Relative susceptibility of the Bikaner and Delhi populations of mustard aphid, *Lipaphis erysimi* (Kalt.) (Homoptera: Aphididae), and its predator, *Coccinella septempunctata* L. (Coleoptera: Coccinellidae), to different insecticides

Susceptibilité relative des populations de Bikaner et Delhi du puceron de la moutarde, *Lipaphis erysimi* (Kalt.) (Homoptera: Aphididae) et de leur prédateur, *Coccinella septempunctata* L. (Coleoptera: Coccinellidae), à différents insecticides

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Résumé de l'article

Une étude a été menée pour connaître l'efficacité de cinq insecticides sur des populations du puceron de la moutarde, *Lipaphis erysimi* (Kalt.), de Delhi et de Bikaner en utilisant le procédé de résidu de feuille, ainsi que sur *Coccinella septempunctata* L. en condition semi-naturelle. L'acétamipride et le thiaméthoxame se sont révélés plus toxiques que les autres insecticides. Après 24 h, les valeurs de LC50 pour la population de Bikaner en réaction aux différents insecticides étaient de 7,0, 6,0, 4,0, 3,0 et 2,0 ppm pour le carbosulfan, la bifenthrine, l'imidaclopride, l'acétamipride et le thiaméthoxame, respectivement. De même, l'ordre décroissant de toxicité pour la population de Delhi était l'acétamipride (7,0 ppm), le thiaméthoxame (9,0 ppm), l'imidaclopride (15,0 ppm), le carbosulfan (32,0 ppm) et la bifenthrine (36,0 ppm). Les valeurs de toxicité relatives ont démontré que dans les deux populations, le thiaméthoxame et l'acétamipride avaient la plus haute toxicité. Le carbosulfan et la bifenthrine étaient fortement toxiques aux larves de coccinelles et ont provoqué la mortalité de 100 % de la population en condition semi-naturelle, tandis que les néonicotinoïdes, l'acétamipride et le thiaméthoxame ont engendré moins de mortalité. Cela démontre la tolérance des coccinelles aux néonicotinoïdes en condition semi-naturelle.
Relative susceptibility of the Bikaner and Delhi populations of mustard aphid, *Lipaphis erysimi* (Kalt.) (Homoptera: Aphididae), and its predator, *Coccinella septempunctata* L. (Coleoptera: Coccinellidae), to different insecticides

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A study was undertaken to assess the effectiveness of five insecticides against the Delhi and Bikaner populations of mustard aphid, *Lipaphis erysimi* (Kalt.), using the leaf dip method, and against *Coccinella septempunctata* L. in semi-field conditions. Acetamiprid and thiamethoxam were found to be more toxic than other insecticides. After 24 h, the LC$_{50}$ values for the Bikaner population against different insecticides were 7.0, 6.0, 4.0, 3.0 and 2.0 ppm for carbosulfan, bifenthrin, imidacloprid, acetamiprid and thiamethoxam, respectively. Similarly, the descending order of toxicity for the Delhi population was acetamiprid (7.0 ppm), thiamethoxam (9.0 ppm), imidacloprid (15.0 ppm), carbosulfan (32.0 ppm) and bifenthrin (36.0 ppm). The relative toxicity values suggest that in both populations, thiamethoxam and acetamiprid show the highest toxicity. Carbosulfan and bifenthrin were highly toxic to coccinellid grubs and resulted in 100% mortality in semi-field conditions, whereas the neonicotinoids acetamiprid and thiamethoxam showed less mortality. It showed the tolerance of coccinellidae against neonicotinoids under semi-field conditions.

