

Original Research Article

Baseline susceptibility and resistance monitoring for Novaluran 10% EC against *Spodoptera frugiperda* (J. E. Smith)

ABSTRACT

Study—A study was undertaken to assess the susceptibility of fall armyworm (FAW), *Spodoptera frugiperda* (J. E. Smith) to Novaluran (10% EC) by diet overlay bioassay method. Novaluran 10% EC was tested *in vitro* against *S. frugiperda*, obtained from four important ~~maize-producing~~maize-producing tracts in Tamil Nadu. The susceptible population was obtained from FAW laboratory at TNAU which was in the 150th generation and further, reared up to 157th ~~generations~~generation. The LC₅₀ and LC₉₅ values of Novaluran 10% EC to the susceptible population decreased from 0.832 to 0.746 ppm and 2.134 to 1.451 ppm, respectively. The susceptibility index of Novaluran was 1.115 and 1.470. The F157 population of *S. frugiperda* was used to conduct the preliminary discriminating dosage in Novaluran 10% EC and was 0.746 ppm. Resistance monitoring studies of *S. frugiperda* from Coimbatore, Perambalur, Salem, and Theni districts revealed that the LC₅₀ values of Novaluran 10% EC ranged from 0.792 and 0.930 ppm for Theni and Coimbatore, respectively. Novaluran 10% EC had the highest resistance ratio of 1.246 fold in the Coimbatore field population and the lowest resistance ratio of 1.061 fold in the Theni field population when compared with the TNAU FAW laboratory susceptible population of *S. frugiperda*.

Formatted: Font: 11 pt

Formatted: Font: 11 pt

Keywords: Baseline insecticide susceptibility; discriminating dose; *S. frugiperda*; Novaluran 10% EC; resistance ratio; resistance monitoring; susceptibility index

1. INTRODUCTION

The fall armyworm (FAW), *Spodoptera frugiperda* (J.E. Smith), (Lepidoptera: Noctuidae) is a voracious insect pest endemic to the Western Hemisphere, mainly in South America Pogue (2002) [1]. It is one of the most quickly spreading and invasive maize pests in Africa and Asia (Tambo et al.,2021; Ramasamy 2022) [2-3]. *S. frugiperda* has become a pest species due to biological traits like polyphagy, hidden larval feeding habits, high reproductive

capability, adult dispersal, and many generations per year (Li et al 2020; ~~bird-Bird~~ et al 2021)[4-5]. *S. frugiperda* is an economically important pest that infects maize and other Gramineae-family crops (Andrews, 1988) [6]. Fall armyworm is a polyphagous insect that causes substantial damage to cereal and vegetable crops (Goergen et al., 2016; Roger et al., 2017) [7-8]. Moths have ~~a~~-migratory ~~behavior-behaviour~~ as well as a more confined dispersion habit. Before oviposition, they can travel beyond 500 km (300 miles) (Prasanna et al., 2018) [9]. FAW was initially detected in West Africa in late 2016 (Goergen et al., 2016) and has since spread to 44 African countries (Bhusal et al., 2019) [7,10]. Sharanabasappa and Kalleshwaraswamy (2018) reported the first appearance of this novel invasive pest FAW in India [11]. ~~The fall armyworm was first detected in sugarcane in Tamil Nadu (Srikanth et al., 2018)[12]. The invasion of the fall armyworm threatened India's food security at the time.~~ It feeds on at least 186 host plants and is a major pest of economically important crops such as corn, sorghum, and rice (Casmuz et al. 2010) [13]. FAW has evolved resistance to 29 insecticides with six distinct mechanisms of action (Chao et al., 2019) [14]. In every farming system where pesticides are frequently employed, resistance is ~~the~~-a major concern, and monitoring the target pest's susceptibility is critical for successful integrated pest management (IPM) and insecticide resistance management (IRM). Insecticide use is literally expanding in maize fields in ~~the~~-recent days. As a result, the goal of this study was to monitor the fall armyworm resistance development to Novaluran 10% EC in populations obtained from several maize production districts of Tamil Nadu. Studies on the resistance levels of the freshly built FAW in Tamil Nadu settings will help stronger and more effective IPM decision management systems. In this regard, attempts were made to determine the resistance levels of *S. frugiperda* against Novaluran 10% EC in maize.

