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Insecticide resistance trend of Pymetrozine against Brown Plant Hopper populations of rice in different agro-climatic zones of Tamil Nadu

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Abstract
The widespread use of synthetic insecticides to control *Nilaparvata lugens* (Stål) (Brown Plant Hopper, BPH), a major threat to rice production in Asia, has unfortunately resulted in the evolution of insecticide resistance in this pest over the past decades. This study investigated insecticide use patterns and assessed the susceptibility of *N. lugens* populations to Pymetrozine in major rice-growing regions of Tamil Nadu, India. Results revealed significant deviations from recommended insecticide doses, with farmers frequently applying higher than recommended rates of Organophosphorus compounds, Neonicotinoids, and other insecticide groups. This excessive use can increase the risk of environmental contamination, enhance the selection pressure for insecticide resistance, and potentially reduce efficacy. Bioassays revealed significant variations in Pymetrozine susceptibility among BPH populations regions of Tamil Nadu. The population from Villupuram exhibited the highest susceptibility, while the Coimbatore population displayed the highest level of resistance with a Resistance Ratio value of 20.17, indicating a significant increase in resistance compared to the laboratory susceptible strain. These findings highlight the urgent need for improved insecticide use practices, including the adoption of Integrated Pest Management (IPM) strategies and regular monitoring of insecticide resistance in BPH populations to ensure sustainable rice production in Tamil Nadu.

Keywords: Pymetrozine, resistance, susceptibility, efficacy, monitoring, *Nilaparvata lugens*

Introduction
Rice (*Oryza sativa* L.), a vital staple food for nearly half the world's population, primarily in Asia and the Pacific, plays a crucial role in global food security. Its high calorific content and economic significance make it a cornerstone of many economies. To keep pace with the rising global demand, driven by population growth, annual rice production must increase by 1.1% until 2031, as projected by the World Economic Forum. A significant challenge to rice production and productivity in India is due to the infestation of various insect pests throughout the crop's life cycle. Over 100 insect species attack rice, and 20 of these can cause substantial economic damage (Pathak and Khan, 1994) [10]. The rice crop is subjected to sustain damage by considerable number of pests among them. The brown plant hopper (BPH), *Nilaparvata lugens* Stal (Delphacidae: Homoptera) is one of the economically migratory pests of rice paddy fields in India and they damage plants directly by sucking the plant sap and by ovipositing in plant tissue causing plant wilting or hopper burn (Turner *et al*., 1999) [14]. Damage to the rice crop is caused directly by feeding on the phloem (Sogava, 1992) and indirectly by transmitting plant viral disease like grassy stunt viruses (Powell *et al*., 1995) [11]. The control of *N. lugens* has primarily relied on various types of insecticides (Endo and Tsurumachi, 2001) [3]. This BPH can often develop resistance to various insecticides used in our country. Moreover, the resistance has already developed to various types of insecticides in almost all Southeast Asia countries, including south China, which have been considered as the region of *N. lugens* origin (Nagata, 2002; Matsumura *et al*., 2008; Wen *et al*., 2009; Latif *et al*., 2010) [6, 8, 9, 15]. Hence, it is necessary to monitor the resistance levels to various insecticides in the progenitor populations to manage *N. lugens* populations in a more efficient manner. In this aspect, the resistance levels Pymetrozine (the most commonly used insecticide in local strains *N. Lugens*) was monitored as the first step towards the effective control of *N. lugens* in different Agroclimatic Zones of Tamil Nadu.

Materials and Methods
A survey was conducted in different Agro-climatic zones of Tamil Nadu to identify the primary pesticides used by farmers to manage the Brown Plant Hopper (BPH) and other rice pests. The survey aimed to assess the prevalence of different pesticide types and application methods. Among the numerous insecticides used to control BPH in these areas, the most commonly and frequently used insecticide were selected for this study: thiamethoxam, pymetrozine, buprofezin, quinalphos, clothianidin, acephate, fipronil, ethofenprox, triflumezopyrim and imidacloprid. From the study most commonly and widely used insecticide was evaluated for insecticide resistance.
The BPH collected from different villages of selected districts in selected Agroclimatic zone of Tamil Nadu sites during 2022 were used, with more than 200 healthy female adults and 600 nymphs collected from the collar region using an aspirator or sweep nets. These were initially kept in plastic container with rice plants and the shoots alone