Key words: *Coccinella septempunctata*, *Lipaphis erysimi*, populations, insecticides, safety, toxicity

Susceptibilité relative des populations de Bikaner et Delhi du puceron de la moutarde, *Lipaphis erysimi* (Kalt.) (Homoptera: Aphididae) et de leur prédateur, *Coccinella septempunctata* L. (Coleoptera: Coccinellidae), à différents insecticides

Une étude a été menée pour connaître l’efficacité de cinq insecticides sur des populations du puceron de la moutarde, *Lipaphis erysimi* (Kalt.), de Delhi et de Bikaner en utilisant le procédé de résidu de feuille, ainsi que sur *Coccinella septempunctata* L. en condition semi-naturelle. L’acétamipride et le thiaméthoxame se sont révélés plus toxiques que les autres insecticides. Après 24 h, les valeurs de LC$_{50}$ pour la population de Bikaner en réaction aux différents insecticides étaient de 7,0, 6,0, 4,0, 3,0 et 2,0 ppm pour le carbosulfan, la bifenthrine, l’imidaclopride, l’acétamipride et le thiaméthoxame, respectivement. De même, l’ordre décroissant de toxicité pour la population de Delhi était l’acétamipride (7,0 ppm), le thiaméthoxame (9,0 ppm), l’imidaclopride (15,0 ppm), le carbosulfan (32,0 ppm) et la bifenthrine (36,0 ppm). Les valeurs de toxicité relatives ont démontré que dans les deux populations, le thiaméthoxame et l’acétamipride avaient la plus haute toxicité. Le carbosulfan et la bifenthrine étaient fortement toxiques aux larves de coccinelles et ont provoqué la mortalité de 100 % de la population en condition semi-naturelle, tandis que les néonicotinoïdes, l’acétamipride et le thiaméthoxame ont engendré moins de mortalité. Cela démontre la tolérance des coccinelles aux néonicotinoïdes en condition semi-naturelle.

Mots clés: *Coccinella septempunctata*, insecticides, *Lipaphis erysimi*, populations, sécurité, toxicité

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INTRODUCTION
Mustard, Brassica juncea L., is an important oil seed crop, the second most important after groundnut in India (Bartaria et al. 2001). Every effort is being made to raise its yield by adopting modern agricultural practices, such as the use of high yielding varieties, judicious use of fertilizers and assured irrigation, in order to meet the growing demand for oils. However, these efforts are nullified if the crop is not protected from damage caused by insect pests. More than three dozens of insect pests infest the crop at various growth stages (Rai 1976). Of these, the mustard aphid, Lipaphis erysimi (Kalt.), is considered to be a limiting factor in the successful cultivation of rapeseed mustard, causing yield reductions of up to 91.3% (Singh and Sachan 1994; Sharma and Kashyap 1998; Gupta et al. 2003) and oil contents reductions of up to 15% (Verma and Singh 1987). Mustard aphid colonies feed on new shoots, inflorescence and the underside of leaves. Therefore, it is essential to keep this pest under control in order to reap a profitable harvest. To control this pest, different insecticides have been evaluated and recommended by researchers (e.g. Bakhetia et al. 1988; Kumar et al. 2007; Boopathi et al. 2010; Boopathi and Pathak 2011). Testing insecticides on insect pests and associated natural enemies has become common practice over the last two decades. However, the number of natural enemies introduced into tests and the number of preparations tested on a particular species are variable. The predacious coccinellid beetles, commonly known as ladybird beetles, are considered to be of great economic importance in the agro-ecosystem. They have been successfully employed in the bio-control of many injurious insects (Agarwal et al. 1988). In the field, mustard aphid populations are naturally controlled, to a large extent, by its predator Coccinella septempunctata L., which plays a vital role in reducing the population of mustard aphids in the field (Kalra 1988). To control mustard aphid successfully while preserving C. septempunctata, insecticides should be applied at appropriate doses and at the right time. Keeping these points in mind, the present study aimed to assess the effectiveness of five different insecticides belonging to different chemical groups. These were studied against Delhi and Bikaner populations of L. erysimi under laboratory condition and in a semi-field evaluation against grubs of C. septempunctata.

