RELATIVE SUSCEPTIBILITY OF LIFE STAGES OF COTTON WHITEFLY
BEMISIA TABACI (GENN.) TO PYRIPROXYFEN

KELAGERI S S, MAHAPATRO G K*, SUBRAMANIAN S, CHITRA SRIVASTAVA AND RAJNA S

Division of Entomology, ICAR-Indian Agricultural Research Institute (IARI), New Delhi 110012, India
*IARI Regional Station, Pune 411067, Maharashtra, India
*Email: gagan_gk@rediffmail.com (corresponding author)

ABSTRACT

The relative susceptibility of lifestages of seven Bemisia tabaci (Genn.) populations from major cotton growing regions in India to pyriproxyfen has been studied. The results revealed that adults were unaffected at significantly high concentration i.e., 200 mg/l (field recommended dose, <10% mortality), hence adult bioassay was not done. Indore and Amravati populations were the most and least susceptible ones for both egg and nymphal stages; and all the populations were found susceptible to pyriproxyfen with RR ratio of <5, except for Amravati one, revealing low level of resistance with RR ratio (5.04- egg; 5.09- nymph) computed deploying the LC\textsubscript{90} and LC\textsubscript{50} values.

Key words: Bemisia tabaci, pyriproxyfen, bioassay, LC\textsubscript{90}, LC\textsubscript{50}, adults, egg, nymph, juvenile hormone analogue, relative resistance ratio

The cotton whitefly Bemisia tabaci (Gennadius) is a pest of global significance affecting wide range of crops including field, vegetable, fruit and ornamental crops (Kanakala and Ghanim 2019; Horowitz et al., 2020). Indirectly B. tabaci affects crops through vectoring more than 114 virus species (Simon, 2003). Use of insecticides is the major control measure against B. tabaci, although the rapid development of insecticide resistance by B. tabaci has resulted in frequent pest outbreaks. It has evolved resistance to most of the commonly used insecticides (Basit 2019, Horowitz et al., 2020; Mota-Sanchez and Wise, 2019). Involvement of biorational insecticides in the spray schedule reduces selection pressure and encourages natural enemies. Juvenile hormone analogues (JHAs) are the synthetic analogues of juvenile hormone, and these are considered to be effective and environment friendly (Mohandass et al., 2006). Pyriproxyfen is a pyridine based juvenile hormone analogue i.e., 4-phenoxyphenyl (RS)-2-(2-pyridyloxy) propyl ether, it targets JH binding site receptors in insects by mimicking the action of juvenile hormone and thus keeping it in the immature stage (Sullivan and Goh, 2008). Pyriproxyfen has been proven as an effective molecule for managing B. tabaci with strong ovicidal action, inhibiting adult emergence and translaminar activity against eggs; and the egg hatchability of the treated female gets suppressed (Ishaaya and Horowitz, 1992). In this study the relative susceptibility of seven B. tabaci populations from cotton growing regions of India to pyriproxyfen has been evaluated.

