

**ORIGINAL RESEARCH ARTICLE****Morphological description of *Dirhinus spp.*, a parasitoid of black soldier fly (*Hermetia illucens*) pupae detected in Bondo**

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Abstract

The farming of the black soldier fly (BSF) as a feed insect is on an increasing trend in Kenya. However, the enterprise is now under threat from parasitoids of the pupae stage. The objectives of this communication are to create awareness on a recently described wasp parasitoid discovered within the Jaramogi Oginga Odinga University Black Soldier fly farm in Bondo; and highlight possible mitigation management strategies. The parasitoid was isolated from pupa stage and characterized using a binocular microscope to observe and measure various body parts such as body length without antennae, antenna length, forewing length, head capsule width, thorax width, and abdomen width. Results indicated that the body length without antennae was 5.15 mm, body length with antennae was 7.97 mm, antenna length was 1.5 mm, forewing length 2.81 mm while the widths of the head capsule, thorax, and abdomen were 1mm each. The parasitoid was subsequently identified by use of dichotomous keys to belong to order Hymenoptera, family Chalcididae, and Genus *Dirhinus*. The study recommends further characterization of the parasitoid to species level by molecular techniques and use of suggested sustainable management methods to contain the parasitoid menace within BSF farms.

Keywords: Insect rearing, black soldier fly, pupae, parasitoids, morphology, Kenya

1.0 Introduction

The scarcity of conventional sources of protein such as fish, meat, and soybean has made the feeding of an ever-increasing population a real challenge (Diehl et al., 2014). Sustainable alternatives such as farming of edible insects is regarded as a potential solution to attaining both food and the elusive nutritional security (Veldkamp, 2012). In Kenya, the farming of the black soldier fly as a source of protein for feed production is on the increase chiefly due to the non pest status, high reproductive and feed conversion rates and ability to valorize diverse organic wastes (Nyakeri et al., 2017).

One of the pioneer black soldier fly farms is located at Jaramogi Oginga Odinga University of Science and Technology main campus in Bondo (GPS: 0° 14' 19.00" N; 34° 16' 9.98" E) (Kenya Google Maps, 2023). The black soldier fly colony was started in 2015 by attraction of wild black



soldier flies (Nyakeri *et al.*, 2017b). The colony has passed through several successful generations over the years. However, after seven successful years, we report the citing of a parasitoid that infects the pupa stage and prevents emergence of adults. This parasitoid threatens black soldier fly farming not only in Bondo but also in Kenya at large.

Wasp parasitoids have been reported in other African countries among them Nigeria, Mali, Ghana and Tanzania. These parasitoids were also reported to attack the pupa stage (Devic and Maquart, 2015; Maquart *et al.*, 2020). In Kenya, there are also records of wasp parasitoids in agricultural crop farms. However, this is the first record of a parasitoid attack in a Kenyan insect farm.

2.0 Materials and methods

2.1 Detection of parasitoid infection

It started with the observation that pupa took unusually long to emerge into adults. Upon inspection by gentle squeezing of the abdomen of sampled pupa from different pupation trays revealed unusual softness and tenderness compared to normal pupation which is characterized by progressive hardening of the pupa as the embryo develops within the pupal casing. This was followed by actual opening of the pupa casing by cutting off the puparium end to reveal a creamy to yellow coloured fluid. Further inspection of the pupation trays revealed presence of the parasitoids. Twelve parasitoids were randomly selected and picked using a pair of forceps from the affected pupation trays, preserved in 70% ethanol, and taken to the microbiology laboratory for detailed microscopic observation according to the procedure developed by Noyes (1982).

2.2 Parasitoid microscopic analysis.

The samples were dried in an oven at 104 °C for 4 hours. After desiccation, the samples were then prepared for microscopic morphological examination (Noyes (1982)). Magnification was done using a binocular microscope (Make: Coslab® model: SZM-115 LED) to observe and measure different parameters. Measurements were taken for body length with antennae, body length without antennae, antenna length, forewing length, head capsule width, thorax width, and abdomen width to the nearest millimeter by placing each specimen on a millimeter paper. Photos of the magnified insect and parts were taken using a SONY lens G 20 X Optical Zoom digital camera. The data was processed using Excel 2013 software.

2.3 Parasitoid identification

Identification was done using the identification keys developed by Delvare and Aberlenc (1989) and Delvare and Copeland (2018). The Delvare and Aberlenc (1989) key enabled identification of the order and family of the parasitoid while Delvare and Copeland (2018) facilitated the identification of the genus as it is more precise and specific.



3.0 Results

3.1 Measurement of the body parts and insect size.

The measurement obtained for body length without antennae, body length with antennae, antennae length, forewing length and widths of head capsule, thorax and the abdomen are summarized in table I.

