Original Article

SCANNING ELECTRON MICROSCOPIC CHANGES IN THE SEMINAL VESICLE OF THE ADULT MALE INSECT Odontopus varicornis (HETEROPTERA: PYRRHOCORIDAE) IN RELATION TO REPRODUCTION

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Abstract

Pest control massive use of synthetic pesticides has imposed so many detrimental effects on the environment and cause intoxication of non-targeting organisms. An attractive alternative solution to this problem is to employ natural products for control in agroecosystem, in the form of a preventive strategy against pest attack on agricultural products. Hence, the present study aimed to find out the scanning electron microscopic (SEM) changes in the seminal vesicle of the male insect Odontopus varicornis treated with sublethal concentration (LD_{50}, 0.15% ) of Piperidine for 48h. The present results revealed that characteristic SEM changes in the seminal vesicle treated insect than control insect, such as shrunken with thick and prominent myoethelial cells and also thickened and highly pycnotic than control. Disintegration of secretory vesicles and secretory substances were observed in the present study.

INTRODUCTION

Many plant species are known to possess biological activity that is frequently assigned to the secondary metabolites. Among these, essential oils and their constituents have received considerable attention in the search for new biopesticides. Many of them have been found to possess an array of properties, including insecticidal activity, repellency, feeding deterrence, reproduction retardation and insect growth regulation against various insect species (Cheng, et al., 2004 and Traboulsi, 2005). One of the most important and challenging aspects in the pesticidal research is the development of new and effective approaches for controlling various insect pests (Chang, 2001 and Ficker, et al., 2003). Synthetic pesticides have been effectively used to control insects and plant diseases; however, increasing concern over their environmental effects has highlighted the need for the development of an alternative types of selective control or methods for crop protection with/ without reduced use of conventional pesticides (Hayes and Laws, 1991).

The pesticide is applied to a compound for its action on repelling, attracting and sterilizing action on insects. The poisons affect the normal functions of specific cells and tissues of insects, just as they are known to do in human and other higher animals. Basically, some chemical process in the animal affected so as to bring about changes in its function. These changes are secondary to the original process that was affected and are frequently mistaken for the initial action of the poison (Vasantharaj David and Kumaraswami, 1982). In male insects, the reproductive organs are visibly either as a direct effect or as an indirect effect of chemicals. Induced sterility in treated males may be due to complete cessation of spermatogenesis resulting in the loss of fertility due to the presence of dominant lethals (Balakrishnan, 1990 and Ramanathan, 1995).

Insect, the male reproductive system consists of a pair of testicles connected by ducts to a gonopore. In many groups, including in Hymenoptera, there are regions along the ducts such as the seminal vesicles and the secretory accessory glands, where spermatozoa are stored until mating (Vinicius Albano Araujo et al., 2005). Some insects, such as stingless bees (Kerr, 1948; Ferreira, 1966; Colleto, 2000) and sand flies, have no accessory glands, while in others, the epithelial cells of the seminal vesicle or of the ejaculatory duct have the organelles typically associated with a secretory function.

The products, to these epithelial cells have the same function as those of the accessory glands (Cantacuzene, 1972; Riemann and Thomson 1976; Ferreira, 1977; Couche and Gillott, 1988; Cruz-Landim and Fausto et al., 2000). In other insects, the seminal vesicle apparently produces no secretion (Odhiambo,1969; Cruz-Landim and Dallacqua,
The seminal vesicle of *Odontopus varicornis* appears to be white oval shaped body (Selvisabhanayakam, 1995). Therefore, it has been programmed in the present study to find out the effect of phytopesticide Piperidine on the seminal vesicle of adult male insect, *Odontopus varicornis*.

**MATERIALS AND METHODS**

**Insects**

The insects collected from the fields and gardens were reared in wooden cage, each measuring about 30 × 22 × 28 cm at the laboratory temperature of 29 ± 2°C and relative humidity of 80 ± 5%. The insects were fed daily with soaked cotton seeds (*Bombax ceiba*) as well as with seeds of its higher plant, *Stericula foetida* and *Gossypium* sp. An additional food of the pieces of chow-chow (*Sechium edule*) was also given to these insects. The insect's cage was cleaned properly, every alternative day, by removing the excreta and other waste materials. The egg laid by them was transferred to another cage, and thus a continuous culture was maintained.

