



E-ISSN: 2320-7078

P-ISSN: 2349-6800

JEZS 2019; 7(6): 1245-1250

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Received: 11-09-2019

Accepted: 15-10-2019

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## Studies on biology, feeding habits and natural enemies of fall armyworm, *Spodoptera frugiperda*, a new invasive pest in India

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**Abstract**

Fall Armyworm, *Spodoptera frugiperda* (J.E. Smith), a new invasive insect pest is native to Americas and it was recorded as devastating pest of Maize in African Countries in 2016. In India, the occurrence of FAW was reported from Karnataka state during May 2018. In Southern Agroclimatic zone of Andhra Pradesh, Fall Armyworm was first noticed in Chittoor District during August 2018 on Maize. Subsequently on other crops viz., Ragi, bajra, fodder maize, sugarcane. In the fields of early stages of Maize crop, egg masses that are covered with cream coloured hair and scraping symptoms on the leaves were observed. The scraping symptoms caused by first and second instars, shot holes and voracious cut feeding by later instars were recorded. The larvae were collected from maize fields of Chittoor district, culture was maintained in the laboratory, studied the characters of egg, six larval instars, pupa and adult stages. The confirmation as *Spodoptera frugiperda* was done with the key identification character viz., "distinct inverted Y shaped suture on head region and exact square shaped pustules on 8<sup>th</sup> segment of the abdomen". The cadavers of *S. frugiperda* with tree top symptom were noticed in maize fields of Chittoor District, the pathogen was identified as Nuclear Polyhedrosis Virus (NPV). Like wise from the cadavers collected from Anantapur District, Cytoplasmic Polyhedrosis Virus (CPV) infection was confirmed. *B.t* infected caterpillars were also seen in maize fields and the pathogen was confirmed. During 2019 kharif, the cadavers infected with *Metarhizium anisopliae* were collected from maize fields. *S. frugiperda* larvae were treated with NPV, *B.t*, *Nomuraea rileyi* and *Metarhizium anisopliae* in the laboratory at  $1 \times 10^7$  POBs or Spores per ml concentration and found the larvae highly susceptible to all the pathogens.

**Keywords:** Fall armyworm, maize, damage, Egg, larva, pupa, adult, NPV, CPV, *B.t*.

**Introduction**

Fall Armyworm, *Spodoptera frugiperda* (J.E. Smith), which is native to Americas and it was recorded as devastating pest of Maize in African Countries in 2016. In India, the occurrence of FAW was reported from Karnataka state during May 2018. In Southern zone of Andhra Pradesh, Fall Armyworm was first observed during August 2018 as voracious defoliator in kharif Maize in the fields of Regional Agricultural Research station (RARS), Tirupati and farmers fields of Madanapalli Division. Subsequently, the pest was observed on other crops viz., Ragi, bajra, fodder maize, fodder grasses and sugarcane. In case of sugarcane, the newly planted crop during February 2019, the infestation was noticed. However, Maize and fodder grasses were observed as suitable, preferred hosts compared to Ragi, Bajra and Sugarcane. Farmers in Tirupati region have cut down the severely infested fodder maize during September – October months. During 2019 kharif also, Fall Armyworm is recorded as major pest in southern zone of Andhra Pradesh. Dumas *et al.* (2015) reported that Fall Armyworm consists of two strains adapted to different host plants. One is Maize strain feeds predominantly on Maize, cotton and sorghum while second is rice strain that feeds primarily on rice and pasture grasses.

According to Reports of USAID (United States Agency for International development) – CYMMIT (The International Maize and Wheat Improvement Center) – CGIAR (Consultative group on International Agricultural research) (2018), Fall Armyworm is capable of attacking more than 80 different plant species but has major preference for Maize.