MATERIALS AND METHODS

Bemisia tabaci populations were collected from seven cotton growing locations and reared on cotton (Gossypium hirsutum L.) in the Insect Proof Climate Control Chamber, Division of Entomology, IARI (27±2°C, 60-70% RH, and photoperiod 14:10- L:D). The whiteflies collected from Leucaena leucocephala from Pusa campus, reared under laboratory condition, served as the susceptible check. The details of populations are- Amravati (21.02°N 77.48°E), Guntur (16.22°N,80.30°E), Hisar (29.09°N, 75.87°E), Indore (22.8°N,75.73°E), Ludhiana (30.54°N, 75.48°E), New Delhi (28.64°N,77.17°E), Sriganganagar (29.54°N,73.54°E), and susceptible (laboratory-28.38°N 77.09°E) one. Commercial formulation of pyriproxyfen 10 EC (Lano®, Sumitomo Chemical India) procured from the market was diluted with deionized water to make 1% stock solution for use in bioassay. Seven concentrations with three replications were set and studies carried out following the modified Insecticide Resistance Action Committee (IRAC) protocols (https://irac-online.org/). All the important lifestages viz., egg, nymph (N2) and adults were used; lethal effects in egg and nymphal stage (2\textsuperscript{nd} instar) were tested following the IRAC method No. 016 (formally method No 12c) (https://irac-online.org/), whereas adults were tested with leaf dip bioassay- modified IRAC method by Naveen et al. (2017). The estimates of lethal concentrations and 95% confidence intervals were determined by log–dose probit analysis using
Pyriproxyfen showed limited efficacy against adults of *B. tabaci*, but maintaining the insect in its immature stage, with suppression of embryogenesis and adult formation (Ishaaya and Horowitz 1992). Pyriproxyfen is known to cause the external deformities in emerged adults such as twisted wings and legs in case of *Plodia interpunctella* (Ghasemi et al., 2010), apart from external deformities pyriproxyfen reduces the size of ovaries due to the reduction in the synthesis and supply of lipid and protein (Ghasemi et al., 2010). There are many reports of pyriproxyfen resistance in *B. tabaci* - Egg bioassays by Devine et al. (1999) reported very high level of resistance (6500-fold) among Israeli populations; low to moderate level of resistance (11-fold) in Chinese populations (Luo et al., 2010); moderate resistance from Al Hassa Oasis (Saudi Arabia) (Hajjar et al., 2019); and high resistance (89.71-folds) from cotton field populations of Arizona (Ma et al., 2010) and Australian (96.9 fold) (Hopkinson et al., 2019). Similarly with nymphal bio assay studies, Basit et al. (2013) reported low level of resistance from Pakistan; moderate level of resistance (30.08-fold) from West Bengal, India was observed (Roy et al., 2019); and very high level of resistance (1100-fold) in Israeli populations (Devine et al., 1999). Pyriproxyfen resistance in *B. tabaci* might be a case of metabolic resistance involving cytochrome P450 monooxygenases (P450s) and glutathione S-transferases (GSTs) (Ma et al., 2010; Ghanim et al., 2007; Nauen et al., 2015).

The present results are in contrast with the resistance development data available from other parts of the world, as it has been observed that the Indian populations are highly susceptible to the pyriproxyfen at both egg and nymphal stages. This might be due to the less selection pressure exerted as pyriproxyfen is not among the mainstream insecticides. Another factor is the dominance of B biotype of *B. tabaci* in the Indian subcontinent, as confirmed by the earlier studies (Ellango et al., 2015; Mandal et al., 2016). It was observed that cases of strong resistance to pyriproxyfen have been associated with the Q rather than the B biotype (Dennehy et al., 2005; Horowitz and Ishaaya, 2014). This fact derives support from the data that in Israel considerable reduction in pyriproxyfen resistance was observed since 2009. Since then studies had shown a significant shift in the biotype ratios i.e. the B biotype has become predominate over the Q (Crowder et al., 2011; Horowitz and Ishaaya, 2014). Recent studies involving the *B. tabaci* populations from major cotton growing regions of India revealed the LC$_{50}$ values of 52 to 956 and 26 to 194 mg/ l for imidacloprid and thiamethoxam; while pyrethroids viz., cypermethrin
Table 1. Log dose probit mortality data for pyriproxyfen against egg stage of different *Bemisia tabaci* populations

