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BIOMETRIC DISCRIMINATION OF THE LARVAL INSTARS AND SEXES OF *MALADERA MATRIDA* (COLEOPTERA: SCARABAEIDAE: MELOLONTHINAE)

D. BEN-YAKIR', L. KOREN' AND A.R. HARARI[2]

[1]*Department of Entomology, Institute of Plant Protection, ARO, The Volcani Center, P.O. Box 6, Bet Dagan 50250, Israel*

[2]*Department of Entomology, The Hebrew University of Jerusalem, Faculty of Agriculture, P.O. Box 12, Rehovot 76100, Israel*

ABSTRACT

Maladera matrida Argaman was first reported from Israel in the late 1970's and was subsequently described as a new species. Shortly thereafter, this species has become a serious pest of ornamental plants and agricultural crops. In this study we identified external anatomical features for distinguishing between the three larval instars and between the two sexes of pupae and adults collected in the field. Larval instars may be divided as follows: first instar, head width less than 1.8 mm; second instar, head width above 1.9 mm and ratio of body length to head width below 3; and third instar, body length above 9 mm. Male pupae may easily be recognized by an ampulla covering the developing genitalia on the posteroventral abdominal segments. Males usually had longer antennal laminae (mean = 823 |iin, range 750-900 |im) than females (mean = 704 |J.m, range of 570-790 (xm); however, in about 10% of the beetles this length overlapped between the sexes (range 750-790 |i.m).

KEY WORDS: *Maladera matrida,* morphology, biometrics.

INTRODUCTION

Maladera matrida Argaman was first reported from Israel in the late 1970's and was subsequently described as a new species (Argaman, 1986,1990). The adult beetles are feeding on foliage and flowers, while the larvae ("white grubs") feed in the soil on roots, tubers and ground nuts (Gol'berg et al., 1989). In the late 1980's, peanut growers in the northwestern Negev region suffered significant economic losses due to infestation with this pest. As a result, intensive biological studies of *M. matrida* were undertaken in that region. A prerequisite for these studies was acquiring the ability to distinguish between the larval instars and determine the sex of live, field-collected insects at various stages of development.

Most scarabaeids have three larval instars, which differ from each other mainly by size (Richter, 1958). The late third-instar larva (prepupa) ceases feeding, ejects the accumulated excrement, and its body color changes to yellowish white (Hallock, 1932). In some members of the subfamily Melolonthinae, males can be recognized at the larval stage by the presence of terminal ampullae, and at the pupal stage by the three-lobed ampullaceous protuberance covering the developing genitalia on the posteroventral abdominal segments (Fleming, 1972;

Tashiro, 1987). Determining the sex of the adult Japanese beetle, *Popillia japonica* Newman, is based on slight differences in the shape of the tibia and tarsus of the prothoracic legs (Fleming, 1972). The last three antennal segments of *M. matrida* are expanded, as in other scarabaeid beetles, into bladelike structures named laminae (Fig. 1). In *Maladera holosericea* Scop. and *Serica brunea* L., which are closely related to *M. matrida*, the antennal laminae of males are longer than those of females (Balachowsky, 1962). Ascher et al. (1990) reported that males of *M. matrida* also have longer antennal laminae than females, and they used this character to determine the sex of cold-immobilized or CO_2-anesthetized beetles. In his original description of *M. matrida*, Argaman (1986) reported that the tip of the abdomen was rounded in males and pointed in females.

The aim of this study was to identify morphological features of *M. matrida* which can be used to distinguish between the three larval instars and to identify the sex of live specimens.

MATERIALS AND METHODS

Insects were collected in peanut fields in the northwestern Negev region during the summer of 1990 and 1991. Grubs and pupae were collected from the top 30 cm of soil, while adults were collected by black-light traps. Specimens were kept in moist sandy soil at room temperature and examined within 24 h after collection. When live insects were measured, they were either cold-immobilized or CO_2-anesthetized.