**Material and Methods**

In kharif 2018 Maize crop, Fall Armyworm, *Spodoptera frugiperda* incidence was noticed for the first time during August month in Chittoor district of Southern zone of Andhra Pradesh. In

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In B. kothakota and Santhipuram Mandals of Chittoor district, the symptoms and insect stages were recorded. Close observations were made on egg masses, and damage symptoms in the Maize crop present in different stages in RARS, Tirupati and nearby villages during *kharif* 2018. The larvae and egg masses were collected from the fields and cultured in the laboratory. The biology and life cycle studies were taken up under laboratory conditions of 25 – 35 °C and 65 to 80% Relative Humidity. Grownup larvae were collected from Maize fields, fed with Maize whorls daily in rearing glass jars. Fully grown larvae were provided with soil to pupate. The pupae were arranged in a petriplate in insect rearing cages. The maize whorls of small plants were kept in conical flasks having water and these flasks were arranged in cages. Fifty per cent honey dipped cotton swabs were provided for the sake of emerging adults to suck honey. The adults laid egg masses on Maize whorls. These egg masses were utilized for studying the life cycle. Morphology of eggs, different larval instars, pupae and adults was studied and documented. The symptoms and feeding habits were studied in field conditions in the Research Station and also farmers fields of Chittoor District during *kharif* 2019 also.

During the surveys made for the incidence of *S. frugiperda*, also observed for any natural occurrence of cadavers infected with pathogens viz., Nuclear Polyhedrosis Virus (NPV), Cytoplasmic Polyhedrosis Virus (CPV), *Bacillus thuringiensis* (*B.t*) and Fungus. The cadavers were carried to the laboratory and characterized. The pathogens were isolated. The NPV infected cadavers were macerated with minimum quantity of water, centrifuged at 500RPM to remove debris as supernatant and at 5000 RPM to obtain NPV polyhedra as sediment. For CPV also same procedure followed. *B.t* was isolated on Luria bertani Agar Medium and fungus on Sabourauds Dextrose Agar medium. Pathogenicity tests with isolated pathogens were carried out at  $1 \times 10^7$  POBs or spores per ml.

## Results and Discussion

### Symptoms of damage Observed in the Field Surveys in Southern Zone of Andhra Pradesh and in the trials conducted at Regional Agricultural Research Station, Tirupati

In the fields of early stages of Maize crop, egg masses that are covered with cream coloured hair and scraping symptoms on the leaves were observed. While the tuft of hair is removed, cream – golden yellow coloured, spherical eggs were seen. After hatching from eggs, the neonate larvae were observed to feed on that leaf for 1 or 2 days, later dispersed to adjacent leaves. With the close observation in the fields, the scrapings are confirmed that done by first and second instar larvae. Severely scraped leaves were seen coming to drying. In mild days, adult moths also observed sitting in the whorls may be for oviposition. In the fields where Maize crop is with matured leaves (above 1 month age of the crop), along with egg masses, scrapes on older leaves (some were dried), the matured leaves with series of different shaped shot holes and severely cut whorls (Like cut with sickle or grazed by animal) with excreta were observed. In the plants with fresh excreta, when the whorl is opened, 1 to 4 larvae were found.

The leaves with series of shot holes were confirmed to be done by third and fourth instars whereas severely cut whorls done by still grownup larvae. Marengo *et al.* (1992) [3] reported that first instar larvae of FAW consume leaf tissue from one side, leaving the opposite epidermal layer intact. By

the second instar, larvae begin to make holes in leaves and eat from the edge of the leaves inward. Feeding in the whorl of corn often produces a characteristic row of perforations in the leaves. They reported cannibalistic behavior in FAW larvae. They recorded 0.2 to 0.8 mean larvae per plant during late whorl stage could reduce maize yield by 5 – 20 per cent. Pannuti *et al.* (2015) [4] reported that in vegetative stage, FAW prefers leaf tissue whereas on mature plants, the larvae tend to settle and feed on ear zone particularly on silk tissues.



