Arthropod Abundance and Diversity in Bt and Non-Bt Rice Fields

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Arthropod Abundance and Diversity in Bt and Non-Bt Rice Fields

FANG-FANG LI,1 GONG-YIN YE,1 QIONG WU,1 YU-FA PENG,2 AND XUE-XIN CHEN1,3


ABSTRACT In a field experiment, possible effects of transgenic Bt rice on arthropod communities under paddy field conditions were assessed for 3 yr in terms of arthropod guild dominance, family composition, dominance distribution of each guild, individuals of each guild, and community indices (including Shannon-Weaver diversity index and dominant concentration index). Our results overall suggested no significant differences between the Bt and control rice plots in these arthropod community-specific parameters. The similarity of arthropod communities in the Bt and control rice plots was apparently high. Based on our findings, we conclude that Bt rice generally exerts no marked negative effects on the arthropod community in paddy fields.

KEY WORDS transgenic, Bt rice, biodiversity, species richness, Shannon-Weaver diversity index

With the global population steadily growing, the amount of arable land is steadily decreasing. Thus, it is essential that sustainable strategies be implemented to use agricultural resources more efficiently to yield an abundant healthy diet. Experience to date with genetically engineered crops has shown that this technology can make substantial contributions toward this goal. Among the first transgenic crops approved for release were Bt maize and Bt cotton, which contain genes encoding insecticidal proteins from the bacterium Bacillus thuringiensis Berliner. These crops have been readily adopted by farmers, have resulted in increased yields and reductions in insecticide applications, and have been sustainable when used with resistance management programs (Shelton et al. 2002). However, emerging controversies surrounding the risks and benefits of this novel technology have limited its benefits in many parts of the world (Obrzycki et al. 2001, Schuler et al. 2001, Groot and Dicke 2002, Conner et al. 2003, Carrière et al. 2004).

Rice is the target crop for many improvement programs because it is the staple diet for nearly two billion people worldwide and the major food for over one half of those living in Asia (Khush 1997). In 2002, rice production in China reached 177 million tons, of which 3.1 million tons was exported, producing revenues of U.S. $578 million (http://www.irri.org/science/ricestat/index.asp). These numbers represent only a fraction of what might be available if rice plants were not subject to insect attack. Many rice varieties have been genetically transformed with genes encoding various Bt crystal (Cry) proteins and have been shown to be resistant to one or more lepidopteran pests, the most important of which are the yellow stem borer [Scirpophaga incertulas (Walker)], the striped stem borer [Chilo suppressalis (Walker)], and several species of leaf-folders [Marasmia spp. and Cnaphalocrocis medinalis (Guénot)] (Shu et al. 2000, Tu et al. 2000, Ye et al. 2001a, 2001b, 2003). Field trials of Bt rice started in China in 1998, but no Bt rice or other transgenic rice varieties have yet been released for commercialization.

The toxins produced by Bt plants kill only a narrow range of insect species. Many rice varieties have been transformed with genes encoding various Bt crystal (Cry) proteins and are resistant to one or more lepidopteran pests of rice. Thus, one might expect decreased abundance of susceptible lepidopterans and their specialist natural enemies. However, the potential for broader effects exists. Many herbivores may consume the toxin and survive. Predators that consume such herbivores may be adversely affected. For example, Chrysoperla carnea (Stephens) that consumed Spodoptera littoralis (Boisdhuix) reared on a diet with Bt toxin had lower survival rate than C. carnea that fed on S. littoralis reared on a diet without Bt toxin (Hillbeck et al. 1999). Such interactions are likely to be complex, because neither all herbivores that feed on Bt plants uptake the toxin nor are natural enemies always affected by consuming prey that have taken up the toxin (Dutton et al. 2002).

Many studies related to the impact of Bt crops on nontarget organisms have examined the interaction of one or a few species in the laboratory (Sims 1995, Hillbeck et al. 1998a, 1998b, 1999, Zwahlen et al. 2000, Dutton et al. 2002). Some results indicated no significant effects on the nontarget organisms, whereas some found there were negative effects on the natural enemies. However, translating laboratory results to

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the field may be problematic because the concentration of toxin doses used in the laboratory may be higher than the doses that the arthropods encounter in the field, and highly mobile species may spend only a fraction of their life in Bt fields. Despite these limitations, laboratory studies can provide valuable insights into potential effects and the causal mechanisms of patterns in field data.

Most field studies assessing potential impacts of Bt crops have focused on a limited numbers of species (Wilson et al. 1992, Hardee and Bryan 1997, Orr and Landis 1997, Pilcher et al. 1997, Pilcher 1999, Wold et al. 2001, Jasimniski et al. 2003, Liu et al. 2003, Men et al. 2003, Schoenly et al. 2003). Here, we surveyed all of the arthropods on Bt and non-Bt rice at three field sites in Zhejiang Province of China: two in 3 yr and one in 2 yr. Our primary goal was to determine whether major differences occur between the plant-dwelling arthropod community on transgenic Bt and non-Bt rice.

