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Morphological changes induced by thermal treatment and gamma irradiation on the males' hind legs of *Spodoptera littoralis* (Noctuidae; Lepidoptera)

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ABSTRACT

External morphology of males' hind legs of *Spodoptera littoralis* subjected to thermal treatment (33 °C and 37 °C) or/and irradiated with substerilizing doses of gamma radiation (75, 100 and 150 Gy) were studied using scanning electron microscopy (SEM) in the parental generation. Five types of sensilla have been distinguished; three types of trichoid sensilla (T_1,T_2 and T_3), sensilla basiconica and sensilla auricillica, which are considered as olfactory chemoreceptors. Moreover, sensilla chaetica are contact chemoreceptors, whereas sensilla styloconica are thermo–hygro/gustatory mechanoreceptors. The impact of thermal treatment or/and gamma irradiation reflect a clear morphological change in *S. littoralis* legs'-sensilla, claws, spurs and scales. Otherwise, the degree of deformity was thermal and dose dependent, as it increased with an increase of the degree of temperature and dose of irradiation applied. Substerilizing doses 75 and 100 Gy, either alone or combined with thermal treatment 33 °C, have low undesirable effects on the hind legs with successful mobility or courtship behavior. Consequently, synergistic effect of gamma radiation and thermal stress induced successful application in the integrated pest management program for controlling *S. littoralis*.

1. Introduction

The cotton leaf worm, *Spodoptera littoralis* (Boisd.), is a serious pest attacking field and many other crops in Egypt. The insect mobility arises from the power of flight or walking with the three pairs of legs, in order to mate and feed. The insect legs have several types of senseilla which is distributed on them and plays a very important role in the insect behavior.

These receptors are the main tools of insect chemical communications. Receptors are tune feeding preferences (De Boer, 2006). They, also detect host plant odors (Skiri, Stranden, Sandoz, Menzel, & Mustaparta, 2005) and play important roles in insect survival and environmental

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adaptation. Multiple receptors offer many functional advantages to the insects' ability to perceive and respond to different environmental signals by facilitating the detection of different stimuli (Debry & Steullet, 2001). These receptors on legs may be in the form of contact chemoreceptor or mechanoreceptors, which play an important role in the final recognition of host plant (Maher, 2002; Maher, Jolivet, & Thiery, 2001), oviposition site or mating (Ramaswamy, 1988; Städler, 2002).

All sensilla located on the legs appeared to be mechanoreceptors for providing information during waking or other behavior. The male pretatarsal setae usually help to hold the female during mating. (Kim & Yamasaki, 1996).

Several research papers have provided extensive discussion on the general morphology and function of the sensory receptors of Lepidoptera legs such as; Salama, Sharaby, and Abdel-Aziz (1987) on *Heliothis armigera*, Faucheux (1991) on *Homoeosoma nebulella*, Haiba (1998) on *Puthorimaea operculella* and Hazaa, El-Shall, & Alm El-Din, 2006; Hazaa, El-Degwi, & Alm El-Din, 2012 on *S. littoralis*.

Consequently, sensilla are considered the main communication system in insects between the individuals and their external environment. So, sensilla can play an important role in insects' control. The present study aimed to investigate the thermal treatment 33 °C and 37 °C, either alone or in combination with substerilizing gamma radiation dose 75,100 and 150 Gy on the morphology of hind legs receptors of male *S. littoralis* resulting from irradiated full grown pupae by using scanning electron microscopy (SEM). Accordingly, the present study was undertaken to investigate the synergistic effect of heat stress and gamma radiation for providing information necessary for pest management.

2. Materials and methods

2.1. Rearing and maintenance of insects

The culture of the cotton leaf worm, *S. littoralis* was maintained continuously in the insectary of Atomic Energy Authority at Inshas, Egypt. Larvae were reared on fresh castor-oil plant leaves *Ricinus communies* under laboratory controlled conditions of 27 ± 2 °C and 65 ± 5% R.H.

2.2. Thermal treatment

Full grown male pupae were subjected to thermal treatment in two incubator adjusted at 33 °C and 37 °C for 20 h. Similar numbers were kept as control at 25 ± 2 °C. After thermal treatment, pupae were transferred to the laboratory conditions and kept till the adult parent males' emergence.

2.3. Irradiation process

Full grown male pupae were gamma irradiated with 75, 100 and 150 Gy using a Co^{60} gamma irradiation cell located at Nuclear Research Center in Inshas, with a dose rate of 1 Gy/sec. Untreated pupae were kept as control for comparison. The emerged adult males' moths were caged till they were used.

2.4. Association of gamma irradiation and thermal treatment

Groups of full grown male pupae were exposed to thermal treatment 33 °C for 20 h in an incubator and then were irradiated by dose levels; 75,100 and 150 Gy. The emerged parental generation adult moths were caged till they were used.

2.5. Scanning electron microscopy (SEM)

For examination with scanning electron microscope (SEM) the whole hind legs of freshly killed males resulted from (thermal treatment, gamma irradiation and combination treatment) specimens were removed from the head, cleaned by gentle sonification in distilled water and 70% ethanol for 1 min, attached vertically to aluminum stubs and air-dried before being coated with gold in a vacuum evaporator. Sensilla observation and description were made by using (Joel JSM 5400) scanning electron microscope with an accelerating voltage of 10 KV.

3. Results

Spodoptera littoralis has three pairs of legs, one pair on each of the thoracic segments. The kinds of legs in *S. littoralis* are ambulatory legs used for walking. Each leg typically consists of six basic segments, articulating with each other and these are: coxa, trochanter, femur, tibia, tarsus and pretarsus. The tarsus is subdivided into five tarsomeres differentiated from true segments by the absence of muscles. The pre-tarsus consists of a membranous base supporting a median lobe, the arolium, which may be membranous, or partly sclerotized and a pair of claws which articulated with a median process of the last tarsomere. These claws are strong with a broad base and a tough sharp end. The development of the claws is variable; they are more or less equally well developed (Fig. 1A and B). The pre-tarsus also ends with two basal spines (Fig. 1A). At the hind leg, the tibia has two pairs of sharp spurs, one in the middle part of it and the other on the end (Fig. 1E and F). The pair of spurs has one long spur and one short spur. The morphological examination of insect legs by Scanning Electron Microscope showed the presence of five different types of sensillae, three subtypes of sensilla trichoid (T_1, T_2 and T_3), chaetica, styloconica, basiconica, and auricillica.

3.1. The sensilla detected are as follows

3.1.1. Sensilla trichoid (T)

Sensilla trichodea are usually thin with variable lengths and a rather sharply pointed tip articulated with the body wall by a membranous socket so that it would be free to move. It is the most numerous types of sensilla present in the tarsus segment. Three distinct subtypes of sensilla trichoid can be distinguished in the male leg according to the length, curvature and tips. Trichoid 1 (T_1) is the most numerous and largest type, relatively straight, but hooked at the tip. Trichoid 2 (T_2) is shorter and more curved than T_1. Trichoid 3 (T_3) lies among trichoid sensilla are curved with a blunt tip and is the shortest one among the three types. (Fig. 1A).