Fig 1: Egg mass of *S. frugiperda* covered with hair tuft



Fig 2: Scraping on leaves done by neonate larvae of *S. frugiperda*



Fig 3: Series of shot holes observed on Maize due to whorl feeding by *S. frugiperda* larva

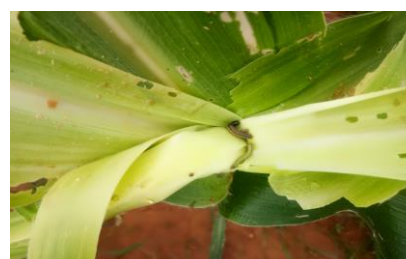


Fig 4: In some whorls, 2-4 larvae found

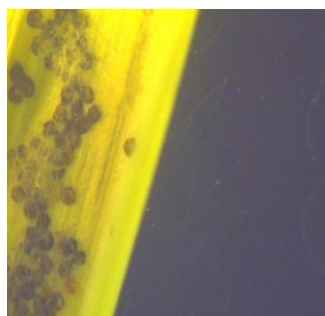


**Fig 5:** Matured *S. frugiperda* larva voraciously feeding by cutting the whorl of Maize. Heavy faecal matter is also seen.

**Egg stage:** Preoviposition of 2-4 days recorded. The eggs were laid in masses on leaves and whorls. Freshly laid eggs were seen in cream colour covered by golden coloured hair tuft. The eggs were laid in masses on upper surface of leaves and on the stem portion. The bottom portion of egg is seen flat and top portion is dome shaped. The eggs are 24 – 30 microns in size (at 20X, 472.8 to 593.3microns in diameter i.e 0.47 to 0.59mm). Before hatching, the eggs turned reddish brown and then black. Egg period recorded was 3 to 5 days. Egg period according to reports of USAID – CYMMIT - CGIAR (2018), egg stage was 2 – 3days, the eggs were dome shaped with bases flattened, 0.4 mm diameter and 0.3 mm height.



**Fig 6:** Cream coloured eggs of *S. frugiperda* turning to reddish tinged



**Fig 7:** Eggs of *S. frugiperda* became flat and turned brownish black before hatching.

#### Larval stage

**First instar:** larva was light green in colour with brown head. At 6.7X magnification, It was measured 0.71mm in length and with 0.16mm head capsule width. The duration of first

instar was 5 days.

**Second instar:** larva was cream coloured with blackish brown head. It was measured 3.5 to 4mm length and 0.5mm head capsule width.

**Third instar:** was light green- cream in colour with three longitudinally light lines. The head and first thoracic segment was black in colour. The length was 7mm and head capsule width was 2mm. On each segment of the body, 6 dark pustule like spots were seen. The side two spots are large in size, middle ones are medium and central spots are small in size. The lateral areas where dark spots are located are dark greenish – brown in colour. Upto penultimate segment of the abdomen, the big spots are dark.

**Fourth instar:** was seen with brownish black body with reddish pigmentation. Head and first thoracic segment was black in colour. Second thoracic segment light black in colour. All the spots arranged as in case of third instar with increase in size and with more darkness. It was measured 16mm length and Head capsule width was 2.5mm.

**Fifth instar:** The colour of the body was yellowish to cream. Colour of the head was brick red with dark brown areas. Thoracic and abdominal segments are similar in colour. The area around the hair was black ring in first thorax only. In remaining segments, Black ring and lighter area present. Light dorsal longitudinal lines became more lighter. Side areas were brown in colour. Fifth instar was 22mm in length, width of head capsule was 3mm.

**Sixth instar:** The body was reddish cream, head was brown, first thorax light brown. Dorsal lines and lateral dark areas became light. Spots became black in colour, surrounding areas reduced in size, ring is black and surrounding area is light in colour. On first thoracic segments, spots are very small. On the abdomen and on 2<sup>nd</sup> thoracic segment, the spots are large. Spiracles were found very black in light brown lateral areas. Length of the body was 28mm and head capsule with 3mm. Total 6 instars were recorded. The key identification characters viz., inverted ‘Y’ shaped epicranial suture on 8<sup>th</sup> abdominal segment are very clearly seen in all the instars of larva.