Table I: Average measured size of different body parts of the parasitoid in millimeters.

Parameters	Average \pm SD
Body length without antennae	5.15 \pm 0.43 mm
Body length with antennae	7.97 \pm 0.58 mm
Antennae length	1.50 \pm 0.00 mm
Forewing length	2.81 \pm 0.23 mm
Head capsule width	1.00 \pm 0.00 mm
Thorax width	1.00 \pm 0.00 mm
Abdomen width	1.00 \pm 0.00 mm

3.2 Description of the parasitoid body parts

The head of the Hymenoptera is orthognathous. The antenna had 11 articles and angled between scape and pedicel while the first article is elongated (Fig. 1 A and Fig 1B). A metafemur at the level of the hind leg is also observed (Fig. 1 A). The thorax also has two pairs of membranous wings including two anterior (forewings) and two posterior (hindwings) (Fig. 1 A). The visible thorax elements include the pronotum, the mesoscutum, and the scutellum (Fig. 1 C). The forewing of the insect is characterized by a reduced number of veins and cells, and is covered by thin layers of setae. The forewing has the following elements: radial sector fold, marginal cells, marginal vein, post marginal vein, submarginal vein, and stigmal vein (Fig. 1 D). The junction of the abdomen and the thorax are reduced. These characteristics point to the identity of the insect as belonging to Order Hymenoptera, Family Chalcididae, and Genus *Dirhinus*.

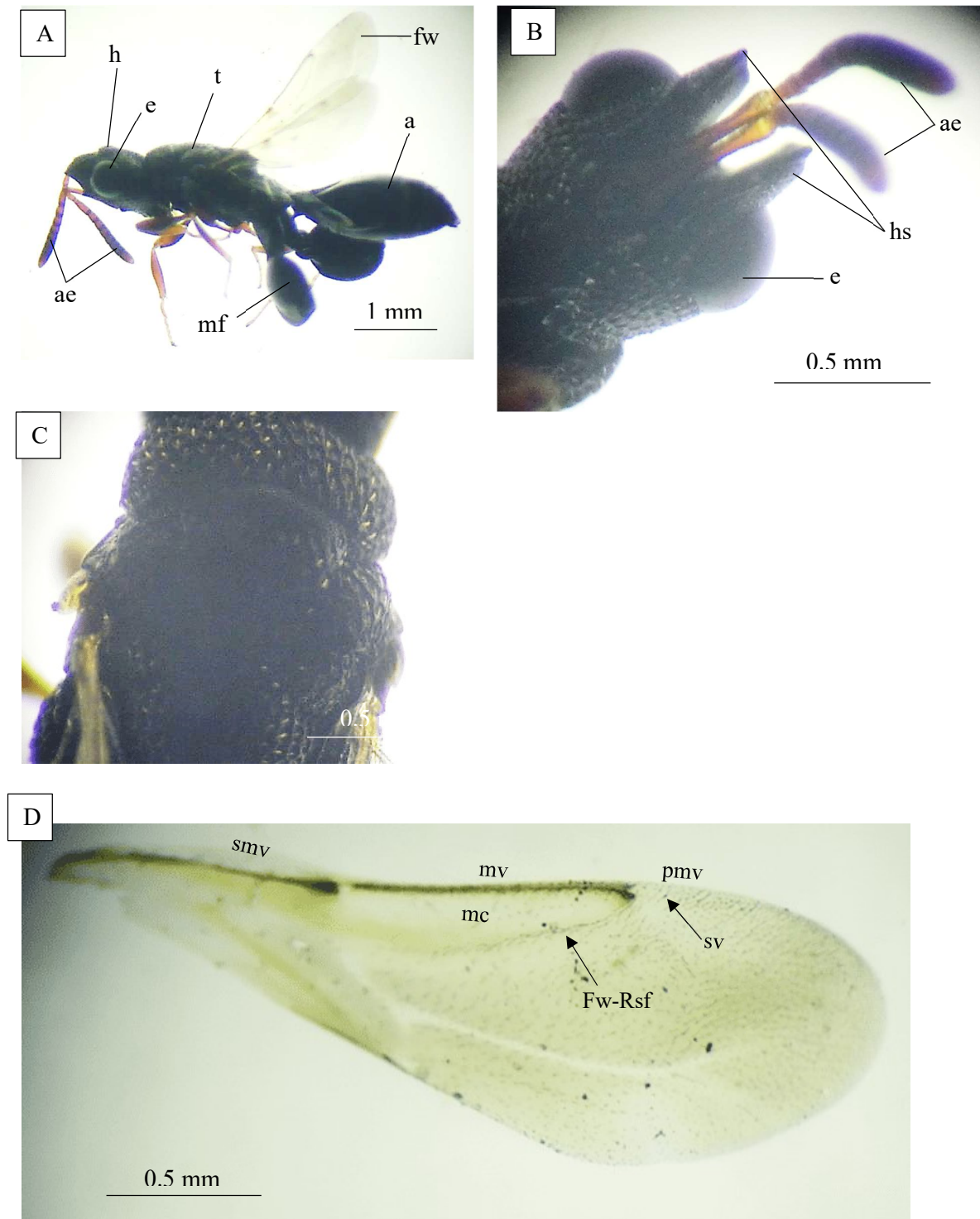
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Figure 1: A: Side view of the insect; B: Ventral view of the head; C: Dorsal view of the thorax; D: Fore wing. Abbreviations: a, abdomen; ae, antennae; e, eye; mf, metafemur; fw fore wing; h, head; hs, horns; msct, mesoscutum; prnt, pronotum; scut, scutellum; Fw-Rsf, Radial sector fold