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Population</th>
<th>df</th>
<th>Slope± SE</th>
<th>( \chi^2 )</th>
<th>LC(_{50}) value mg/l (CI 95%)</th>
<th>Fiducial limit for LC(_{50}) (mg/l)</th>
<th>RR for LC(_{50})</th>
<th>LC(_{90}) value mg/l (CI 95%)</th>
<th>Fiducial limit for LC(_{90}) (mg/l)</th>
<th>RR for LC(_{90})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lab Susceptible</td>
<td>3</td>
<td>1.929± 0.104</td>
<td>2.764</td>
<td>0.018</td>
<td>0.010 to 0.031 (a)</td>
<td>1.00</td>
<td>0.083</td>
<td>0.042 to 1.538 (a)</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Amravati</td>
<td>3</td>
<td>2.171± 0.095</td>
<td>1.261</td>
<td>0.064</td>
<td>0.026 to 0.115 (a)</td>
<td>3.56</td>
<td>0.418</td>
<td>0.188 to 16.826 (a)</td>
<td>5.04</td>
</tr>
<tr>
<td>2</td>
<td>Guntur</td>
<td>3</td>
<td>1.552± 0.089</td>
<td>3.150</td>
<td>0.038</td>
<td>0.031 to 0.045 (a)</td>
<td>2.11</td>
<td>0.253</td>
<td>0.178 to 0.426 (a)</td>
<td>3.05</td>
</tr>
<tr>
<td>3</td>
<td>Hisar</td>
<td>3</td>
<td>1.850± 0.095</td>
<td>1.146</td>
<td>0.052</td>
<td>0.043 to 0.063 (b)</td>
<td>2.89</td>
<td>0.258</td>
<td>0.181 to 0.458 (a)</td>
<td>3.11</td>
</tr>
<tr>
<td>4</td>
<td>Indore</td>
<td>4</td>
<td>2.566± 0.085</td>
<td>2.175</td>
<td>0.020</td>
<td>0.010 to 0.040 (a)</td>
<td>1.11</td>
<td>0.135</td>
<td>0.057 to 6.133 (a)</td>
<td>1.63</td>
</tr>
<tr>
<td>5</td>
<td>Ludhiana</td>
<td>3</td>
<td>1.623± 0.082</td>
<td>1.781</td>
<td>0.049</td>
<td>0.027 to 0.083 (a)</td>
<td>1.72</td>
<td>0.303</td>
<td>0.145 to 3.328 (a)</td>
<td>3.65</td>
</tr>
<tr>
<td>6</td>
<td>New Delhi</td>
<td>3</td>
<td>1.556± 0.079</td>
<td>3.134</td>
<td>0.030</td>
<td>0.023 to 0.039 (a)</td>
<td>1.67</td>
<td>0.201</td>
<td>0.123 to 0.484 (a)</td>
<td>2.42</td>
</tr>
<tr>
<td>7</td>
<td>Sriganganagar</td>
<td>3</td>
<td>1.555± 0.087</td>
<td>2.112</td>
<td>0.056</td>
<td>0.035 to 0.089 (b)</td>
<td>3.11</td>
<td>0.371</td>
<td>0.179 to 3.038 (a)</td>
<td>4.47</td>
</tr>
</tbody>
</table>

Log dose probit mortality data for pyriproxyfen against nymphal stage (2\(^{nd}\) instar) of different *Bemisia tabaci* populations