Pitre and Hogg (1983)<sup>[5]</sup> reported that there are six instars in Fall Armyworm. Larvae attained lengths of 1.7, 3.5, 6.4, 10.0, 17.2 and 34.2mm respectively during the six instars. The duration of larval period recorded was 14days during summer and 30 days during cooler months.



**Fig 8:** instar larva of *S. frugiperda*



**Fig 9:** instar larva of *S. frugiperda*



**Fig 11:** 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> instar larvae of *S. frugiperda*



**Fig 12:** Inverted “Y” shaped clear suture on the dorsal portion of head of larva



**Fig 14:** Four black pustule like spots arranged in exact square on 8<sup>th</sup> abdominal segment of larva

#### **Pupal stage**

Pupa is dark brown in colour with two small spine like structures at the end. Male pupa looked small with 1.1 – 1.3

cm in length and 1.9 to 2.0 mm width (at middle of pupa). Female pupa was comparatively larger with 1.5 – 1.6cm length and 2.0 to 2.3 mm width at middle. Total 10 segments were visible in pupa. The terminal segment width in male was 0.73 to 0.86mm and in female, 0.31 to 0.48mm. Two small hook like structures were present at the end of pupal body and these hooks are widely arranged in female pupa. Pupal period recorded was 9 days. Pitre and Hogg (1983) [5] the pupation of FAW at 2-8cm depth of the soil. They reported that pupal cocoon was oval in shape with 20 – 30 mm in length. The pupa was reddish brown in colour, measuring 14 – 18 mm in length and 4.5 mm in width.



**Fig 15:** Pupae of *S. frugiperda*



**Fig 16:** Tips of Pupae with two hook like structures

**Adult stage:** Adults resembling *Spodoptera litura*. Body size, wing span are found less than *S. litura*. In males, length of the body was 1.5cm, front wing length 1.2cm, hind wing 0.9cm, wing span was 2.9cm. The front wings are darker than in females. In female, body length is 1.5cm but body is robust when compared to males. Fore wing length 1.2cm, hind wing 0.9cm, wing span 3.0 to 3.1cm. (In *S. litura* male, body length 1.5cm, fore wing length 1.4cm, hind wing length 0.9cm, wing span 3.2cm. In female, body 1.9cm, fore wing length 1.6cm, hind wing length 1.1cm, wing span 3.3cm). In markings on front wings (White areas) also, some differences were recorded when compared to *S. litura*. Adult life span recorded was 7 days. According to Pitre and Hogg (1983) [5], the moths of FAW have a wingspan of 32 – 40 mm. In the male moth, the forewing generally is shaded grey and brown with triangular white spots at the tip and near the center of the wing. The forewings of females are less distinctly marked, ranging from an uniform greyish brown and to a fine mottling of grey and brown. The hind wing is iridescent silver – white with a narrow dark border in both males and females.



