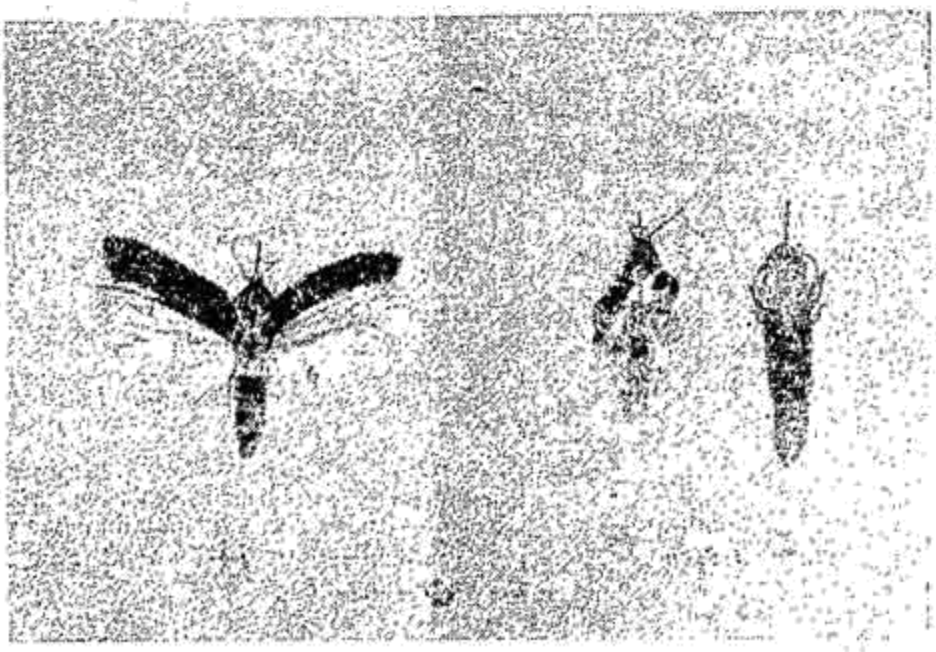

Fig. 1.

A. Normal adult.

B. Wing deformation

C. Eclosion prevention

Similar results were also reported in *Spodoptera litura* (Natesan and Balasubramanian, 1980), *Nephantis serinopa* (Sundaramurthy and Santhanakrishnan, 1979), *Tenebrio molitor* (Soltani *et. al.* 1984). Diflubenzuron is characteriszed by its delayed effects (Mulder and Gijswijt, 1973.) Diflubenzuron treated fresh pupae of *C. cephalonica* exhibited toxic effects only at the time moulting. Manifestation of diflubenzuron toxicity at the time of moulting suggests the involvement of endocrine system. Increase in ecdysone by diflubenzuron treatment was reported by Yu and Terriere (1975 and 1977) in house flies *Musca domestica*, flesh flies *soroophaga bullata* and blow flies *Phrmia regina:* Redfern *et al* (1982) in *Oncopeltus fasciatus* nymphs. On the other hand increase in JH by diflubenzuron treatment was suggested by Subrahmanyam *et al* (1980) in castor semilooper *Achoea janata* Further Ishaaya and Casida (1974) reported increased chitinase and phenol oxidase activity in diflubenzuron treated houseflies.

The criteria of mortality taken into consideration was either deformity in pupal-adult transformation or death of pupa (Table 1). The regression equation of probit analysis was calculated to be $Y = 2.21 x + 1.403$, where Y was probit mortality and x log of amount of diflubenzuron injected. ED_{50} calculated was observed to be 0.043/ μg/pupa with fiducial limits of 0.032-0.056 whereas LD_{50} calculated by taking mean weight of pupa as 30 mg 1.43μ g/g body weight.

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