2. MATERIAL AND METHODS

2.1 Maintenance of Insect Culture

~~Fourth and fifth instar larvae~~ Field populations of *S. frugiperda* were collected from four diverse geographical locations viz., Coimbatore, Perambalur, Salem, and Theni in Tamil Nadu, India (Fig. 1 and Table 1). ~~Fourth and fifth instar larvae were collected from maize plants using forceps.~~ These larvae were reared in an artificial diet in individual feeding ~~box~~ boxes at TNAU FAW laboratory. They were separated from the containers after pupation and placed into adult emergence ~~stage-cages~~ with 10 per cent sugar solution to aid in oviposition. The egg masses that resulted were grown in ~~an~~ artificial diet and the population was kept under controlled circumstances (25°C, 70% relative humidity, and a 14:10 h light/dark photoperiod) without selection pressure (no pesticide exposure). In the study, the ~~field collected~~field-collected population ~~population~~ will be served as a resistant strain of FAW (RS). Further, ~~the~~ susceptible strain of FAW (SS) received from ~~the~~ FAW laboratory without selection pressure upto 157 generations under similar conditions at the Department of Agriculture Entomology, Tamil Nadu Agricultural University, Coimbatore was utilized to compare the median fatal dose with a resistant strain.

Table 1. Background data for field populations of *Spodoptera frugiperda* collected from different sites

Collected Location	Coordinates	Map No.	Reference	Host plant
Perambalur – Tamilnadu	11.33° N, 78.81° E	1		<i>Zea mays</i>
Salem – Tamilnadu	11.62° N, 78.58° E	2		<i>Zea mays</i>
Coimbatore – Tamilnadu	11.01° N, 76.93° E	3		<i>Zea mays</i>

Formatted: Font: Italic

Formatted: Font: Italic

Formatted: Font: Italic

Entomology, TNAU. From the 151st, no selection pressure (or pesticide exposure) was applied until the Fn generation. Based on the doses indicated by the preliminary range finding test, bioassays were performed for the susceptible population. A discriminating dose was determined based on the LC₅₀ and LC₉₅ value obtained for the 'n' generation for susceptible population.

2.5 Statistical Analysis

Finney's probit analysis was used to establish the median lethal doses (LC₅₀) of the pesticide selected, which were validated in POLOPLUS software version 2.0. Susceptibility indices were calculated using the LC₅₀ and LC₉₅ values obtained from the last generation that was not exposed to pesticides. The Susceptibility Index (SI) is the ratio of the first generation LC₅₀ or LC₉₅ to the final generation LC₅₀ or LC₉₅. Regupathy and Dhamu (2001) [16] determined the rate of resistance drop (R) and the number of generations necessary for a ten-fold fall in LC50 value (G).

$$R = \frac{\text{Log(Final LC50)} - \text{Log (initial LC50)}}{n}$$

$$G = 1/R$$

$$\text{ID value} = \left[\frac{\text{Slope of last Generation}}{\text{Slope of first generation}} - 1 \right] \times 100$$

$$RR = \frac{\text{LC50 of resistant Strain (RS)}}{\text{LC50 of susceptibility strain (RR)}}$$

2.5 Monitoring the Insecticide Resistance

A diluted insecticide based on the concentration of the discriminating dose of Novaluran-(0.746 ppm) was applied to the surface of artificial diet in 24 wells using the diet overlay bioassay method against the larval population collected from four fields in Coimbatore, Salem, Perambalur, and Theni.

$$RP = (100 - CM)SE$$

The technique given by (Abbott,1925) [17] was used to calculate the corrected mortality (CM) and standard error (SE).

3. RESULTS AND DISCUSSION

The log concentration probit mortality lines (lcpm) were built for the population of fall armyworm moths obtained from maize fields and raised up to F157 generations without insecticide exposure in FAW laboratory, Department of Agricultural Entomology, Tamil Nadu Agricultural University using baseline data for test insecticide of Novaluran 10% EC. Table 3 shows the LC₅₀ and LC₉₅ values of Novaluran 10% EC against *S. frugiperda* determined by diet overlay bioassay for F151, F152, F153, F154, F155, F156, and F157 generations.