**Fig 17:** Adults of *S. frugiperda*

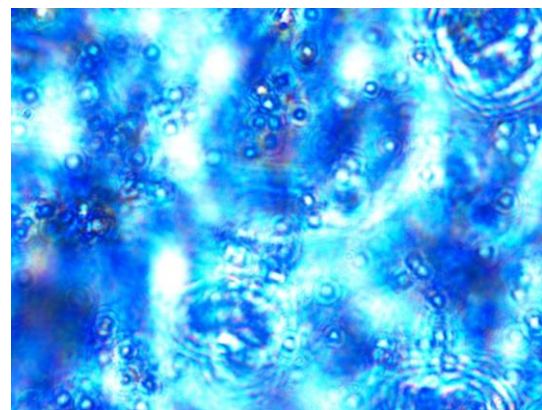


**Fig 20:** CPV and *B.t* infected *S. frugiperda* larvae

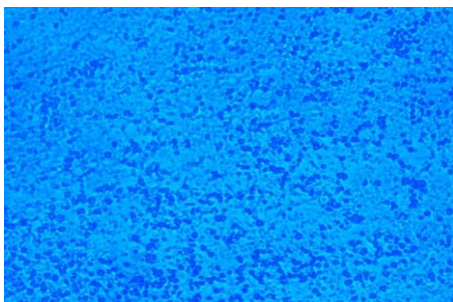
**Natural infection of microbial pathogens to *Spodoptera frugiperda*:** During September – October, 2018, The cadavers of *S. frugiperda* with tree top symptom were noticed in maize fields of Tirupati region, they were carried to the laboratory, made into suspension and studied the characters of POBs and the pathogen was identified as NPV (SfMNPV)



**Fig 18:** NPV infected and dead *S. frugiperda* larva by exhibiting Tree top Symptom on Maize plant



**Fig 21:** Polyhedra of CPV of *S. f* at 400X (Original size of POBs is 17.9 to 21.2 microns) (7.1 to 8.4mm at 400X and 19.1 to 25.1mm at 1000X). Out border is total white in colour. (Binocular Research microscope)

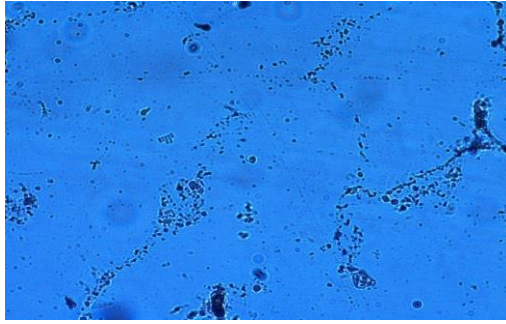


**Fig 19:** Polyhedra of *S. f* NPV at 400X (Original size of POBs is 5 to 8 microns) 92.1 to 3.2mm at 400X and 4.8 to 7.4mm at 1000X). Out border thick and black. (Binocular research microscope)

From the field collected larvae, one CPV infected (suspected as CPV with fragile integument with enlarged disproportionate head) was obtained. Like wise *B.t* infected (suspected as *B.t* with shriveled integument and blackish discolouration) and dead caterpillars were also seen in maize fields, collected and the pathogen was confirmed in the laboratory.



**Fig 22:** *B.t* infected and dead *S. frugiperda* larva on maize plant (Natural infection during kharif 2018)

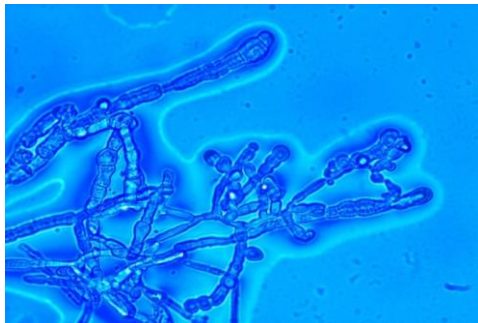


**Fig 23:** Spores and crystals of *B.t* from *S. f* at 400X

During September 2019, in *kharif* maize, white mummified cadavers were found and it is confirmed as *Metarhizium anisopliae* infection.



**Fig 24:** Mummified fall Armyworm larva with *Metarhizium anisopliae*



**Fig 25:** Conidiophores and chains of conidia of *M. anisopliae*

Third instar *S. frugiperda* larvae were treated with NPV, *B.t*, *Nomuraea rileyi* and *Metarhizium anisopliae* suspensions @  $1 \times 10^7$  POBS/Spores per ml and found highly susceptible to all microbial agents.

Behle and Popham (2012) and Haase *et al.* (2015) studied the efficacy of granulosis virus (SfGV) and multiple nucleopolyhedrovirus (SfMNPV) and reported that SfMNPV has greater potential for use in the management of FAW. All *et al.* (1996) stated that although several pathogens have been shown experimentally to reduce the abundance of Fall Armyworm larvae in corn, only *Bacillus thuringiensis* is feasible and its success depends on having the product on the foliage when the larvae first appear.

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