# SUSCEPTIBILITY OF IMIDACLOPRID RESISTANT WHITEFLY BEMISIA TABACI (GENNADIUS) TO CYANTRANILIPROLE

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### ABSTRACT

The susceptibility of five north Indian populations of *Bemisia tabaci* (Genn.) against a neonicotinoid insecticide, imidacloprid and a comparatively novel, diamide insecticide cyantraniliprole has been analysed in this study. It was found that the Sriganganagar and Bathinda populations were moderately resistant to imidacloprid with  $LC_{50}$  values 845.55 and 765.65 mg/ l, respectively; while, the  $LC_{50}$  value for cyantraniliprole was around 4.5 and 4.3 mg/ l for both the populations. The relative resistance ratio for imidacloprid was 14.53 and 13.16 for Sriganganagar and Bathinda populations, respectively, whereas, for cyantraniliprole, no resistance was observed. The pairwise comparisons of  $LC_{50}$  so f various populations did not exhibit any cross-resistance. The results of the study demonstrate the possibility of using cyantraniliprole as an alternative for imidacloprid resistant *B. tabaci* populations in north India.

Key words: *Bemisia tabaci*, cross-resistance, imidacloprid, cyantraniliprole,  $LC_{50}$ , north India, susceptibility, toxicity, Sriganganagar, Bathinda, relative resistance

Bemisia tabaci (Gennadius) is a serious pest in tropical, subtropical and low altitude temperate regions of the world (Ran et al., 2018). Its polyphagous nature, high reproductive potential, and the vectoring capacity of B. tabaci necessitate effective management (Horowitz and Ishaaya, 1996). Existence of a wide genetic variability, confirmed *B. tabaci* as a cryptic species complex consisting of 44 genetic groups at present (Kanakala and Ghanim, 2019). The persistence of this pest has led to widespread adoption of newer chemicals (Elbert et al., 2008). Several neonicotinoid insecticides namely imidacloprid, acetamiprid, nitenpyram, thiamethoxam, clothianidin etc. were developed to control hemipteran pests across the world (Bass et al., 2015). The systemic and translaminar properties of these neonicotinoids have enhanced their use in managing sucking insect pests (Horowitz et al., 1998). Though imidacloprid was the first neonicotinoid to be found effective against B. tabaci (Nauen and Denholm, 2005), its non-judicious and unilateral use has led to the development of resistance, which is a predictable outcome of insecticide use in the field (Kranthi, 2005). Severe infestation and outbreaks of B. tabaci, as well as several instances of resistance to imidacloprid from from India, since the last two decades (Sethi and Dilawari, 2008; Naveen et al., 2017). Among the 132 cases of imidacloprid resistance registered in the Arthropod Pesticide Resistance Database (APRD), 110

are field evolved resistance (Mota-Sanchez and Wise, 2020). Thus, it became necessary to bring an alternative to this neonicotinoid insecticide for managing *B. tabaci* in the field. An anthranilic diamide insecticide, cyantraniliprole with a novel mode of action, activating ryanodine receptors and thus the calcium channel activation, has targeted hemipteran insects (Cordova et al., 2006; Sattelle et al., 2008) and found effective with an acute toxicological profile (Ran et al., 2018). In this study, the susceptibility of *B. tabaci* populations from six locations in north India against the neonicotinoid, imidacloprid and the diamide, cyantraniliprole is evaluated.

#### MATERIALS AND METHODS

*Bemisia tabaci* populations were collected from the cotton fields from six locations in north India and reared on cotton plants (*Gossypium hirsutum* L.) in Insect Proof Climate Control Chamber (IPCCC), Division of Entomology, IARI (27±2°C, 60-70% RH, photoperiod 14:10 L:D). The whiteflies collected in 2013 from *Leucaena leucocephala* from Pusa campus, under laboratory condition, served as the susceptible check. The details of the *B. tabaci* populations collected are New Delhi (28.64°N 77.17°E), Bathinda (30.20°N 74.95°E), Indore (22.8°N 75.73°E), Hisar (29.09°N 75.87°E), and Sriganganagar (29.52°N 74.78°E). Commercial formulations of insecticides, imidacloprid (Confidor, Bayer Crop Science) and cyantraniliprole (Benevia, DuPont) were used for bioassay studies. The insecticides procured from the market were diluted with deionized water to make 1% stock solution and serial dilutions were prepared. Seven concentrations with three replications were set and susceptibility studies were carried out following the modified Insecticide Resistance Action Committee (IRAC) protocols.

