



## ***In vitro* Assessment of Neonicotinoids and Pyrethroids against Tea Mosquito Bug, *Helopeltis antonii* Sign. (Hemiptera: Miridae) on Guava**

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### **Article History**

### **Abstract**

The tea mosquito bug (TMB), *Helopeltis antonii*, is an emerging pest of horticultural crops, specially on guava and moringa. Insecticides are indispensable component for the management of insect pests. Exploration of new molecules with shortest waiting period may pave way for managing TMB in fruit and vegetable crops with nil/low residue. Until now there are no recommended insecticides available under Central Insecticides Board & Registration Committee (CIB&RC) against TMB on guava. In view of the above facts, new molecules with a low waiting period and are recommended by CIB&RC on tea, viz., Clothianidin 50% WDG, Thiacloprid 21.7% SC, Bifenthrin 10% EC, and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, were chosen and evaluated against TMB under *in vitro* condition. Clothianidin 50% WDG recorded the highest mortality of 100.00 per cent at 72 hours after treatment (HAT), and the lowest LC<sub>50</sub> value (0.328 ppm, fiducial limits: 0.144-0.515 ppm) and LT<sub>50</sub> value (10.49 h, fiducial limits: 5.444-14.551 h), followed by Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, Thiacloprid 21.7% SC, and Bifenthrin 10% EC. The results showed that the Clothianidin 50% WDG and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, were highly effective, with the lowest LC<sub>50</sub> and LT<sub>50</sub> values. Since TMB occurs from new flushing to fruiting stage of guava, a minimum of two sprays are mandatory to have quality fruit yield. Hence, application of Clothianidin 50% WDG followed by Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC on need basis will help to reduce the impact of TMB on guava.

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**Keywords: Tea mosquito bug, Guava, Neonicotinoids, Pyrethroids, In vitro assay**

## Introduction

The tea mosquito bug (TMB), *Helopeltis* spp., belongs to the family Miridae is predominantly found in the old-world tropic regions. The first report of TMB was at Java on tea in 1847 (Rao, 1970). There are 41 species of *Helopeltis* identified globally, of which 26 are found in Africa and 15 were observed in the Australasian region (Stonedahl, 1991; Sundararaju and Sundarababu, 1999). In India, three species of TMB, viz., *H. antonii* Signoret, *H. bradyi* Waterhouse, and *H. theivora* Waterhouse, were recorded to cause economic losses on several crops (De Silva, 1957; Stonedahl, 1991; Sundararaju, 1996). *Helopeltis antonii*, *H. bradyi*, and *H. theivora* were first described by Signoret in Sri Lanka during 1858, Waterhouse in Malaya, and Assam during 1886, respectively. In southern India, the first record of *H. antonii* was on neem and guava (Rao, 1915), whereas the outbreak of *H. theivora* was found on tea in 1920 (Rao, 1970). Among them, *H. antonii* is the predominant species (Sundararaju and Bakthavatsalam, 1994; Sundararaju, 1996) and is a major pest of guava, moringa, neem, cashew, tea, cacao, pepper, and cardamom (Aravinthraju *et al.*, 2022). Both nymphs and adults of TMB suck the sap from young shoots, flowers, and fruits/pods, which leads to necrotic lesions causing up to 61.79 per cent fruit loss in guava (Patil and Naik, 2004) and 74 to 100 per cent loss in moringa (Mala *et al.*, 2020). Chemical control plays a pivotal role for the management of TMB. There are no insecticides against TMB on guava and moringa under CIB&RC. However, Bifenthrin 8.0% SC @ 40 a.i./ha, Clothianidin 50% WDG @ 60 a.i./ha, Thiacloprid 21.70% SC @ 90 a.i./ha, Thiamethoxam 25% WG @ 25 a.i./ha, and Thiamethoxam 12.6% + Lambda-cyhalothrin 9.5% ZC @ 33 a.i./ha are recommended by CIB&RC against TMB on tea. In the current study, *In vitro* assays were conducted to assess the efficacy of the above insecticides against TMB on guava, so as to make them available for TMB management on guava.