3.1 Baseline susceptibility

The median LC₅₀ and LC₉₅ values for the F151 population against Novaluran were 0.832 ppm and 2.134 ppm, respectively. The median LC₅₀ and LC₉₅ values for the F157 population for Novaluran was 0.746 ppm and 1.451 ppm. The LC₅₀ and LC₉₅ values were observed to be lowering with following generations and stabilizing for F156 and F157 generations, indicating that susceptibility increased with succeeding generations.

The calculated LC₅₀ and LC₉₅ values of novaluran revealed that susceptibility steadily increased with successive generations from F151 to F157 (LC₅₀: 0.832 to 0.746 ppm) and (LC₉₅: 2.134 to 1.451 ppm). After F157 generation, the susceptibility index of novaluran was 1.115 and 1.470 ppm. Resistance drop (R) of novaluran was -0.009. A negative R value suggested that susceptibility increased with successive generations. The number of generations needed for a 48-fold drop in LC₅₀ was 108 (Table 2). Based on the baseline toxicity values obtained for the F157th generation of fall armyworm moth reared in insecticide-free conditions, a preliminary discriminating dose (DD) of 1.451 ppm novaluran was determined. Based on the existing range data, a tentative discriminating dosage of 1.45 was determined to be served for detecting novaluran resistance in field populations of Tamil Nadu, India, including Coimbatore, Perambalur, Salem, and Theni.

According to Vinothkumar Bojan et al. (2023), the LC₅₀ value of novaluran 10% EC is 0.91ppm in field collected population in Coimbatore, India [18]. In Israel, Rami Horowitz et al. (2022) found that the LC₅₀ value of novaluran 5% SG is 0.06ppm in susceptible population [19]. According to Rebeca Gutiérrez-Moreno (2019), the Puerto Rican population displayed extraordinary field-evolved resistance to triflumuron (20-fold), with an LC₅₀ of 0.08ppm [20]. Thirawut et al. (2023) revealed that in Southeast Asian nations, the LC₅₀ value of a field population of 5 locations against Lufenuron 5% EC is TM2019 (2.359), SN2019 (2.267), SP2021 (4.558), TL2021 (1.034), KC2022 (0.259), WS2022 (0.209) and the resistance co-efficient values indicated that *S. frugiperda* developed low resistance to only one pesticide (lufenuron) among the other pesticides [21].

Formatted: Font: Italic

Table 2. Baseline susceptibility of *S. frugiperda* to Novaluran 10% EC by Diet overlay bioassay method

Generation	Chisquare (Σ 2)	Slope	LC50 (ppm)	Fiducial Limit		LC95 (ppm)	Fiducial Limit	
				LL	UL		LL	UL
F151	0.114	4.204	0.865	0.786	0.952	2.125	1.444	3.126
F152	0.224	4.235	0.847	0.769	0.934	2.088	1.423	3.062
F153	0.153	4.275	0.783	0.705	0.870	1.911	1.354	2.696
F154	0.445	4.400	0.774	0.697	0.859	1.850	1.334	2.565
F155	0.316	4.624	0.768	0.695	0.850	1.755	1.310	2.351
F156	0.807	5.619	0.749	0.685	0.819	1.498	1.215	1.845
F157	0.923	6.037	0.746	0.683	0.806	1.451	1.162	1.813

LL – Lower Limit; UL – Upper Limit

Table 3. Susceptibility Index of *S. frugiperda* to Novaluran 10% EC

Generation	LC50 (ppm)	LC95 (ppm)	Susceptibility Index		Rate of Resistance Decline		Slope function I/D %
			LC50	LC95	R	G	
F151	0.865	2.125	1.159	1.464	-0.009	-108.903	43.571
F157	0.746	1.451	1.000	1.000			

Table 4. Resistance Ratio of Novaluran 10% EC to different locations of *S. frugiperda*