The lethal effects of imidacloprid and cyantraniliprole were tested following the leaf dip bioassay, modified IRAC method by Naveen et al., (2017). The concentrations of insecticides in mg/l were prepared from 1% stock solutions and water was kept as an untreated control. Cotton leaves from thirty to forty days old plants, which were grown without any infestation were used. Leaves were given a slanting cut at the petiole keeping a length of about 1 cm, completely immersed in the insecticide solutions for 20 sec, and then air-dried (30-45 min). The dried leaves were then kept in the 2% agar slants in petriplates. Using an aspirator, adult whiteflies were collected and briefly anaesthetized by CO<sub>2</sub> for 10-15 sec. The insects were transferred on to the treated leaves in petriplate and mortality was recorded after 72 hr. Insects with no sign of movement was recorded as dead. The estimates of lethal concentrations and 95% confidence intervals were determined for adult bioassay by log- dose probit analysis using PoloPlus 2 (LeOra Software, Petaluma, CA). Relative toxicity of insecticides was estimated following Dhole et al. (2017).

## **RESULTS AND DISCUSSION**

The susceptibility studies were carried out keeping the laboratory population of *B. tabaci* as the baseline. The median lethal concentration of imidacloprid and cyantraniliprole is given in Table 1. In all the bioassays, mortality in control was <7%. The LC<sub>50</sub> value of neonicotinoid for the susceptible population is estimated as 58.19 mg/ l. The relative resistance ratio of imidacloprid is highest in Sriganganagar (14.53) followed by Bathinda (13.16) populations with  $LC_{_{50}}$  values 845.852 mg/ 1 and 765.654 mg/ l, respectively. The LC<sub>50</sub> value of imidacloprid for all the populations is significantly different from the susceptible one considering the overlap of 95% fiducial limits. Moreover, the LC<sub>50</sub> value for New Delhi one (120.173 mg/l) is also significantly different. The results suggested the existence of a high amount of heterogeneity in response of each population to imidacloprid. The susceptibility of populations to imidacloprid based on the  $LC_{50}$  values are in the order of New Delhi >Indore>Hisar>Bathinda>Sriganganagar.

Log dose-probit mortality data displayed susceptibility of all populations against cyantraniliprole. The populations with maximum resistance to imidacloprid viz., Sriganganagar and Bathinda, have shown high susceptibility to cyantraniliprole (LC<sub>50</sub> -4.575 and 4.308 mg/l, respectively); the highest level of susceptibility was in Indore population ( $LC_{50}$  - 3.320 mg/l); based on  $LC_{50}$  values, susceptibility is in the order: Indore>Hisar>Bathinda>Sriganganagar>New Delhi. The relative resistance factor of none of the populations has crossed 2.5, indicating no resistance development (Table 1). High susceptibility of B. tabaci populations towards cyantraniliprole is clear from relative susceptibility. When relative toxicity is compared for the two insecticides, cyantraniliprole was 32x more toxic than imidacloprid in the case of laboratory susceptible population itself. Further, the relative toxicity was higher for cyantraniliprole by 185 and 177x in Sriganganagar and Bathinda populations than the neonicotinoid insecticide, imidacloprid. Relative resistance ratios are used to know the resistance status of each population to respective insecticides (Table 4)- and those ranging between  $5.0 < RR \le 10.0$ is categorized as low level of resistance,  $(10.0 < RR \le$ 40.0) as moderate level, and RR  $\geq$ 40.0 as a high level of resistance (Liu et al., 2010). In the present study, Sriganganagar (14.53) and Bathinda (13.16) populations showed a moderate level of resistance whereas Hisar population (RR-7.24) showed a low level of resistance against imidacloprid (Table 4). Paired comparisons of the log LC<sub>50</sub>s of insecticides tested showed no correlation between imidacloprid and cyantraniliprole (r=0.671) indicating no sign of cross-resistance.