of the forewing; mc, marginal cell; mw, marginal vein; pmv, post marginal vein; smw, submarginal vein; sv, stigmal vein; t, thorax;

4.0 Discussion

This study aimed at describing the morphology of a parasitoid that attacks pupa of the Black Soldier Fly (*Hermetia illucens*). The description enabled the identification of the parasitoid up to the genus level of *Dirhinus*. Genus *Dirhinus* was first reported in Kenyan agricultural areas using malaise traps (Delvare and Copeland, 2018). In the study four species of the genus were identified from samples collected in Central, Rift valley and Eastern Kenya namely *Dirhinus quadrhinus*; *Dirhinus gigasetosus*; *Dirhinus kambae*; *Dirhinus maasaii* and *Dirhinus leakeyorum* (Delvare and Copeland, 2018). However, three of the species were isolated in Rift Valley making it the most speciose sampled site in the study. However, the study did not identify the hosts of the parasitoids in these areas. It is also significant that the study did not report the presence of the parasitoid in Nyanza despite carrying out sampling in the region. Therefore, this is the first-time genus *Dirhinus* is being reported in Nyanza region.

Fruit flies are reported to be commonly parasitized by different species of genus *Dirhinus*. These include *D. giffardii* which has been introduced into different parts of the world, *D. inflexus* whose host is the *Glossina* spp. The hosts pupate within the soil and when a female parasitoid finds a suitable host pupa, she deposits eggs within the intersegmental areas of the pupa (Bradley and Sheppard, 1984). This is known as an idiobiont ectoparasitoid. The substrate storage section in the BSF facility at JOOUST has a lot of fruit flies, and therefore it is possible that the fruit flies introduced the parasitoids to the BSF facility. However, this needs to be confirmed by collecting puparia from different potential hosts to correctly associate the parasitoid with the source host.

The detection of the parasitoid in a black soldier fly pupae indicates that the parasitoid may be a generalist rather than a specialist (Jones *et al.* 2015). Generalists have diverse food sources and for this parasitoid, the food sources include BSF pupae. This may be adaptive as like other animals, insect species are vulnerable to habitat loss due to conversion of natural areas into small farms or large-scale agroforestry initiatives, and to the effects of global warming (Jeffs and Lewis, 2013).

It is however significant to note that the parasitoid was not detected within larval populations of BSF in the facility. This may indicate that the parasitoid is specific with respect to stage of development of the host. According to Bradley and Sheppard (1984), the wriggling activity of larvae and prepupa discourages female parasitoids from ovipositing on them. Again, it is also known that the larvae stage produces pheromones that deter infection and infestation by other organisms and enable them to colonize an area (Bradley and Sheppard, 1984).

Morphological differences were noted between samples collected in this study and the ones reported by Delvare and Copeland (2018). For instance, the body length of this insect is about 5.15 mm, whereas the species described by Delvare and Copeland (2018) were *Dirhinus*



quadrhinus (4.2 mm); *Dirhinus gigasetosus* (3.5 mm); *Dirhinus kambae* (3.65 mm); *Dirhinus maasaii* (4.2 mm) and *Dirhinus leakeyorum* (3.05 mm). This shows that the parasitoid found in the JOOUST Black Soldier Fly farm in Bondo is much larger than those found in the agricultural farms and may be a completely different species. Variations in some specimens belonging to the same species but sampled in different environments have been noted by other authors such as Delvare et al. (2019). Indeed, these authors noted that specimens of *Eniacomorpha hermetiae* sampled in Ghana had the flagellum, fore, and mid legs darker and browner than those from Kenya.

In view of the importance of BSF, the parasitoid description down to the species level is necessary. This would determine whether it is novelty as a new species or otherwise. In any case, the results of this study are promising and could help in the development of management strategies.

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