Location	N	Regression Equation	LC50 (ppm)	Fiducial Limit		LC50 of susceptible Population (ppm)	Resistance Ratio
				LL	UL		
Coimbatore	180	$y=5.108+3.465x$	0.930	0.827	1.047	0.746	1.246
Salem	180	$y=5.131+3.373x$	0.908	0.812	1.091	0.746	1.217
Perambalur	180	$y=5.282+4.954x$	0.878	0.800	0.953	0.746	1.176
Theni	180	$y=5.494+4.874x$	0.792	0.722	0.868	0.746	1.061

3.2 Resistance Ratio

The bioassay was carried out against *S. frugiperda* field populations collected in from different District of Tamil Nadu (Coimbatore, Perambalur, Salem, and Theni). For the test insecticides (Novaluran 10% EC), log concentration probit mortality (lcpm) lines were fitted to resistance populations acquired across sites. The median lethal concentration (LC50) values for each region's F1 *S. frugiperda* generation were computed.

The LC50 values in ppm for the Coimbatore, Salem, Perambalur, and Theni populations in Novaluran 10% EC were 0.930, 0.908, 0.878, and 0.792, respectively. The resistance ratios (RRs) were determined using the susceptible population's LC50 (0.746 ppm) and revealed a 1.176 (Perambalur), 1.217 (Salem), 1.246 (Coimbatore), and 1.061 (Theni) fold increase in resistance as compared to the susceptible population (Table 4).

4. CONCLUSION

The current study unveiled that populations of the *S. frugiperda*, gathered from distinct maize cultivar regions in Tamil Nadu namely Perambalur, Salem, Coimbatore,

and Theni, exhibited variations in their sensitivity to Novaluran. These differences were attributed to factors like changes over time, geographical diversity, varying response to the chemical's toxicity, the dosage employed, and the manner in which the test insecticide was used. In comparison to the population from Salem, Perambalur, and Theni, the Coimbatore sample shown greater resistance to Novaluran 10% EC.

REFERENCES

1. Pogue, Michael. "World revision of the genus *Spodoptera* Guenée." (2002).
2. Tambo, Justice A., Monica K. Kansiime, Ivan Rwomushana, Idah Mugambi, Winnie Nunda, Catherine Mloza Banda, Shingirayi Nyamutukwa, Fernadis Makale, and Roger Day. "Impact of fall armyworm invasion on household income and food security in Zimbabwe." *Food and energy security* 10, no. 2 (2021): 299-312.
3. Ramasamy, Maruthadurai, Bappa Das, and R. Ramesh. "Predicting climate change impacts on potential worldwide distribution of fall armyworm based on CMIP6 projections." *Journal of Pest Science* 95, no. 2 (2022): 841-854.
4. Li, Xi-Jie, Ming-Fei Wu, Jian Ma, Bo-Ya Gao, Qiu-Lin Wu, Ai-Dong Chen, Jie Liu et al. "Prediction of migratory routes of the invasive fall armyworm in eastern China using a trajectory analytical approach." *Pest Management Science* 76, no. 2 (2020): 454-463.
5. Bird, Lisa, Melina Miles, Adam Quade, and Helen Spafford. "Insecticide resistance in Australian *Spodoptera frugiperda* (JE Smith) and development of testing procedures for resistance surveillance." *Plos one* 17, no. 2 (2022): e0263677.
6. Andrews, Keith L. "Latin american research on *Spodoptera frugiperda* (Lepidoptera: Noctuidae)." *Florida entomologist* (1988): 630-653..
7. Goergen, Georg, P. Lava Kumar, Sagnia B. Sankung, Abou Togola, and Manuele Tamò. "First report of outbreaks of the fall armyworm *Spodoptera frugiperda* (JE Smith)(Lepidoptera, Noctuidae), a new alien invasive pest in West and Central Africa." *PloS one* 11, no. 10 (2016): e0165632.