MATERIALS AND METHODS
Insecticides
Commercial formulations of the insecticides used for the present investigation were obtained from their respective manufacturers: imidacloprid (Confidor 17.8 SL %, Bayer CropScience Limited, Mumbai), acetamiprid (Baadshah 20% SP, Hindustan Pulverising Mills, Jammu), thiamethoxam (Actara WG 25%, Syngenta, Mumbai), bifenthrin (Talstar 10% EC) and carbosulfan (Marshall 20% EC) were obtained from FMC India Private Limited, Kanchipuram. Different concentrations of various insecticidal emulsions were prepared by using double distilled water for the dilution of emulsifiable concentrations using the serial dilution technique described in Shankarganesh et al. 2009.

Insetcs
Apterous specimens of L. erysimi were collected from mustard fields of the Indian Agricultural Research Institute, New Delhi, and from farmers’ fields near the Central Arid Zone Research Institute, Bikaner, Rajasthan. Grubs and adults of C. septempunctata were collected from experimental farms of the Indian Agricultural Research Institute, New Delhi, in 2010.

Laboratory bioassay
Apterous aphid individuals were kept under controlled conditions at 27 ± 1°C and 60 ± 5% RH on mustard leaves. Healthy female aphid individuals were removed from the mustard twig and kept separately under laboratory conditions for preconditioning 30 min before treatment. Aphids were exposed to insecticidal residues on mustard leaves. Leaf discs of approximately 3 cm diam. were cut from well-grown leaves. The leaf discs were thoroughly washed with water, dipped in the required emulsion concentration for 20 sec and then shade dried. The treated leaf discs were then transferred to clean jars (15 cm × 10 cm). Twenty aperous females were placed in each jar, and each treatment was replicated twice. Simultaneously, a control was created by treating the leaf discs with water. For the toxic effect assessment, mortality counts were made 24 h after treatment (AT). Moribund insects were also counted as dead. Five to seven concentrations of each insecticide were tested to obtain a concentration-probit mortality curve. Estimates of LC50 with 95% confidence intervals (CI) were determined by log-dose probit analysis (Finney 1971, using PoloPlus 2.0 (LeOra software 2005)). Relative toxicity was calculated using the standard procedure described in Shankarganesh et al. 2007.

Semi-field bioassay
To study the effect of residual toxicity of insecticides on the grubs of C. septempunctata, mustard variety ‘Pusa Bold’ was sown following standard agronomic practices. Thereafter, we allowed natural infestation of L. erysimi to take place. Field-recommended doses of thiamethoxam (0.005%), acetamiprid (0.004%), imidachloprid (0.005%), bifenthrin (0.016%) and carbosulfan (0.04%) were sprayed on mustard twigs infested with aphids; they were then allowed to properly dry. Thereafter, grubs of C. septempunctata collected from the field were exposed to insecticide-treated mustard twigs along with the aphids. Treated twigs and C. septempunctata grubs were covered with specially-designed cages to prevent coccinellid grubs from escaping. Mortality counts were made after 24 h AT and live grubs were collected for further study.

RESULTS AND DISCUSSION
Among the five different insecticides tested against the Delhi and Bikaner populations of L. erysimi using the leaf dip method, acetamiprid and thiamethoxam were found to be more toxic, whereas carbosulfan was less toxic than other insecticides. The LC50 value of the different insecticides in the Bikaner population was 7.0, 6.0, 4.0, 3.0 and 2.0 ppm for carbosulfan, bifenthrin, imidacloprid, acetamiprid and thiamethoxam, respectively (Table 1). Similarly, the ascending order of toxicity for the Delhi population...
was acetamiprid (7.0 ppm), thiamethoxam (9.0 ppm), imidacloprid (15.0 ppm), carbosulfan (32.0 ppm) and bifenthrin (37.0 ppm). The relative toxicity for the Bikaner population of *L. erysimi* suggested that thiamethoxam was 3.5, 3.0, 2.0 and 1.5 times more toxic than carbosulfan, bifenthrin imidacloprid and acetamiprid, respectively. Similarly, for the Delhi population, thiamethoxam was 3.55, 4.0 and 1.66 times more toxic than carbosulfan, bifenthrin and imidacloprid. The same trend was observed when we compared the toxicity of acetamiprid with other insecticides. Acetamiprid was 2.33, 2.0 and 1.33 times more toxic than carbosulfan, bifenthrin and imidacloprid to the Bikaner population. The Delhi population of *L. erysimi* was more tolerant to bifenthrin and carbosulfan than other insecticides. The relative toxicity shows that acetamiprid was 4.57, 5.14, 2.14 and 1.60 times more toxic than carbosulfan, bifenthrin, imidacloprid and thiamethoxam, respectively.

