First report of *Exorista xanthaspis* (Wiedemann, 1830) (Diptera: Tachinidae), a larval-pupal parasitoid on invasive pest, *Spodoptera frugiperda* (J. E. Smith) in maize from India

OMPRAKASH NAVIK*, T. VENKATESAN and T.R. ASHIKA
ICAR-National Bureau of Agricultural Insect Resources, Post Bag No. 2491, H. A. Farm Post, Bellary Road, Hebbal, Bengaluru – 560 024, Karnataka, India
*Corresponding author E-mail: omnavikm@gmail.com

ABSTRACT: For the first time, the tachinid fly, *Exorista xanthaspis* (Wiedemann, 1830) (Diptera: Tachinidae), was found to parasitize the larvae of fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae) collected from maize fields in Karnataka, India. The field survey conducted during 2018-19 revealed the occurrence of *E. xanthaspis* on the larvae of *S. frugiperda* with the parasitism rate of 1.85 to 4.55% in maize fields. The identity of tachinid parasitoid was confirmed by amplifying Cytochrome Oxidase I gene (CO1-658 bp) and DNA bar-coding.

KEY WORDS: Parasitoid, *Spodoptera frugiperda*, Tachinid fly

Field surveys were conducted in major maize growing areas in 2018-19 from July to November 2019 in Chikkaballapur, Karnataka. The different stages of *S. frugiperda* larvae were collected from the infested maize fields based on the feeding injury on maize leaf and presence of frass (larval excreta) on maize leaf whorl. The larvae were pulled out from the infested maize leaf whorl and placed in the plastic bags along with maize leaf. However, younger larvae were collected along with a portion of maize leaf and...
kept in sealed plastic bags. *Spodoptera frugiperda* larvae were identified based on inverted ‘Y’ shaped marking on head and four dot square on upper surface of last abdominal segment (EPPO, 2015; Prasanna et al., 2018). On reaching laboratory, the larvae were segregated instar wise and later reared individually on the artificial diet as per methodology developed by Ballal et al. (1995) in growth chamber (27 ± 2°C; RH 65 ± 5%; 12L: 12D h photoperiod) till parasitoid emergence or adults of *S. frugiperda*. The emerged parasitoids from infested larvae were collected and preserved in 95% ethanol for molecular identification.

**Molecular Based Identification of Tachinid Parasitoid**

**DNA Extraction and Sequencing**

DNA extraction was done using a hind leg of the tachinid parasitoid using Qiagen DNeasy® kit, following the manufacturer’s protocols. The DNA extracts were subjected to Polymerase Chain Reaction (PCR) amplification of a 658bp region near the 5’ terminus of the COX1 gene following the standard protocol (Hebert et al., 2003). Primers used were: forward primer (LCO 1490: 5’- GGTCACAACATCATAAAGATATTGG-3’), and reverse primer (HCO 2198: 5’- TAAACTTCAGGGT GACCAAATACTCATCA-3’). PCR reactions were carried out in 96-well plates, 50µL reaction volume containing: 5 µL GeNeiTm Taq buffer, 1 µL GeNeiTm 10mM dNTP mix, 2.5 µL (20 pmol/µL) forward primer, 2.5 µL (20 pmol/µL) reverse primer, 1 µL GeNeiTm Taq DNA polymerase (1 U/µL), 2µL DNA (50 ng/µL), and 36µL sterile water. Thermocycling consisted of an initial denaturation of 94°C for 5 min, followed by 30 cycles of denaturation at 94°C for 1 min, annealing at 46°C for 1 min and extension at 72°C for 1 min. PCR was performed using a C1000™ Thermal Cycler. The amplified products were analyzed on 1.5% agarose gel electrophoresis as described by Sambrook and Russell (2001). The amplified products were sequenced. The species was bidirectional sequenced and checked for homology, insertions and deletions, stop codons, and frame shifts by using NCBI BLAST and ORF finder. The sequence was uploaded to GenBank and the Barcode of Life Database (Ratnasingham and Hebert, 2007).

In total, 92 larvae of *S. frugiperda* were collected from the infested maize fields and the tachinid parasitoids emerged from the pupae of *S. frugiperda* and they were found to be larval-pupal parasitoids (Fig. 3). The extent of parasitism by the parasitoid was 1.85 to 4.55% on larvae during rainy season in maize habitat (Table 1).

### Table 1. Collection locality and parasitism rate of *Exorista xanthaspis* on larvae of *Spodoptera frugiperda*

| Species          | Locality                          | Parasitism (%) |
|------------------|-----------------------------------|----------------|
| *Exorista xanthaspis* | Karnataka: Chikkaballapur, 06.vii.2019 (13°22' N 77°44' E) | 1.85           |
| *Exorista xanthaspis* | Karnataka: Chikkaballapur, 08.viii.2019 (13°30’ N, 77°35’ E) | 4.55           |

**Occurrence of *E. xanthaspis* has been reported on many noctuid moths of crop pests in India. The tachinid fly was found to cause 3% parasitism on the larvae of *Amsacta moorei* Butler in Rajasthan (Verma, 1983). Parasitism of *E. xanthaspis* was recorded on *A. albistriga* (Walker) on groundnut from Karnataka (Veenakumari et al., 2008) and *H. armigera* infesting different field crops in India (Bhatnagar, 1982). However, *E. xanthaspis* was found to have expanded...
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its host range by parasitizing the invasive *S. frugiperda* in maize. *Exorista xanthaspis* is also known to attack abundantly the larvae of various species of genus *Spodoptera* with different parasitism rate worldwide (CABI, 2019). Verma (1983) reported 3% larval parasitism of *A. moorei* by *E. xanthaspis* in Rajasthan. The higher parasitism (8%) of this tachinid was observed on *S. exigua* larvae in cotton fields in Turkey (Efil and Kara, 2004). In Africa, another tachinid fly, *Palexorista zonata* appeared as a primary parasitoid with 12% larval parasitism in maize within two years of invasion of *S. frugiperda* (Sisay et al., 2018). We recorded lower parasitism (4.55%) of *E. xanthaspis* in the early stage of pest invasion, as the pest is yet to establish in the native environment and parasitism level of local parasitoids may improve in future. Recently, the occurrence of tachinid, *E. sorbillans* was reported with negligible levels of parasitism of fall armyworm in India (Sharanabasappa et al., 2019). Approximately 100 tachinid species have been incorporated in the biological control programme of crops or forest pests with partial or complete success (Grenier, 1988). Therefore, recruiting the local parasitoids or conserving them to suppress the population of invasive pest could be an alternative to the chemical approach. In the present study, we report the new association of *E. xanthaspis* with *S. frugiperda* for the first time and further a new addition to the parasitoid assemblages of fall armyworm in India. Conservation of the tachinid parasitoids may limit and regulate the pest population in structuring the natural control of pest in maize habitat.

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