Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2017; 5(5): 1445-1449

Received: 06-07-2017
Accepted: 07-08-2017

Mahantesh Kapasi
Assistant Professor of Entomology, Agricultural College Kalaburagi, UAS, Raichur, Karnataka, India

Bheemanna M
Professor and Head, PRFQAL, UAS, Raichur, Karnataka, India

Guruprasad GS
Scientist, ARS, Gangavati, UAS, Raichur, Karnataka, India

Vijaykumar Ghante
Scientist, MARS, UAS, Raichur, Karnataka, India

Correspondence
Mahantesh Kapasi
Assistant Professor of Entomology, Agricultural College Kalaburagi, UAS, Raichur, Karnataka, India

Baseline susceptibility of sulfoxaflor 24 SC insecticide on paddy brown planthopper, (*Nilaparvata lugens*, Stal) population of Northeastern Karnataka

Mahantesh Kapasi, Bheemanna M, Guruprasad GS and Vijaykumar Ghante

Abstract
Baseline susceptibility of sulfoxaflor 24 SC insecticide was investigated by collecting field populations of brown planthopper from different locations of northeastern Karnataka during 2014-15 and 2015-16. All the selected populations differed in their susceptibility to sulfoxaflor. In general, Gangavati and Sindhanur BPH population recorded higher LC_{50} values of 29.95 and 27.75 ppm respectively, followed by Ballari (26.16 ppm), Manvi (25.03 ppm) and Devadurga (22.68 ppm). Lowest LC_{50} value was observed in population collected from Kembhavi (21.56 ppm) during 2014-15. The similar trend was noticed during 2015-16 season. The comparison studies were made with dinotefuran 20 SG and buprofezin 25 SC insecticides for a population collected from Gangavati.

Keywords: *Nilaparvata lugens*, Insecticide resistance, baseline susceptibility and sulfoxaflor

Introduction
The brown planthopper (BPH), *Nilaparvata lugens* (Stål) (Hemiptera: Delphacidae) is continuing to be a serious pest of rice in Asia. In 1927 it was first time reported as sporadic pest on rice crop of Guntur district in Andhra Pradesh, India [1]. A considerable loss in yields due to this pests were also reported in West Bengal [2], Uttar Pradesh [3, 4] and Punjab [5] of India. BPH regarded as a endemic pest of rice in Tungabhadra and Cauvery command areas of Karnataka, India [6].

BPH suck sap directly from growing plants, and the affected plants become chlorotic, and the leaves dry up gradually, leading to the death of plants. This feeding damage is commonly referred as 'hopper burn'. BPH also act as a vector of rice grassy stunt virus and rice ragged stunt virus. The loss in grain yield ranges from 10 to 70 per cent due to infestation of BPH [7]. Outbreak of this pest often leads to total loss of the rice crop, if no effective control measures were taken up. The management of this pest has always been emphasized and largely relied on insecticides as a first line of defense in spite of their drawbacks [8, 9]. Insecticide is the only tool that is reliable for emergency action when insect pest population exceeds the economic threshold. Several potent insecticides have been recommended for managing the sucking pests, but the arbitrary use of insecticides has resulted in the development of resistance in insects to insecticides, resurgence, secondary pest outbreaks, disruption of the natural enemy complex, loss in biodiversity and environmental pollution [10]. Paddy BPH found to have developed resistance to the recommended organophosphate and organochlorine insecticides [11, 12]. However, in the recent past field level failure of neonicotinoids and carbamates were also noticed in this pest [13, 14]. Under such circumstances new molecules selectively to target pests are required to be evaluated for the justification of chemical control as the first line of defense. Sulfoxaflor is a new and safer insecticide from a novel, a new class of chemistry known as sulfoximines.

Sulfoxaflor is a systemic insecticide and acts as an insect neurotoxin. It is the only member of a sulfoximines class of chemicals. Sulfoxaflor targets the central nervous system of the insects as a agonist at nicotinic acetylcholine receptors (nAChRs) with much lower toxicity to mammals similarly to neonicotinoids. The sulfoximines are very good in managing wide range of sap-feeding insect pests that are resistant to neonicotinoids class of insecticides.

The sulfoxaflor intoxicated insects symptoms are initially excitatory and include tremors, antennal waving and leg extension or curling, followed by partial or complete paralysis and death. In consideration with these features, Insecticide resistance action committee (IRAC) has placed sulfoxaflor in a mode of action subgroup (Group 4C) that is separate from the neonicotinoids (Group 4A) [15].