## Materials and Methods

### Mass culturing of TMB

The mass rearing of TMB, *H. antonii* was carried out as per the protocols of Sundararaju and John (1992). The adults and nymphs were collected from moringa fields, Horticultural College and Research Institute, Periyakulam (10°12' N Latitude and 77°58' E Longitude), Theni, Tamil Nadu, India and introduced into aluminum rearing cages measuring 47.5×47.5×47.5 cm, containing guava (variety: Lucknow 49) shoots of 15 to 20 cm long with young leaves placed in glass jar filled with distilled water. The cage was designed with three closed sides and a closed top, while one side was equipped with a movable lid for convenient access. The guava shoots within the cage were regularly replaced in alternative days, guided by the observation of feeding punctures and drying of shoots. From the culture, the adults were utilized for conducting *in vitro* assays.

### *In vitro* assay

Based on the preliminary range finding test for four insecticides, viz., Clothianidin 50% WDG, Thiacloprid 21.7% SC, Bifenthrin 10% EC, and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, the concentration that gave 30 and 100 per cent mortality were chosen and further dilutions were made to work out the LC<sub>50</sub>. The insecticides, viz., Clothianidin 50% WDG at 0.30, 0.60, 1.20, 2.40, 4.80, and 9.60 ppm; Thiacloprid 21.7% SC at 0.50, 1.00, 2.00, 4.00, 8.00, and 16.00 ppm; Bifenthrin 10% EC at 0.25, 0.50, 1.00, 2.00, 4.00, and 8.00 ppm; and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC at 0.20, 0.40, 0.80, 1.60, 3.20, and 6.40 ppm, were utilized for the assessment of relative toxicity against TMB. For the *in vitro* assay, a transparent mylar film container (30 cm height × 12.5 cm width) was used. Healthy guava shoots (terminal four leaves) were collected from the field, washed thoroughly with distilled water, and air dried. The different concentrations of each insecticide were prepared using distilled water, and sprayed on the guava shoots using a fine atomizer. After spraying, the guava shoots were air dried and tightly placed using wet cotton in a glass vial (6 cm height) which was filled with distilled water (15 ml). Then, thirty adults of *H. antonii* (five days old) were selected from the stock culture and released separately into each cage containing guava shoots. Shoots sprayed with water was maintained as control. Each treatment was replicated three times, and adult mortality was observed every 24 h up to 72 HAT and LC<sub>50</sub> was worked out. The observation on mortality was recorded at 12 h interval up to 72 HAT at highest concentration for each insecticide viz., Clothianidin 50% WDG (9.60 ppm), Thiacloprid 21.7% SC (16.00 ppm), Bifenthrin 10% EC (8.00 ppm), and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC (6.40 ppm) to work out the LT<sub>50</sub>. The group, mode of action and waiting period of selected insecticides were listed below.

S. No.	Insecticide*	Group	Mode of action	Waiting period (Day(s))
1.	Clothianidin 50% WDG	Neonicotinoid (Nitro-substituted)	Nicotinic acetylcholine receptor (nAChR) competitive modulator	5
2.	Thiacloprid 21.7% SC	Neonicotinoid (Cyano-substituted)	nAChR competitive modulator	7
3.	Bifenthrin 10% EC	Pyrethroids	Sodium channel modulator	11
4.	Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC	Neonicotinoid (Nitro-substituted) + Pyrethroids	nAChR competitive modulator + Sodium channel modulator	1

\*Approved by CIB&RC for Tea mosquito bug on tea

### Data analysis

Using Abbott's formula (Abbott, 1925), the percentage mortality in the test insects was corrected in relation to the control mortality. A completely randomized design (CRD) was used for the experiment. Lethal concentration and time for 50 per cent mortality *i.e.*,  $LC_{50}$  and  $LT_{50}$  were calculated using SPSS version 16.0, based on Finney's (1971) probit analysis method and expressed in ppm.