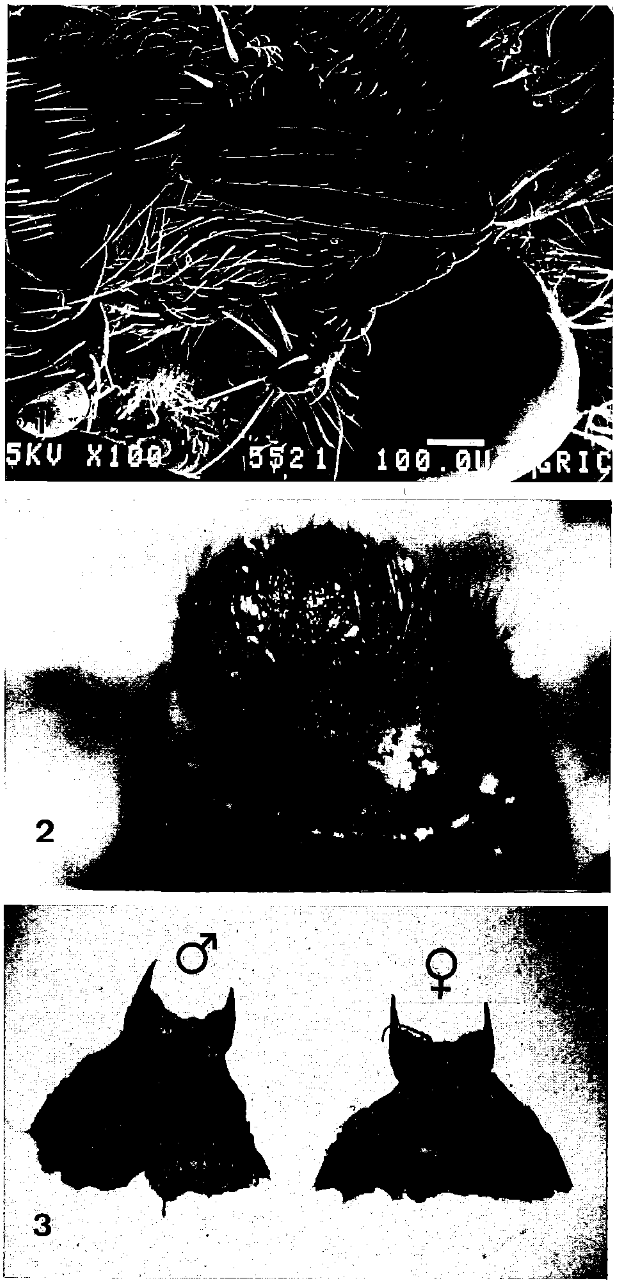
Larval instar discrimination

Grubs of *M. matrida* were found to have on their posteroventral abdominal segments a rasteral pattern (a complex of definitely arranged bare surfaces, hairs and spines) with a single transverse curved row of spines (= palidia; Fig. 2). This rasteral pattern is similar to that of the Asiatic garden beetle, *Maladera (= Autoserica) castanea* (Arrow) (Hallock, 1932), a species that does not occur in Israel, and differs from that of white grubs of other genera (Tashiro, 1987). Therefore, the rasteral patterns of all collected grubs were examined to avoid confusion with those belonging to other genera. We studied 120 grubs, of various sizes. The body length was measured with a flexible tape-measure to an accuracy of 0.5 mm. The head-capsule width was measured with the aid of a stereoscope equipped with an ocular micrometer to an accuracy of 0.1 mm. For comparison, the same morphological characters were measured on 100 laboratory-reared grubs of each instar whose stage was determined by closely following their development. Larvae at the prepupal stage were excluded because they can easily be distinguished from other larval stages by the change of color.

Secondary sexual characteristics

Larvae and pupae were examined for external morphological differences that may indicate their sex (Fig. 3). When differences were found, insects were reared to maturity and their sex was determined by dissection.

Three males and three females were examined for external anatomical differences that may indicate their sex with a scanning electron microscope (model JSM 35C, JEOL Ltd., Tokyo, Japan). Prior to the examination, beetles were dried and coated with a layer of carbon (200 Å) followed by a layer of gold (150 Å). The length of the antennal laminae of 100 females and 100 males was measured with the aid of a stereoscope to an accuracy of 0.02 mm. In order to achieve



Figs. 1–3. *Maladera matrida*. 1. Male antenna (×100). 2. Third-instar grub; rasteral pattern on the posteroventral abdominal segments (×30). 3. Pupae; posterior ventral abdominal segments of male and female (×10).

accurate measurements, antennae were removed and glued flat on a double-sided adhesive tape. The body weight of 100 beetles was determined to an accuracy of 1 mg and then their sex was determined as above.

RESULTS AND DISCUSSION

Larval instar discrimination

Three peaks were noted in the distribution of the grubs' head widths (Fig. 4). The group of grubs at the lowest range of head widths (1.4–1.7 mm) was clearly separated from the rest of the population. Three peaks were also noted in the distribution of the grubs' body length (Fig. 5). The group of grubs at the highest range of body length (9–18 mm) was clearly separated from the rest of the population. Only two groups were noted in the distribution of the ratio of the grubs' body length to head width (Fig. 6). Two thirds of the grubs had a ratio of 2.1–2.9, while the other third had a ratio of 4.1–7.3. These results were compared to data

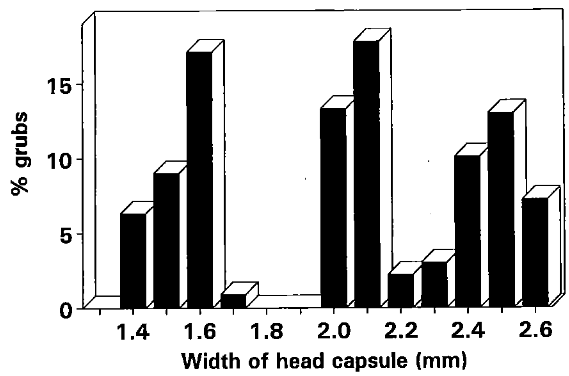


Fig. 4. Frequency distribution of head width in a field population of *Maladera matrida* grubs (n = 120).