The Bikaner population of *L. erysimi* was found to be more susceptible to these insecticides than the Delhi population, which was 6.00, 4.57, 4.50, 3.75 and 2.33 times more tolerant to bifenthrin, carbosulfan, thiamethoxam, imidacloprid and acetamiprid, respectively. Furthermore, in both populations, acetamiprid showed more toxicity than other insecticides (Table 2).

In another study, the effect of field-recommended doses of insecticides on grubs of *C. septempunctata* showed that carbosulfan and bifenthrin were highly toxic and resulted in 100% mortality within 24 h. Neonicotinoids, thiamethoxam and acetamiprid were less toxic compared with imidacloprid. Pupal mortality was high when using thiamethoxam, whereas adult metamorphosis was found to be high with acetamiprid treatment. Imidacloprid had a negative effect on adult emergence. When we exposed adults of *C. septempunctata* to these insecticides, thiamethoxam resulted in less mortality; all other insecticides showed a negative effect within 24 h (Table 3).

In the present study, insecticides belonging to different chemical groups, namely imidacloprid, acetamiprid and thiamethoxam (neonicotinoids), bifenthrin (synthetic pyrethroid) and carbosulfan (carbamate), were studied for their efficacy against the mustard aphid, *L. erysimi*, and its associated predator, *C. septempunctata*. The results revealed that the susceptibility in both populations varied from one group to another, indicating that the mode of action of these insecticides is entirely different from one another. The superior efficacy of neonicotinoids such as acetamiprid and thiamethoxam in controlling the mustard aphid was reported by several authors. Maximum control of mustard aphid was obtained with the application of 100 g ha\(^{-1}\) thiamethoxam 25% WG, followed by 150 ml ha\(^{-1}\) imidacloprid 17.8% SL. Conventional insecticides were found less effective than newer insecticides for controlling mustard aphid (Kumar et al. 2013). The effective control of mustard aphid was achieved with the application of acetamiprid at 0.02% (Chinnabai al. 1999; Gour and Pareek 2003; Singh and Verma 2008; Singh and Singh 2009; Dhaka et al. 2008; Mandal and Mandal 2010). Imidacloprid and bifenthrin were 8.93 and 1.10 times more toxic than deltamethrin, respectively (Devee and Baruah 2010). The same trend was observed when we compared the toxicity of acetamiprid with other insecticides. Acetamiprid was 2.33, 2.0 and 1.33 times more toxic than carbosulfan, bifenthrin, imidacloprid and thiamethoxam, respectively.

### Table 1. Toxicity of different insecticides against the Delhi and Bikaner populations of the mustard aphid, *Lipaphis erysimi*.