Materials and Methods

Transgenic Bt Rice Lines and Their Parental Cultivars. Two lines of transgenic indica rice with the cry1Ab gene, namely B1 and B6, were tested along with their corresponding untransformed parental cultivar Jiazao935. Two homogenous lines of transgenic indica rice containing the cry1Ab/cry1Ac genes, i.e., TT9-3 and TT9-4, were tested along with their corresponding untransformed parental cultivar IR72. One line of transgenic japonica rice containing the cry1Ab gene, KMD1, was tested along with its corresponding untransformed parental cultivar Xiushui11.

Field Experiments. The experiments were conducted in 2003, 2004, and 2005 at three different sites where field trials of Bt rice are permitted in Zhejiang province in China.

The first set of experiments was conducted at the Experimental Farm of Zhejiang University at Hangzhou (120.19 E, 30.26 N), consisting of three plots each of B1, B6, and the control line (Jiazao935). Plot sizes were 20 by 25 m.

The second set was planted at a local experimental field in the suburbs of Jiande (119.27 E, 29.49 N), consisting of three plots each of TT9-3, TT9-4, and the control line IR72. Plot sizes were 20 by 35 m.

The third set was carried out at the Experimental Farm of the China Rice Research Institute at Fuyang (119.95 E, 30.07 N), with three plots each of KMD1 and the Xiushui11 control. Plot sizes were 20 by 35 m.

All experiments were designed as randomized complete block with three replications for each treatment. For each experiment of every year at each test site, seedlings were manually transplanted with one seedling per plant or hill spaced 16.5 by 16.5 cm apart. Each experimental plot was bordered on all sides by an unplanted walkway 50 cm wide, and the entire experimental field of each location was planted with five border rows of the nontransgenic rice plants.

Agronomic practices such as fertilization and irrigation for growing rice were the same as followed by local farmers.

Sampling. Numbers of arthropods were sampled randomly at each site every 15 d during the rice growing season each year. Arthropods on the plants were collected by vacuum-suction machine, modified according to Carino et al. (1979), and were taken to the laboratory, stored in 95% ethanol, identified to species, and counted.

Data Analysis. Most of the arthropods sampled were identified to species. The Shannon-Weaver diversity index $H'$ and dominance concentration index $C$ were calculated. The Shannon-Weaver diversity index $H'$ (Shannon and Weaver 1949),

$$H' = -\sum pi \log pi$$

where $pi$ is the proportion of the ith species in the total sample and was calculated as a measure of the arthropod community diversity (all species of insects) and pest and natural enemy subcommunities in the treatment plots. The dominance concentration index was calculated as $C = \sum (pi)^2$.

The arthropods were split into five guilds: phytophages, parasitoids, predators, detritivores, and others. The dominance distribution is the percentage of each subcommunity among the total communities. All data on population densities of insects from different treatments in the field were analyzed using analysis of variance (ANOVA), and means were compared with the least significant difference (LSD) as calculated by SAS software (SAS Institute 2001).

<table>
<thead>
<tr>
<th>Site</th>
<th>Year</th>
<th>Line</th>
<th>$H'$</th>
<th>$C$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hangzhou</td>
<td>2003</td>
<td>B1</td>
<td>3.156</td>
<td>0.132</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B6</td>
<td>3.069</td>
<td>0.161</td>
</tr>
<tr>
<td></td>
<td></td>
<td>935</td>
<td>3.070</td>
<td>0.172</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>B1</td>
<td>3.465</td>
<td>0.107</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B6</td>
<td>3.266</td>
<td>0.121</td>
</tr>
<tr>
<td></td>
<td></td>
<td>935</td>
<td>3.277</td>
<td>0.125</td>
</tr>
<tr>
<td></td>
<td>2005</td>
<td>B1</td>
<td>3.266</td>
<td>0.132</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B6</td>
<td>3.099</td>
<td>0.167</td>
</tr>
<tr>
<td></td>
<td></td>
<td>935</td>
<td>3.142</td>
<td>0.174</td>
</tr>
<tr>
<td>Jiande</td>
<td>2003</td>
<td>TT9-3</td>
<td>1.479</td>
<td>0.515</td>
</tr>
<tr>
<td></td>
<td></td>
<td>TT9-4</td>
<td>1.536</td>
<td>0.474</td>
</tr>
<tr>
<td></td>
<td></td>
<td>IR72</td>
<td>1.685</td>
<td>0.431</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>TT9-3</td>
<td>2.518</td>
<td>0.225</td>
</tr>
<tr>
<td></td>
<td></td>
<td>TT9-4</td>
<td>2.140</td>
<td>0.322</td>
</tr>
<tr>
<td></td>
<td></td>
<td>IR72</td>
<td>2.905</td>
<td>0.182</td>
</tr>
<tr>
<td></td>
<td>2005</td>
<td>TT9-3</td>
<td>2.451</td>
<td>0.227</td>
</tr>
<tr>
<td></td>
<td></td>
<td>TT9-4</td>
<td>2.161</td>
<td>0.333</td>
</tr>
<tr>
<td></td>
<td></td>
<td>IR72</td>
<td>2.358</td>
<td>0.291</td>
</tr>
<tr>
<td>Fuyang</td>
<td>2004</td>
<td>KMD1</td>
<td>2.436</td>
<td>0.224</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Xiushui11</td>
<td>1.645</td>
<td>0.429</td>
</tr>
<tr>
<td></td>
<td>2005</td>
<td>KMD1</td>
<td>1.469</td>
<td>0.535</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Xiushui11</td>
<td>1.352</td>
<td>0.566</td>
</tr>
</tbody>
</table>