**Piperidine**

Piperidine was purchased from Aldrich chemical company (Molecular formula: C_5H_11N, Density: 0.8629 g/mol, Molecular wt: 85.15 g/mol, Bolling point: 106°C).

**Scanning electron microscopic study**

For scanning Electron Microscopy (SEM), the seminal vesicle of anesthetized specimens were removed and fixed in Karnovsky solution (2% paraformaldehyde, and 2.5% glutaraldehyde in 0.2 M sodium cacodylate buffer). The tissues were then dehydrated in a graded ethanol series (70% - 100%) and in 100% acetone (1:1) solution, followed by four washes in 100% acetone. After drying, the samples were assembled on aluminium stubs, coated with gold examined and photographed with Joel JSM-P15 Scanning Electron Microscope (Wood Ward, 1972).

**RESULTS AND DISCUSSION**

The male reproductive system of *Odontopus varicornis* consist of a pair of testes, seminal vesicle, vasa deferentia, a common ejaculatory duct, an erection fluid reservoir or bulb like ampulla, a pair of short oval shaped accessory glands opening into the reservoir (ampulla) and an aedeagus. The morphology of testis were whitish organs lying symmetrically in the cavity between second and third abdominal segments. The testes were composed of eight spindle shaped testicular follicles. The entire testis was surrounded by a connective tissue which holds the testicular follicles together and was richly supplied with tracheoles and fat body (Fig. 1).

The seminal vesicle of the control insects exhibited tubular structure with myoepithelial cells. The tubules were externally covered with thick folded cuticle. The epithelial cells were also associated with minute secretory vesicle and secretory globules. The epithelial layer was consisted of numerous pinocytotic pits and microvilli also proves the epithelium to enhance it’s secretory activity. The lumen contained secretory substances and numerous secretory vesicles (Fig. 2). The effects of phytopesticides, Piperidine showed considerable changes in the scanning electron micrograph such as an entire tubule was appeared to be shrunken with thick and prominent myoepithelial cells than the control insect. The rope like structure of the plasma membrane was found to be highly pycnotic and disintegrated with columnar epithelial cells. The muscular layer also thickened and highly pycnotic. Disintegration of secretory vesicles and secretory globules were observed (Fig. 3). Hence, this gland was not enhanced in the synthetic and secretory activity. Pinocytotic pits were not observed in the treated insects, suggested that the mechanism of secretion may became impaired in the treated insects (Fig. 4).

**Fig. 1.** Anatomy of the male reproductive system of *Odontopus varicornis*

![Fig. 1. Anatomy of the male reproductive system of *Odontopus varicornis*](image)

**Fig. 2.** Scanning electron micrograph showing the seminal vesicle of control insect x 1000

![Fig. 2. Scanning electron micrograph showing the seminal vesicle of control insect x 1000](image)
In the present study, the pesticide treated insects showed considerable change was appeared to be shrunken with thick and prominent myoepithelial cells than the control insect. Plasma membrane was found to be highly pycnotic and disintegrated with columnar epithelial cells. The muscular layer also thickened and highly pycnotic. Secretory vesicles and secretory globules were not found in the gland, not enhanced in the synthetic and secretory activity. Pinocytotic pits were not observed in the treated insects, suggested that the mechanism of secretion may became impaired in the treated insects. The SEM changes in the seminal vesicles of treated insects have shown numerous folded cuticle and the epithelial cells were not distinct. The secretory globules were found to be less which may be attributed due to less amount of secretory substances. These changes might be caused the reproductive physiology of Odontopus varicornis. Similar results have been also reported by (Sumathi,2002) exposed to endosulfan for Gryllotalpa africana, (Lousia, 2010) has exposed to pygidial secretion for Odontopus varicornis, (Vivekananthan, 2011) has exposed to vijay neem for Mylabris indica, respectively. In the present study, it has been observed that the reduction in the size, structure of seminal vesicle were attributed due to the Piperidine (phytopesticide) that brought about reproductive disturbances and sterility in the test insect, Odontopus varicornis than the control insect.

It is evident from the present study that the SEM changes in the seminal vesicle showed shrunken tubule with thick myoepithelial cells, the rope like structure of the plasma membrane, which was observed to be highly pycnotic and disintegrated of secretory vesicles and secretory globules were observed in treated insects for 48hours. Further, the drastic changes which alter the function and motility of sperms which intern affects the process of reproduction.

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