As new molecules are developed for use in managing insect pests, it is necessary to develop baseline susceptibility data which would not only help in fixing the dosages for effective management but also in understanding the level of resistance developed by the pest and any possible cross resistance there in, could be assessed in advance. As the information available on the baseline susceptibility of sulfoxaflor molecules against paddy BPH is being limited. Thus, the present study was taken up to assess the susceptibility of BPH populations from different paddy growing locations of northeastern karnataka, India to sulfoxaflor 24 SC insecticide.

Materials and methods

The present investigations were undertaken during 2014-15 and 2015-16 in the Agricultural Research Station, Gangavathi, UAS, Raichur to study the baseline susceptibility of sulfoxaflor 24 SC against the paddy BPH population of major paddy growing areas of Northeastern Karnataka *viz*., Kembhavi, Devadurga, Manvi, Gangavati, Sindhanur and Ballari.

Test insects

The population of brown planthopper was collected from unsprayed farmers field from each selected location during early morning hours in a plastic box containing cut stems of paddy and brought to the laboratory immediately. Collection was made with the help of mouth sucking aspirator and after collection the top of the box was covered with muslin cloth to facilitate aeration.

Mass rearing of the test insect

The BPH susceptible plant variety, IET 18288 (Gangavathi Sanna) was used for experiment. The thirty days old seedlings from trays were transplanted to plastic pots of 60 cm diameter. Five plants per pots were transplanted and the nutrient requirement of transplanted paddy seedlings was met out as per the requirement. The pots transplanted with seedlings were then placed into BPH rearing wooden cages (60 cm x 66.5 cm x 72.5 cm) covered with wire mesh [16]. Four pots per cage were maintained for the rearing and multiplication of BPH culture. These rearing wooden cages were placed in a shaded area. Further, the cage stands were placed in trays with water to prevent ants from entering the cage.

The collected hopper population were brought to the laboratory and carefully transferred to rearing cages containing healthy plants in pots. Two cages for each location were used for rearing and multiplication of collecting population. The cages were examined periodically for the presence of predators and other insect species. Whenever the predators or other species of insects were observed in the cages, they were removed promptly to facilitate development of BPH population. The collected population of different location was reared up to F_1 generation in the laboratory on paddy seedlings.

Test insecticides for bioassay

The test insecticide, sulfoxaflor 24 SC was used for baseline susceptibility studies on paddy brown planthopper populations collected from the different locations and was compared with the dinotefuran 20 SG and buprofezin 25 SC insecticides by using BPH population of Gangavati location. Insecticide solutions were prepared from the formulated products using distilled water. At the initial stage, bracketing or preliminary range-finding tests was done to arrive required concentrations of insecticides.

Bioassay

The third instar nymphs of each location reared in laboratory were selected and exposed to graded concentrations of test insecticide *i.e.*, sulfoxaflor 24 SC, whereas, for bioassay studies of dinotefuran 20 SG and buprofezin 25 SC, only Gangavathi population was used as a comparison. The bioassay method followed for BPH was the whole plant dip bioassay developed and recommended by the Insecticide Resistance Action Committee (IRAC) method No. 5 with slight modifications [17].

Fifteen to twenty days old seedlings raised in small cups of 10 centimeter diameter were used for bioassay. The required concentrations of test insecticide were prepared freshly for one liter volume. Small cups containing fifteen to twenty days old seedlings were inverted and seedlings were dipped in the prepared solution completely for 10 seconds. It was ensured that the all parts of plants were in contact with the test solution. Three replicates were maintained for each concentration of insecticide along with water treated as a control. After dipping, cups were reverted and seedlings were allowed to dry for 10 - 15 minutes. Immediately after drying, plants along with cups were placed into transparent plastic circular tubes. Uniform sized ten third instar nymphs collected from rearing cages of a particular location using an aspirator were introduced into each treated plants in plastic tube and retained using muslin cloth tied with rubber band on top. The treated insects were maintained at room temperature.

The mortality of hoppers was recorded at 72 hours after the treatment. The nymphs were considered dead if they were unable to show movement after gentle prodding with a fine brush.

Analysis

Percentage of mortality for each concentration of test insecticide and control were computed and corrected per cent mortality was calculated by using Abbott's formula [18]. Whenever, the mortality in control exceeded 20 per cent, the experiment was repeated once again. The corrected mortality data of each test insecticide of each location was subjected to probit analysis using EPA probit analysis program version 1.5 for calculation of LC_{50} and LC_{90} values. The bioassay studies were conducted in two cropping seasons *i.e.,* 2014-15 and 2015-16 for all seven district populations against the test insecticides.