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Population</th>
<th>df</th>
<th>Slope± SE</th>
<th>( \chi^2 )</th>
<th>LC(_{50}) value mg/l (CI 95%)</th>
<th>Fiducial limit for LC(_{50}) (mg/l)</th>
<th>RR for LC(_{50})</th>
<th>LC(_{90}) value mg/l (CI 95%)</th>
<th>Fiducial limit for LC(_{90}) (mg/l)</th>
<th>RR for LC(_{90})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lab Susceptible</td>
<td>3</td>
<td>1.565± 0.269</td>
<td>2.824</td>
<td>0.054</td>
<td>0.041 to 0.073 (a)</td>
<td>1.00</td>
<td>0.176</td>
<td>0.105 to 0.316 (a)</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Amravati</td>
<td>3</td>
<td>2.543± 0.416</td>
<td>0.874</td>
<td>0.275</td>
<td>0.193 to 0.396 (c)</td>
<td>5.09</td>
<td>0.878</td>
<td>0.540 to 4.244 (b)</td>
<td>4.99</td>
</tr>
<tr>
<td>2</td>
<td>Guntur</td>
<td>4</td>
<td>3.562± 0.541</td>
<td>2.863</td>
<td>0.155</td>
<td>0.136 to 0.176 (b)</td>
<td>2.87</td>
<td>0.355</td>
<td>0.285 to 5.171 (a)</td>
<td>2.02</td>
</tr>
<tr>
<td>3</td>
<td>Hisar</td>
<td>4</td>
<td>3.603± 0.589</td>
<td>0.985</td>
<td>0.173</td>
<td>0.151 to 0.195 (b)</td>
<td>3.20</td>
<td>0.393</td>
<td>0.318 to 0.575 (b)</td>
<td>2.23</td>
</tr>
<tr>
<td>4</td>
<td>Indore</td>
<td>3</td>
<td>2.362± 0.397</td>
<td>0.485</td>
<td>0.079</td>
<td>0.065 to 0.095 (a)</td>
<td>1.46</td>
<td>0.275</td>
<td>0.206 to 1.024 (a)</td>
<td>1.56</td>
</tr>
<tr>
<td>5</td>
<td>Ludhiana</td>
<td>4</td>
<td>3.236± 0.569</td>
<td>2.142</td>
<td>0.191</td>
<td>0.169 to 0.216 (c)</td>
<td>3.54</td>
<td>0.424</td>
<td>0.340 to 0.634 (b)</td>
<td>2.41</td>
</tr>
<tr>
<td>6</td>
<td>New Delhi</td>
<td>3</td>
<td>2.949± 0.488</td>
<td>0.062</td>
<td>0.137</td>
<td>0.117 to 0.159 (b)</td>
<td>2.54</td>
<td>0.373</td>
<td>0.285 to 0.614 (a)</td>
<td>2.12</td>
</tr>
<tr>
<td>7</td>
<td>Sriganganagar</td>
<td>3</td>
<td>3.880± 0.599</td>
<td>1.150</td>
<td>0.187</td>
<td>0.166 to 0.210 (c)</td>
<td>3.46</td>
<td>0.400</td>
<td>0.327 to 0.568 (b)</td>
<td>2.27</td>
</tr>
</tbody>
</table>

Chi-square values non-significant- \( p=0.05 \) (table value 3df=7.81, 4df=9.49); Letters in parentheses indicate significant difference in lethal concentrations.
and deltamethrin showed LC\textsubscript{50} values of 10 to 1362 and 10 to 760 mg/l respectively. Similarly OP insecticides triazophos, monocrotophos and chlorpyrifos showed LC\textsubscript{50} values ranging from 53 to 1429, 88 to 3934, 12 to 220 mg/l, respectively (Naveen et al., 2017). Novel insecticides fipronil and flonicamid showed LC\textsubscript{50} values of 6.56 to 20.80 and 23.35 to 749.91 mg/l (Romila et al., 2019) and for cytraniliprole it was 1.80 to 4.57 mg/l (Rajna et al., 2021). Comparison of these LC\textsubscript{50} values with those of pyriproxyfen from the present study viz., 0.018 to 0.064 mg/l for egg and 0.054 to 0.275 mg/l for nymph reveal the supremacy of the pyriproxyfen, suggesting that it can be used as a stage specific insecticide in IRM programmes.

ACKNOWLEDGEMENTS

This study is a part of the doctoral research work of the first author entitled “Toxicity variations of insecticides against cotton whitefly *Bemisia tabaci* (Gennadius)”. The senior author thanks the Division of Entomology, ICAR-IARI for the facilities provided.

FINANCIAL SUPPORT

The senior author is thankful to the Ministry of Tribal Affairs, GOI, for providing financial assistance in the form of National Fellowship for Higher Education.

AUTHOR CONTRIBUTION STATEMENT

CS, MGK, and SS conceived and designed research, KSS and RS conducted experiments and analyzed data, KSS wrote manuscript and MGK corrected manuscript.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

REFERENCES


lateral flow test to detect metabolic resistance in *Bemisia tabaci* mediated by CYP6CM1, a cytochrome P450 with broad spectrum catalytic efficiency. Pesticide Biochemistry and Physiology 121: 3-11.