<table>
<thead>
<tr>
<th>Insecticide</th>
<th>Heterogeneity*</th>
<th>Regression equation (Y)</th>
<th>LC(_{50}) (ppm)</th>
<th>Fiducial limit</th>
<th>Relative toxicity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Thiamethoxam</td>
</tr>
<tr>
<td>Bikaner</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Acetamiprid</td>
</tr>
<tr>
<td>Carbosulfan</td>
<td>7.564</td>
<td>Y=7.6704+0.8542x</td>
<td>7.0</td>
<td>0.0006-0.0010</td>
<td>3.50</td>
</tr>
<tr>
<td>Bifenthrin</td>
<td>12.63</td>
<td>Y=6.4545+0.4534x</td>
<td>6.0</td>
<td>0.0001-0.0011</td>
<td>3.00</td>
</tr>
<tr>
<td>Imidacloprid</td>
<td>10.436</td>
<td>Y=8.5145+1.0334x</td>
<td>4.0</td>
<td>0.0001-0.0007</td>
<td>2.00</td>
</tr>
<tr>
<td>Acetamiprid</td>
<td>6.2683</td>
<td>Y=8.3053+1.046x</td>
<td>3.0</td>
<td>0.0001-0.0005</td>
<td>1.50</td>
</tr>
<tr>
<td>Thiamethoxam</td>
<td>12.585</td>
<td>Y=8.7865+1.2457x</td>
<td>2.0</td>
<td>0.0001-0.0003</td>
<td>1.00</td>
</tr>
<tr>
<td>Delhi</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Acetamiprid</td>
</tr>
<tr>
<td>Carbosulfan</td>
<td>6.8041</td>
<td>Y=7.6426+1.0566x</td>
<td>32.0</td>
<td>0.0024-0.0041</td>
<td>3.55</td>
</tr>
<tr>
<td>Bifenthrin</td>
<td>3.3495</td>
<td>Y=7.6447+1.0821x</td>
<td>37.0</td>
<td>0.0028-0.0047</td>
<td>4.00</td>
</tr>
<tr>
<td>Imidacloprid</td>
<td>4.5940</td>
<td>Y=8.4937+1.2310x</td>
<td>15.0</td>
<td>0.0012-0.0017</td>
<td>1.66</td>
</tr>
<tr>
<td>Acetamiprid</td>
<td>6.2683</td>
<td>Y=8.3053+1.0464x</td>
<td>7.0</td>
<td>0.0050-0.0009</td>
<td>0.77</td>
</tr>
<tr>
<td>Thiamethoxam</td>
<td>12.585</td>
<td>Y=8.7865+1.2457x</td>
<td>9.0</td>
<td>0.0050-0.0014</td>
<td>1.00</td>
</tr>
</tbody>
</table>

Data were not found to be significantly heterogeneous at \( P = 0.05 \). \( Y = \) Probit kill; \( x = \) log concentration; \( LC = \) Lethal concentration to induce 50% mortality; Relative toxicity = \( LC_{50} \) of reference insecticide / \( LC_{50} \) of standard insecticide.

### Table 2. Relative resistance of the Bikaner and Delhi populations of mustard aphid to various synthetic insecticides

<table>
<thead>
<tr>
<th>Insecticide</th>
<th>LC(_{50}) (ppm)</th>
<th>Relative resistance*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Delhi</td>
<td>Bikaner</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Delhi</td>
</tr>
<tr>
<td>Carbosulfan</td>
<td>7.0</td>
<td>32.0</td>
</tr>
<tr>
<td>Bifenthrin</td>
<td>6.00</td>
<td>37.0</td>
</tr>
<tr>
<td>Imidacloprid</td>
<td>4.0</td>
<td>15.0</td>
</tr>
<tr>
<td>Acetamiprid</td>
<td>3.0</td>
<td>7.0</td>
</tr>
<tr>
<td>Thiamethoxam</td>
<td>2.0</td>
<td>9.0</td>
</tr>
</tbody>
</table>