Results and Discussion

The results are presented insecticide wise for the year 2014-15 and 2015-16.

Susceptibility of paddy BPH populations to sulfoxaflor 24 SC insecticide

The data on the LC_{50} values of sulfoxaflor to different geographic populations of *N. lugens* for two years are presented in the Tables 1 and 2. The results indicated that there has been marked difference in LC_{50} values among the different location populations.

During 2014-15, the median lethal concentrations of sulfoxaflor to six field populations of paddy BPH ranged from 21.56 to 29.95 ppm. Gangavati population recorded a maximum LC_{50} value (29.95 ppm), followed by the population collected from Sindhanur (27.75 ppm), Ballari (26.16 ppm), Manvi (25.03 ppm) and Devadurga (22.68 ppm). Lowest LC_{50} value was observed in population collected from Kembhavi (21.56 ppm). The LC_{90} values followed the similar trend as that of LC_{50} values obtained during 2014-15 (Table 1).

During 2015-16, also similar trend of median lethal concentrations of sulfoxaflor was noticed for six field populations of BPH as observed during 2014-15 with range from 22.39 to 31.83 ppm. Maximum LC_{50} value of 31.83 ppm was obtained with Gangavathi population and was followed by the population collected from Sindhanur (29.99 ppm), Ballari (27.30 ppm), Manvi (26.25 ppm) and Devadurga (24.22 ppm). Lowest LC_{50} value was observed in population collected from Kembhavi (22.39 ppm). (Table 2). The literatures pertaining to the susceptibility of BPH populations to sulfoxaflor insecticide are limited.

Present findings of sulfoxaflor are in contradicting with the results of Ghosh *et al* [19] who reported a comparatively lower LC_{50} values of sulfoxaflor (0.382 ppm to 2.986 ppm) to selected BPH populations of West Bengal. The variations in the results may be due to a number of factors *viz*., geographical location of the insect population collected, variation in the bioassay methodology and generation of the insect population subjected to study.

In areas like Gangavati and Sindhanur, the rice crop is grown both during *Kharif* and *Rabi* seasons with high intensive agronomic inputs. The use of pesticide is also much higher compared to the usage in other rice growing areas of Karnataka. As the insecticide resistance development is a selection process, the level and rate of development of resistance are determined by the frequency of application of insecticides [20]. Therefore, BPH populations in areas around Gangavati and Sindhanur, which are subjected to greater selection pressure, were expected to possess higher levels of resistance to insecticides compared to the populations from Kembhavi, Devadurga, Manvi and Ballari areas. It was evident from the results that BPH populations from different regions of northeastern Karnataka differed significantly in their response to sulfoxaflor insecticides. Apart from the frequency of applications, the insecticide usage pattern, cropping pattern in different regions, and even the genetic variation in populations which are widely separated might have contributed to the observed variations in responses of BPH populations to sulfoxaflor insecticide. The significant intra-regional variation in susceptibility of different populations has been reported in Taiwan [21], Japan [22], Korea [23], United Kingdom [24] and India [25-27].

Susceptibility of paddy BPH population to dinotefuran 20 SG and buprofezin 25 SC insecticides

LC_{50} value of dinotefuran 20 SG against population collected from Gangavati was 35.48 ppm and 39.88 ppm during 2014-15 and 2015-16, respectively. However, the LC_{50} value of buprofezin 25 SC against population collected from Gangavati was 108.54 ppm 124.83 ppm during 2014-15 and 2015-16, respectively (Table 3). The LC_{50} values of dinotefuran 20 SG and buprofezin 25 SC in the present study were considerably higher as compared to earlier reports of Basanth *et al* [28] who reported the 3.499 ppm (dinotefuran) and 3.116 ppm (buprofezin) for Gangavati population. The continuous and indiscriminate use of insecticides year by year in paddy crop might be resulted in increased LC_{50} values in our studies.

Though the dinotefuran 20 SG is a relatively new neonicotinoid compound and it is not commonly used by the farmers even in Gangavati areas, the observed resistance to this could be attributed to the cross resistance from imidacloprid which is extensively used in these areas which need to be studied. Reports of Wang *et al* [29] and Zewen *et al* [30] suggested that the resistant strain selected with imidacloprid showed substantial cross-resistance to imidaclothiz, thiacloprid and acetamiprid, and slight levels of cross resistance to dinotefuran and thiamethoxam.