### Results and Discussion

The mortality of adult TMB in response to various concentrations of selected insecticides indicated that Clothianidin 50% WDG showed a range of 51.72 to 100 per cent, followed by Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC (41.38 to 96.55%), Thiacloprid 21.7% SC (44.83 to 93.10%), and Bifenthrin 10% EC (31.03 to 82.76%) at 72 HAT (Fig. 1). Clothianidin 50% WDG consistently exhibited the highest mortality across all observations, reaching 96.67 per cent within 48 HAT. Among the selected insecticides, Clothianidin 50% WDG recorded the lowest  $LC_{50}$  value of 0.328 ppm (fiducial limits: 0.144-0.515 ppm) at 72 HAT (Table 1), followed by Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, Thiacloprid 21.7% SC, and Bifenthrin 10% EC with  $LC_{50}$  values of 0.335 ppm (0.163-0.519 ppm), 0.765 ppm (0.286-1.291 ppm), and 0.857 ppm (0.452-1.372 ppm), respectively. Clothianidin 50% WDG displayed the lowest  $LT_{50}$  value of 10.49 h (fiducial limits: 5.444-14.551 h) (Table 2). Followed by, Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, Thiacloprid 21.7% SC, and Bifenthrin 10% EC with  $LT_{50}$  values of 13.31 h (7.546-17.957 h), 14.40 h (7.977-19.534 h), and 19.16 h (10.858-25.687 h), respectively.

Aravinthraju *et al.* (2021) reported that the Clothianidin 50% WDG at 120 g/ha was most effective, with 83.75 per cent reduction over control on guava under field condition. Buprofenzin 25% SC at 1000 ml/ha (74.90%) and Thiacloprid 21.7% SC at 500 ml/ha (70.78%) were on par with each other and equally effective next to Clothianidin 50% WDG. While, Thiamethoxam 25% WG at 100 g/ha, Profenophos 50EC at 250 ml/ha, Spinosad 45% SC at 750 ml/ha, and Emamectin benzoate 5% SG at 500 g/ha were less effective. According to Samanta *et al.* (2017), thiamethoxam 12.60% and lambda-cyhalothrin 9.5% ZC reduced the TMB population with lowest shoot infestation in two locations *viz.*, Marapur Tea Estate, Terai (Location I) with a shoot infestation of 2.64 and 1.78 per cent, and Mogolkata Tea Estate, Dooars (Location II) with 2.38 and 1.79 per cent during 2012 and 2013, respectively.

The outcomes suggest that Clothianidin 50% WDG showed the highest mortality and the lowest  $LC_{50}$ , followed by Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC, Thiacloprid 21.7% SC, and Bifenthrin 10% EC. Furthermore, Clothianidin 50% WDG (5 days) and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC (one day) were reported to have shorter waiting periods compared to Thiacloprid 21.7% SC (7 days) and Bifenthrin 10% EC (11 days). Shorter waiting periods (Clothianidin and Thiamethoxam + Lambda-Cyhalothrin) may provide practical advantages in terms of flexibility in harvest schedules and overall crop management.

### Conclusion

Clothianidin 50% WDG and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC reported with highest mortality, lowest  $LC_{50}$ ,  $LT_{50}$  values and waiting period against TMB. Since, TMB occurs from flushing to fruiting stage on guava, it requires minimum two sprays to manage the population. Use of different insecticides with varying modes of action can help manage resistance development in TMB populations and

hence, the Clothianidin 50% WDG and Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC can be recommended on need basis in rotation. Also, further research under field conditions can strengthen this finding.