clipped off to put on the lid. One to two days old female adults of these populations were used for the bioassay. The field collected populations were used as a starting culture reared on 10 days old TN-1 seedlings transplanted in plastic pots maintained at Department of Entomology, Faculty of Agriculture, Annamalai University, Chidambaram (11.39º N, 79.69º E); it was observed that 25-35 days old plants were ideal for feeding and oviposition. The test insects were mass cultured in the screen house (25± 1°C, 70-80% RH). Adults confined to the 35 to 45 days old potted plants and covered with transparent mesh on all the sides (Heinrichs, 1981). Adults were allowed to oviposit on 55 to 60 days old plants and freshly hatched nymphs were offered young seedlings. Continuous and pure susceptible culture of the *N. lugens* was thus maintained. The bioassays were carried out using seedling dip method- IRAC method No. 5 (IRAC, 2012) and 12 days old rice seedling (TN-1) were used. Six concentrations of insecticide replicated thrice were laid out in a completely randomized block design. Preliminary range-finding tests were done to fix the test dose causing 20-80% mortality approximately for constructing log-concentration-probit mortality (LCPM) lines. The test mortality was corrected against untreated mortality as per Abbott's formula (Abbott, 1925) [1] and subjected to Finney's probit analysis to estimate the median lethal concentrations for the dose-mortality response (Finney, 1971) [4]. The resistance ratio (RR) was calculated by dividing the LC50 value of the field population by that of LC50 of the susceptible one (Baehaki *et al*., 2017; Liao *et al*., 2017) [2, 7].

$$\text{RR at } LC_{50} = \frac{LC_{50} \text{ of Resistant Strain (RS)}}{LC_{50} \text{ of Susceptible Strain (SS)}}$$

Results and Discussion

The data on insecticides use pattern observed among rice farmers in major growing areas of Tamil Nadu revealed significant deviations from recommended doses for most of the insecticides surveyed. Organophosphorus Compounds: Quinalphos and Acephate were widely used, with 40% and 53.33% of farmers, respectively, reporting their application. However, farmers were observed to apply significantly higher doses than recommended, with average application rates exceeding recommended rates by 25% and 40% for Quinalphos and Acephate, respectively. Neonicotinoids: Clothianidin, Imidacloprid, and Thiamethoxam were also widely used, with adoption rates ranging from 66.67% to 73.33%. While the application rates for these insecticides were generally closer to the recommended doses, deviations were still observed, with some farmers applying higher than recommended rates. Other Insecticide Groups like Fipronil (Phenyl Pyrazole) and Pymetrozine (Pyridine Azomethine Derivative) were also commonly used, with adoption rates exceeding 50%. Similar to Organophosphorus compounds, farmers tend to apply higher doses of these insecticides than recommended. Buprefezin (Pyridine Azomethine Derivative) and Ethofenprox (Synthetic Pyrethroid) were used by 56.67% and 16.67% of farmers, respectively, with application rates generally exceeding recommended doses. Triflumezopyrim (Mesoionic insecticide) was used by 26.67% of farmers, with application rates slightly higher than the recommended dose (Table 1).

Higher application rates can lead to increased environmental contamination of water bodies and soil, posing risks to non-target organisms and the excessive use of insecticides can increase the selection pressure for the development of insecticide resistance in target pests. Higher than recommended doses may not always translate to increased efficacy and can even lead to reduced efficacy due to potential phytotoxicity or negative impacts on natural enemies of the target pest.

The acute toxicity bioassays were conducted to assess the susceptibility of *N. lugens* (BPH) populations from different locations in Tamil Nadu to Pymetrozine 50 WG. The LC50 values, along with their 95% fiducial limits, were determined using probit analysis, and the regression equation for each population is provided.

Significant variations in susceptibility to Pymetrozine were observed among the BPH populations. The population from Villupuram exhibited the highest susceptibility with the lowest LC50 value of (4.0041 µg/ml) followed by salem population (4.0682 µg) while the population from Coimbatore showed the highest level of resistance with the highest LC50 value (8.6735 µg/ml) followed Thanjavur population (7.7320 µg/ml). Resistance ratios (RR) were calculated by comparing the LC50 susceptible brown plant hopper strain maintained in the laboratory to the population to the LC50 of different BPH populations under study. The RR values ranged from 9.53 (Villupuram) to 20.17 (Coimbatore), indicating varying levels of resistance across the sampled locations (Table 2). Similarly, Song *et al*., 2023 [13] also reported pymetrozine insecticide resistance in BPH populations of difference provinces China the RR values ranges from 1 to 10.8.