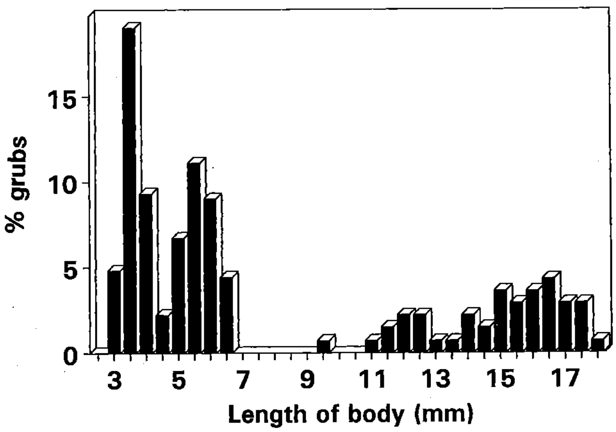


Fig. 5. Frequency distribution of body length in a field population of *Maladera matrida* grubs (n = 120; intervals 0.5 mm).

obtained during morphological measurements of laboratory-reared grubs, in order to define the stadia. It appeared that each instar could be defined by a different anatomical feature with a high degree of confidence: first instar, head width less than 1.8 mm; second instar, head width above 1.9 mm and ratio of body length to head width below 3; and third instar, body length above 9 mm.

Secondary sexual characteristics

We were not able to determine the sex of *M. matrida* at the larval stage. However, male pupae could be easily identified by the ampulla covering the developing genitalia on the posteroventral abdominal segments (Fig. 3).

The weight of beetles ranged widely between 15 and 85 mg, but there was no significant difference between the weight of males (mean ± SD = 42 ± 15 mg; N = 42) and females (mean ± SD = 40 ± 16 mg; N = 58). The range of antennal laminae length was 570–790 µm (mean = 704 µm) in females and 750–900 µm (mean = 823 µm) in males (Fig. 7). However,

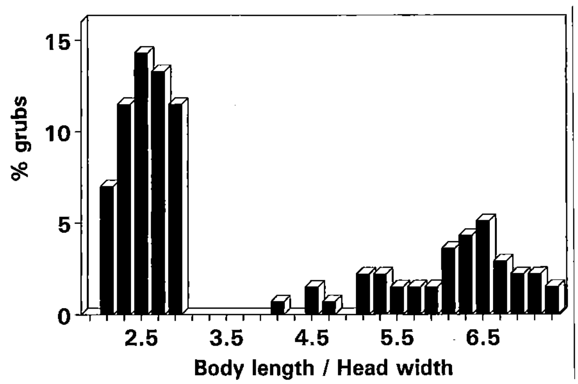


Fig. 6. Frequency distribution of the ratio between body length and head width in a field population of *Maladera matrida* grubs (n = 120; intervals 0.2).

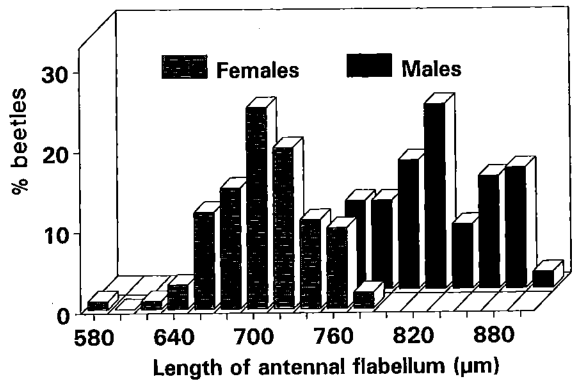


Fig. 7. Frequency distribution of antennal laminae length in a field population of *Maladera matrida* adults (n = 100 of each sex; intervals 20 µm).

although males usually had longer antennal laminae than females, this parameter overlapped between the sexes in the range 750–790 μm, which accounted for about 10% of the beetles we had examined. The length of antennal laminae may still be used to determine the beetles' sex if those falling into the overlap zone will be excluded. Ascher et al. (1990) reported that the length of antennal laminae of *M. matrida* females ranged between 625–750 μm, while in males it was longer than 875 μm. The differences in measurements between the two studies may be due to the fact that while Ascher examined a laboratory-reared population, we examined field-collected specimens.

We did not see differences between males and females of *M. matrida* in the shape of the tibia and tarsus of the prothoracic legs, as had been described for the Japanese beetle by Fleming (1972). We were unable to find differences in the shape of the posterior abdomen between the sexes in *M. matrida* as described by Argaman (1986).

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