Neonicotinoids have been playing a major role in managing sucking pests for more than two decades (Bass et al., 2015); because of its extensive use in a large number of crops, researchers had reported >1000fold resistance (Nauen and Denholm, 2005). A recent study from India reported a resistance factor of 17 for Sriganganagar and 8 for Bathinda population against imidacloprid (Naveen et al., 2017); this study observed that Sriganganagar and Bathinda as the highest resistant populations with a resistance ratio of 15 and 13 folds, respectively to imidacloprid; and an increase in resistance level of imidacloprid in Sriganganagar and Bathinda population developed in Sriganganagar and Bathinda population was anticipated in comparison to the values given in Naveen et al. (2017), but it was confirmed only in Bathinda population, where the resistance

Insecticide	Population	χ²(df)	Slope	LC <sub>50</sub>	95 % fiducial limits	RR	Resistance
				(mg/ l)	LC <sub>50</sub>	(LC <sub>50)</sub>	status
Imidacloprid	Susceptible	1.506 (5)	$2.175 \pm 0.353$	58.187	46.265 - 70.536 (a)	1	
	New Delhi	2.110 (5)	$2.270 \pm 0.559$	120.173	72.638 - 154.798 (b)	2.06	
	Hisar	1.467 (5)	$2.431{\pm}0.352$	421.138	351.835 - 509.733 (d)	7.24	Low
	Bathinda	2.139 (5)	$2.178{\pm}0.358$	765.654	614.421- 928.253 (e)	13.16	Moderate
	Indore	3.311 (5)	$2.755{\pm}0.384$	280.119	232.187-328.032 (c)	4.81	
	Sriganganagar	3.140 (5)	$2.030{\pm}0.355$	845.852	678.083-1045.485 (e)	14.53	Moderate
Cyantraniliprole	Susceptible	0.854 (5)	$1.971{\pm}0.329$	1.803	1.439 - 2.248 (a)	1	
	New Delhi	3.905 (5)	$1.820{\pm}0.297$	4.897	3.622 - 6.257 (b)	2.7	
	Hisar	2.030 (5)	$1.471{\pm}0.263$	3.896	2.603 - 5.237 (b)	2.161	
	Bathinda	3.449 (5)	$1.893{\pm}0.295$	4.308	3.167 - 5.477 (b)	2.399	
	Indore	2.792 (5)	$1.703{\pm}0.255$	3.320	2.349 - 4.307 (b)	1.841	
	Sriganganagar	2.043 (5)	$1.328{\pm}0.260$	4.575	3.038 - 6.310 (b)	2.537	

 Table 1. Log dose probit mortality data for imidacloprid and cyantraniliprole for different *B. tabaci* populations

factor rose to 13 from eight. The Sriganganagar and Bathinda populations were comparatively susceptible to the diamide insecticide cyantraniliprole with an adult LC<sub>50</sub> value of 4.575 (threefold) and 4.308 mg/ 1(two folds) respectively. Thus, cyantraniliprole showed relatively low susceptibility to New Delhi population  $(LC_{50} = 4.897)$ . The higher  $LC_{50}$  value of the New Delhi population may be due to the use of other diamide insecticides such as flubendiamide, chlorantraniliprole etc. against other insect pests in the cotton and vegetable fields. The polyphagous nature of the whiteflies can support the hypothesis of relatively high LC<sub>50</sub> compared to other populations. A report by Dangelo et al. (2017) in Brazil also hypothesized the same when they observed resistance of B. tabaci against cartap hydrochloride and chlorantraniliprole which were not a part of *B. tabaci* management programme, but for other pests in the same ecosystem.

This baseline information on susceptibility obtained from the study can be used for further monitoring of resistance to cyantraniliprole in field conditions. A baseline data was developed in Florida by Carabello et al. (2013) and they found cyantraniliprole to be viable for whitefly resistance management. The adult bioassays of this study provided pooled  $LC_{50}$  values ranging from 0.037 to 0.064 mg/l for nine field-collected populations in Florida. The lethal effects of cyantraniliprole to B. tabaci was also confirmed from China, where eight insecticides including six neonicotinoids were tested and cyantraniliprole was found as most toxic (Wang et al., 2017). The present study indicates a high susceptibility to cyantraniliprole in B. tabaci populations from north India, which suggests that this chemical is a promising alternative for the insecticide resistance management of imidacloprid. Even though cyantraniliprole does not

possess a label claim against whiteflies in cotton and brinjal in India, it is recommended against whiteflies in tomato and gherkins. Considering the high polyphagous nature, the label claim of newer insecticides with a novel mode of action, the label claim can be extended for cotton making them suitable alternatives.

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