Buprofezin, a chitin synthesis inhibitor has been used continuously by the farmers in suppression of BPH for more than five years in areas around Gangavati [31]. Present results indicated that the population of BPH (Gangavati) has acquired a high level of resistance to buprofezin. The observed variation in resistance level could be because of the selection pressures in these areas.

All the field populations of paddy BPH had shown considerable variation in their susceptibility to sulfoxaflor (Figure 1 & 2). In general BPH population of Gangavati and Sindhanur recorded a comparatively higher LC_{50} values, while, populations of Ballari, Manvi, Devadurga and Kembhavi recorded lower LC_{50} values to sulfoxaflor. The present study clearly indicated that sulfoxaflor insecticide had a higher sensitivity and better performance on studying sucking insect pest as compared to other insecticides used in investigations. Hence, sulfoxaflor can be used as a component in integrated resistance management (IRM) approach for the management of paddy BPH insect pest.

Table 1: Log dose probit analysis of sulfoxaflor 24 SC on paddy BPH (*Nilaparvata lugens*) during 2014-15

Location	LC_{50} (ppm)	Fiducial limits		LC_{90} (ppm)	Fiducial limits		Slope	$\chi 2$
		Lower	Upper		Lower	Upper		
Kembhavi	21.56	15.59	29.83	90.14	61.37	113.81	2.18	2.83
Devadurga	22.68	17.00	30.24	93.66	67.09	125.18	2.34	1.12
Manvi	25.03	19.35	32.36	96.41	68.56	128.10	2.51	1.01
Gangavati	29.95	23.30	38.50	110.21	72.86	161.31	2.31	1.46
Sindhanur	27.75	22.00	35.02	100.27	72.08	143.36	2.25	0.38
Ballari	26.16	19.97	34.28	98.18	69.61	135.53	2.60	0.93

ppm – Parts per million

Table 2: Log dose probit analysis of sulfoxaflor 24 SC on paddy BPH (*Nilaparvata lugens*) during 2015-16

Location	LC_{50} (ppm)	Fiducial limits		LC_{90} (ppm)	Fiducial limits		Slope	$\chi 2$
		Lower	Upper		Lower	Upper		
Kembhavi	22.39	16.22	30.91	100.38	69.54	148.21	1.99	2.15
Devadurga	24.22	18.18	32.28	106.71	70.09	163.41	2.31	2.26
Manvi	26.25	19.65	35.08	118.21	76.41	175.08	2.10	1.70
Gangavati	31.83	24.16	41.94	148.02	95.35	245.41	1.98	1.84
Sindhanur	29.99	23.00	39.12	141.38	87.15	231.46	1.86	1.04
Ballari	27.30	19.94	37.37	129.91	83.80	193.26	2.15	2.66

ppm – Parts per million

Table 3: Log dose probit analysis of dinotefuran 20 SG and buprofezin 25 SC on paddy BPH (Gangavathi)

2014-15 season								
Insecticide	LC_{50} (ppm)	Fiducial limits		LC_{90} (ppm)	Fiducial limits		Slope	$\chi 2$
		Lower	Upper		Lower	Upper		
Dinotefuran 20 SG	35.48	27.91	45.09	120.76	80.28	170.56	2.53	0.34
Buprofezin 25 SC	108.54	92.65	127.16	260.61	200.41	360.33	3.34	0.30
2015-16 season								
Insecticide	LC_{50} (ppm)	Fiducial limits		LC_{90} (ppm)	Fiducial limits		Slope	$\chi 2$
		Lower	Upper		Lower	Upper		
Dinotefuran 20 SG	39.88	32.42	49.05	122.21	81.09	173.41	2.67	0.81
Buprofezin 25 SC	124.83	102.84	167.85	383.03	282.47	514.30	2.85	1.31