*Relative resistance = \( LC_{50} \) of *L. erysimi* from Delhi / \( LC_{50} \) of *L. erysimi* from Bikaner
L. was reported by Gesraha (2007). The newer insecticides that were found effective in controlling mustard aphid were also found to be safe for coccinellid populations, just like conventional insecticides. However, Mhaske et al. 2007 found that imidacloprid was biologically safer for predatory coccinellids. Under field conditions, the use of acetamiprid and thiamethoxam is recommended for a safe and effective management of the mustard aphid, and is found to be the least toxic to ladybird beetle populations (Dhaka et al. 2009; Sohail 2011). However, under field conditions, synthetic pyrethroids tend to eliminate non-target insects. Bifenthrin was found to be more toxic to C. undecim-punctata, and both bifenthrin and carbaryl tended to cause 100% mortality in the aphidophagous lady-beetle, Harmonia axyridis (Pallas.), at concentrations equivalent to field rates (James 2003). A decrease in the population of coccinellids and spiders was observed in bifenthrin-treated cotton fields (Balakrishnan et al. 2009).

**CONCLUSION**

The most important aspect of this study is that mustard-growing farmers facing serious yield losses due to the mustard aphid tend to mostly rely on conventional insecticides. However, this study shows that the new insecticides are efficient at controlling aphids while being relatively safe to their natural enemy, *C. septempunctata*, something that is considered to be of great importance. Consequently, the use of these insecticides in mustard has the potential to greatly improve pest management by increasing the conservation of biological control agents (James et al. 2001). Substitution of the currently used conventional insecticides with newer insecticides should go a long way towards minimizing disturbance in beneficial insects and improving biological control in mustard.

Among the five different insecticides tested, acetamiprid and thiamethoxam were found to be more toxic and carbosulfan was the less toxic to both populations of mustard aphid. Furthermore, the Delhi population of *L. erysimi* proved to be more tolerant to bifenthrin and carbosulfan than to other insecticides. Carbosulfan and bifenthrin were highly toxic to grubs of *C. septempunctata*, but in the case of neonicotinoids, thiamethoxam and acetamiprid, they were less toxic as compared with imidacloprid. In conclusion, the use of neonicotinoids for the management of mustard aphid would have less impact on non-target coccinellid beetles. The present study should be validated with further field studies in order to confirm the most efficient pest management strategy.

**ACKNOWLEDGEMENTS**

The authors are highly thankful for the help received from Head, Division of Entomology and Director, IARI, New Delhi.

**REFERENCES**


Table 3. Effect of different field-recommended doses of insecticides on grubs and adults of *Coccinella septempunctata*

<table>
<thead>
<tr>
<th>Insecticide</th>
<th><em>C. septempunctata</em> grub 24 h AT</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% grub mortality</td>
<td>% pupal mortality</td>
<td>% adult emergence</td>
<td>% adult mortality</td>
</tr>
<tr>
<td>Thiamethoxam</td>
<td>30.00 (32.71)c</td>
<td>65.83 (54.34)c</td>
<td>30.00 (32.71)c</td>
<td>20.00 (22.77)b</td>
</tr>
<tr>
<td>Acetamiprid</td>
<td>10.00 (18.43)b</td>
<td>59.26 (51.13)b</td>
<td>36.67 (36.14)b</td>
<td>66.67 (60.00)d</td>
</tr>
<tr>
<td>Imidacloprid</td>
<td>83.33 (70.78)d</td>
<td>58.33 (50.00)b</td>
<td>3.333 (6.14)d</td>
<td>73.33 (66.35)c</td>
</tr>
<tr>
<td>Bifenthrin</td>
<td>100.00 (90.00)e</td>
<td>-</td>
<td>-</td>
<td>100.00 (97.50)e</td>
</tr>
<tr>
<td>Carbosulfan</td>
<td>100.00 (90.00)e</td>
<td>-</td>
<td>-</td>
<td>100.00 (97.50)e</td>
</tr>
<tr>
<td>Control</td>
<td>0.00 (2.50)a</td>
<td>0.00 (2.50)a</td>
<td>100.00 (97.50)a</td>
<td>0.00 (2.50)a</td>
</tr>
</tbody>
</table>

SEm±: 3.68 8.47 4.93 7.21

Figures in parentheses are the arc sine transformed values.

Data analyzed with least squares means, means separated using LSD.


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