8. Day, Roger, Phil Abrahams, Melanie Bateman, Tim Beale, Victor Clotey, Matthew Cock, Yelitza Colmenarez et al. "Fall armyworm: impacts and implications for Africa." *Outlooks on Pest Management* 28, no. 5 (2017): 196-201.
9. Prasanna, B. M., J. E. Huesing, Regina Eddy, and V. M. Peschke. "Fall armyworm in Africa: a guide for integrated pest management." (2018)..
10. Bhusal, Kiran, and Kamana Bhattarai. "A review on fall armyworm (*Spodoptera frugiperda*) and its possible management options in Nepal." *Journal of Entomology and Zoology Studies* 7, no. 4 (2019): 1289-1292.
11. Kalleshwaraswamy, C. M., M. S. Maruthi, and H. B. Pavithra. "Biology of invasive fall army worm *Spodoptera frugiperda* (JE Smith)(Lepidoptera: Noctuidae) on maize." *Indian Journal of Entomology* 80, no. 3 (2018): 540-543.
12. Srikanth, J., N. Geetha, B. Singaravelu, T. Ramasubramanian, P. Mahesh, L. Saravanan, K. P. Salin, N. Chitra, and M. Muthukumar. "First report of occurrence of fall armyworm *Spodoptera frugiperda* in sugarcane from Tamil Nadu, India." *Journal of Sugarcane Research* 8, no. 2 (2018): 195-202.
13. Casmuz, Augusto, M. Laura Juárez, M. Guillermina Socías, M. Gabriela Murúa, Silvina Prieto, Santiago Medina, Eduardo Willink, and Gerardo Gastaminza. "Revisión de los hospederos del gusano cogollero del maíz, *Spodoptera frugiperda* (Lepidoptera: Noctuidae)." *Revista de la Sociedad Entomológica Argentina* 69, no. 3-4 (2010): 209-231.
14. Chao, Wu, Zhang Lei, Liao Chongyu, Wu Kongming, and Xiao Yutao. "Research progress of resistance mechanism and management techniques of fall armyworm *Spodoptera frugiperda* to insecticides and Bt crops." *Plant Diseases and Pests* 10, no. 4 (2019): 10-17.
15. Ahmad, Mushtaq, and Sanobar Gull. "Susceptibility of armyworm *Spodoptera litura* (Lepidoptera: Noctuidae) to novel insecticides in Pakistan." *The Canadian Entomologist* 149, no. 5 (2017): 649-661.
16. Regupathy, A., and K. P. Dhamu. *Statistics work book for insecticide toxicology*. Suriya Desktop Publishers, 1990.

17. Abbott, Walter S. "A method of computing the effectiveness of an insecticide." *J. econ. Entomol* 18, no. 2 (1925): 265-267.
18. Bojan, Vinothkumar, G. Arulkumar, T. Srinivasan, P. S. Shanmugam, V. Baskaran, A. Suganthi, S. Jeyarani, S. V. Krishnamoorthy, N. Muthukrishnan, and N. Sathiah. "Acute and Persistent Toxicity of Newer Insecticide Molecules Against Invasive Pest of Maize, Fall Armyworm Spodoptera frugiperda (JE Smith)." *Madras Agricultural Journal* 110, no. march (1-3) (2023): 1.
19. Horowitz, a. Rami, carolina guzmzn, dganit sadeh, lilach lily mondaca, rui shi, and shlomo sarig. "insecticide resistance management for fall armyworm in maize fields of israel." *agrofor international journal* 7, no. 2 (2022).
20. Gutiérrez-Moreno, Rebeca, David Mota-Sanchez, Carlos A. Blanco, Mark E. Whalon, Henry Terán-Santofimio, J. Concepcion Rodriguez-Maciel, and Christina DiFonzo. "Field-evolved resistance of the fall armyworm (Lepidoptera: Noctuidae) to synthetic insecticides in Puerto Rico and Mexico." *Journal of economic entomology* 112, no. 2 (2019): 792-802.
21. Thirawut, Supangkana, Woravit Sutjaritthammajariyangkun, Artit Rukkasikorn, Pruetthichat Punyawattoe, Uraporn Noonart, and Youichi Kobori. "Pesticide susceptibility monitoring of fall armyworms (Spodoptera frugiperda (JE Smith)): a simple methodology for information-sharing among Southeast Asian countries." *CABI Agriculture and Bioscience* 4, no. 1 (2023): 1-8.