Conclusion

The findings suggest the development of Pymetrozine resistance in BPH populations across Tamil Nadu. The observed variations in susceptibility likely reflect the differential selection pressures exerted by varying levels of Pymetrozine use in different regions. The high RR values observed in some populations highlight the need for effective resistance management strategies to prevent further escalation of resistance. The findings also highlight the need for improved insecticide use practices among rice farmers in Tamil Nadu. By promoting IPM strategies and providing farmers with accurate information and training, it is possible to minimize the risks associated with excessive insecticide use and ensure the sustainable management of rice pests.

Further Study

Further research is necessary to investigate the specific mechanisms of resistance in these BPH populations. This may involve identifying mutations in target sites, characterizing metabolic resistance mechanisms, and exploring alternative control options.

Disclaimer (Artificial Intelligence)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text to image generators have been used during the writing or editing of manuscript.

Table 1: Insecticides Use Pattern in major rice growing regions of Tamilnadu

Group/ Insecticide	Percent Farmers*	Farmers Dose		Recommended Dose	
		(ml/g/litre)	(ml/g/ acre)	(ml/g/litre)	(ml/g/ acre)
Organo Phosphorous Compounds					
Quinalphos 25 EC	40.00± 5.77	3.50±0.52	750.00±105.00	3.00	600.00
Acephate 75 SP	53.33±7.73	4.00±0.54	800.00±100.48	2.00	400.00
Neonicotinoids					
Clothianidin 50 WG	66.67±7.73	0.3±0.42	60.00±8.40	0.12	24.00
Imidacloprid	73.33±10.0	0.50±0.10	100.00±19.00	0.25	50.00
Thiamethaxam 50 WG	66.67±5.46	0.40±0.08	80.00±20.20	0.20	40.00
Phenyl Pyrazole					
Fipronil	56.67±7.73	4.00±0.62	800.00±124.20	3.00	600.00
Pyridine Azomethine Derivative					
Pymetrozine 50 WG	83.33±10.0	0.5±0.08	100.00±16.32	0.60	120.00
Buprefezin					
Buprefezine 25 SC	56.67±5.77	2.00±0.42	400.00±20.84	1.60	320.00
Synthetic Pyrethroid					
Ethofenoprox 10 EC	16.67±2.89	2.00±0.18	400.00±32.36	1.50	300.00
Mesoionic (Zwitterionic) insecticide					
Triflumezopyrim 20 WG	26.67±2.89	0.30±0.06	60.00±12.50	0.25	50.00

*mean of 30 farmers; ± SD

Table 2: Acute toxicity of Pymetrozine 50 WG to *N. lugens* populations from different locations of Tamil Nadu

Insecticides	Regression Equation	□2	LC 50 (ppm)	Fiducial Limits		LC 95	Fiducial Limits		RR
				LL	UL		LL	UL	
Cuddalore	Y= 3.0231x+2.9424	0.0418	5.3091	4.2081	6.6982	30.0751	13.3959	67.5213	12.64
Villupuram	Y= 3.0359x+3.2056	1.9622	4.0041	3.2015	5.0081	18.0780	10.6007	30.8294	09.53
Mayiladuthurai	Y = 3.0263x+2.7497	3.4239	5.9441	4.9517	7.1353	23.2286	13.4176	40.2133	14.15
Thanjavur	Y = 3.0453x+2.4928	2.9037	7.7320	6.1726	9.6854	33.6780	15.8634	71.4981	18.40
Pudukkottai	Y= 2.8922x+3.0975	2.1256	5.0027	3.9242	6.3777	30.3500	13.1552	70.6195	11.91
Salem	Y= 3.3245x+2.9991	0.9441	4.0682	3.3465	4.9456	14.6427	9.7299	22.0360	9.67
Coimbatore	Y=2.9055x+2.3921	2.4608	8.6735	6.8694	10.9516	34.4688	16.3939	72.4719	20.17

LC- Lethal Concentration, UL- Upper Limit, LL- Lower Limit, RR- Resistance Ratio

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