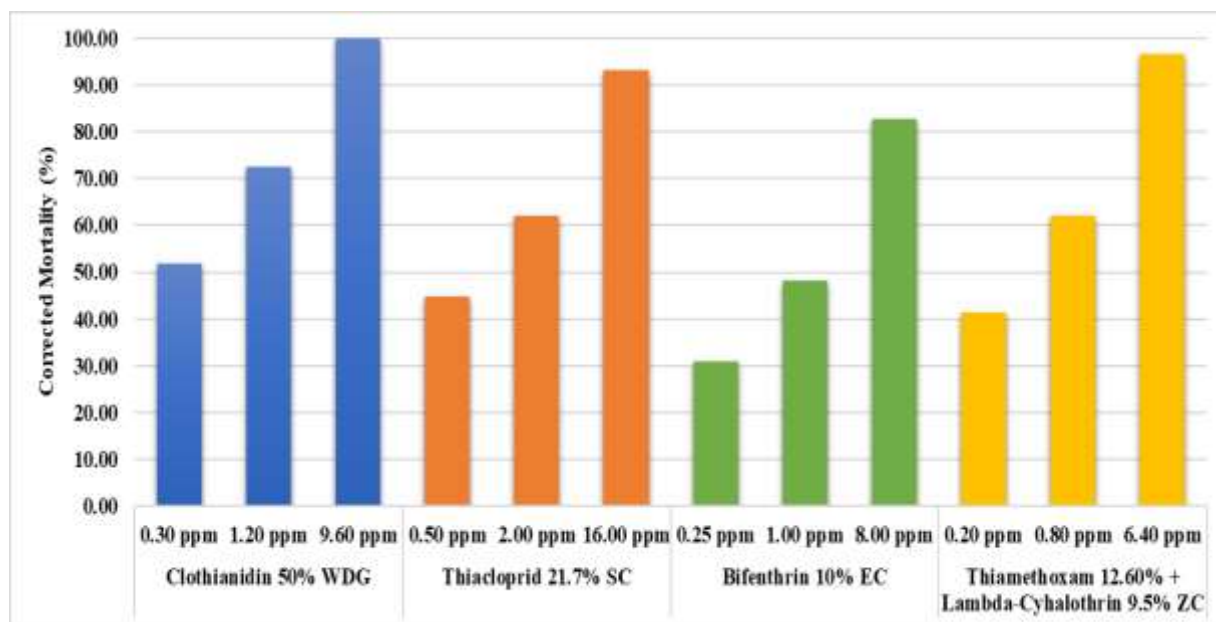


Fig. 1. Effect of Neonicotinoids and Pyrethroids against TMB, *H. antonii* under *in vitro* condition

Table 1. Concentration mortality response of *H. antonii* under *in vitro* condition

S. No.	Insecticide name	LC <sub>50</sub> (ppm)	Fiducial limits (95% confidence)		$\chi^2$	Regression equation
			Lower	Upper		
1.	Clothianidin 50% WDG	0.328	0.144	0.515	2.386	Y = 0.665+1.372X
2.	Thiachloprid 21.7% SC	0.765	0.286	1.291	0.741	Y = 0.114+0.982X
3.	Bifenthrin 10% EC	0.857	0.452	1.372	0.233	Y = 0.063+0.940X
4.	Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC	0.335	0.163	0.519	1.002	Y = 0.562+1.184X

No. of adults used in each treatment = 90

Y = mortality; X = dosage, LC<sub>50</sub> = median lethal concentration

Table 2. Time mortality response of *H. antonii* under *in vitro* condition

S. No.	Insecticide name	Concentration (ppm)	LT <sub>50</sub> (h)	Fiducial limits (95% confidence)		$\chi^2$	Regression equation
				Lower	Upper		
1.	Clothianidin 50% WDG	9.60	10.493	5.444	14.551	1.985	Y = -2.507+2.455X
2.	Thiachloprid 21.7% SC	16.00	14.401	7.977	19.534	0.372	Y = -2.336+2.016X
3.	Bifenthrin 10% EC	8.00	19.160	10.858	25.687	0.756	Y = -2.181+1.701X
4.	Thiamethoxam 12.60% + Lambda-Cyhalothrin 9.5% ZC	6.40	13.309	7.546	17.957	0.679	Y = -2.481+2.207X

No. of adults used in each treatment = 90

Y = mortality; X = time, LT<sub>50</sub> = median lethal time

#### Conflict of interest statement:

The authors declare that they have no conflict of interests.

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