ppm – Parts per million

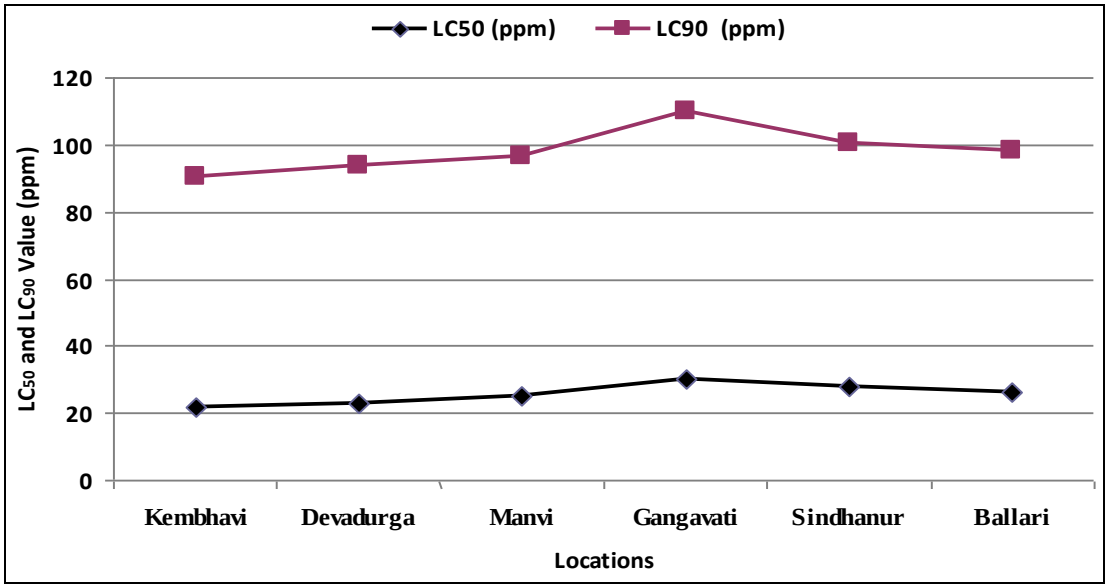

Fig 1: Susceptibility of paddy BPH (*N. lugens*) populations to sulfoxaflor 24 SC insecticide during 2014-15

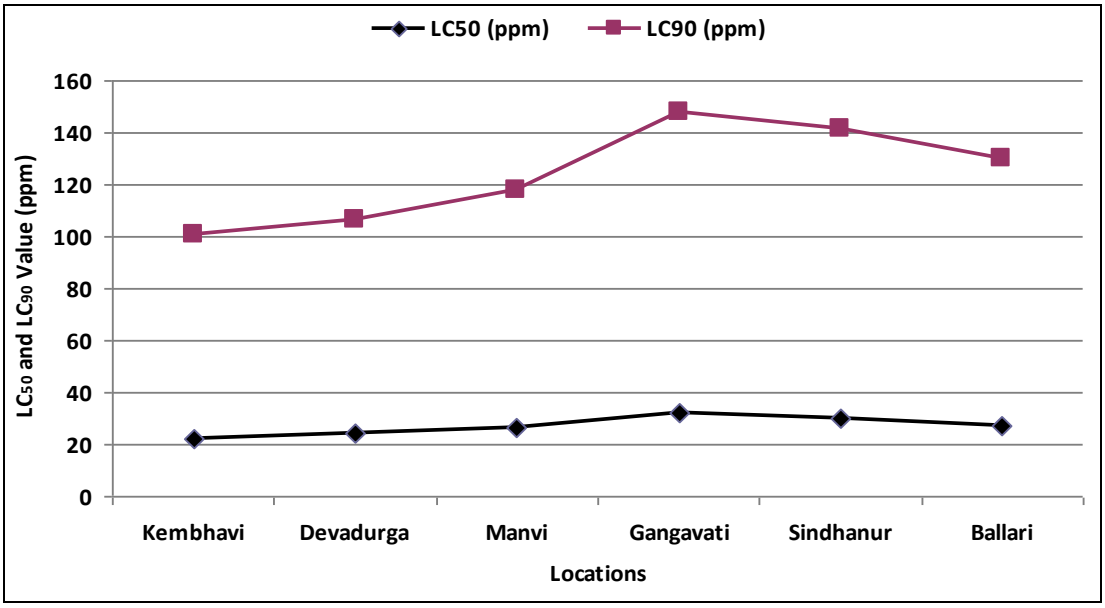

Fig 2: Susceptibility of paddy BPH (*N. lugens*) populations to sulfoxaflor 24 SC insecticide during 2015-16

Acknowledgements
The facilities provided by the Department of Agricultural Entomology, Main Agricultural Research Station, UAS, Raichur, where the investigation was carried out and the funding as well as test insecticide received from Dow Agro Sciences, PVT. Ltd, Mumbai, is gratefully acknowledged.

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