Means ± SE within a column per site and year followed by the same letter are not significantly different based on ANOVA (LSD test, $P \leq 0.05$). $H'$ and $C$ stand for Shannon-Weaver diversity index and dominance conen index, respectively.
Results

Abundance of Arthropods in Rice Fields. Overall, 17,706 individual insects and spiders were recorded in 3 yr in both Bt and non-Bt rice fields.

In Hangzhou, a total 62 species of insects and some spiders in 53 families were observed in the experimental plots during the 3 yr of study. The total number of arthropod individuals in 3 yr at Hangzhou was 4,014 (766, 1,239, and 2,009 in 2003, 2004, and 2005, respectively).

In Jiande, 40 species insects and some spiders in 37 families were observed in the experimental plots dur-

![Temporal dynamics of main indices of arthropod community diversity in plots containing different lines of rice at Hangzhou, China, in 2003, 2004, and 2005. (A–C) Diversity indices. (D–F) Dominant concentration indices. Bars are 95% CIs.](https://bioone.org/journals/Environmental-Entomology)
ing the 3 yr of study. The total number of arthropods in 3 yr at Jiande was 3,800 (1,694, 1,006, and 1,100 in 2003, 2004, and 2005, respectively).

We conducted studies in Fuyang for only 2 yr. A total 46 species of insects and some spiders in 41 families were observed in the experimental plots during the 2 yr of study. The total number of arthropods in 2 yr at Fuyang was 9,892 (1,798 and 8,094 in 2004 and 2005, respectively).

For 3 yr, the total number of arthropods in Bt and non-Bt rice at the three sites were not significantly different. At Hangzhou, the abundance of arthropods in Bt rice was a little lower than in non-Bt rice. In 2003, the number of arthropods in B1 rice was 231, in B6 rice was 186, and in the control (Jiazaor935) was 347. In 2004, there were 369 individuals in B1, 429 in B6, and 391 in the control. In 2005, there were 423 individuals in B1, 620 in B6, and 915 in the control.

At Jiande, the abundance of arthropods in Bt rice was a little higher than in non-Bt rice. In 2003, the number of arthropods in TT9-3 rice was 675, in TT9-4 rice was 588, and in the control (IR72) was 431. In 2004, there were 431 in TT9-3, 342 in TT9-4, and 233 in the control. In 2005, there were 449 in TT9-3, 337 in TT9-4, and 314 in the control.

At Fuyang, the same trend was observed as in Hangzhou. The number of arthropods in Bt rice did not differ significantly from that in non-Bt rice. In 2004, the number of arthropods in KMD rice was 948, whereas in the control (Xiushui11) was 850. In 2005, there were 4,137 in KMD and 3,957 in the control.

Species Richness and Diversity. Some common community indices (including Shannon-Weaver diversity index $H'$ and dominance concentration index $C$) calculated for different plots during the 3 yr are shown in Table 1. Analysis of these indices revealed no significant differences between Bt and non-Bt plants within paddy fields (Table 3).

The temporal dynamics of these main indices of arthropod community diversity revealed the differences between the Bt and non-Bt rice. At Hangzhou, the dominant index of the non-Bt rice was a little higher than those of Bt rice (least significant difference [LSD] test: $P \leq 0.05$ each comparison). The diversity index did not differ significantly between these two plots (Fig. 1). However, the diversity index of the non-Bt rice arthropods in Jiande was a little higher than those of Bt rice (LSD test: $P \leq 0.05$ each comparison), with no significant difference in the dominant index (Fig. 2). At Fuyang, the results of the study revealed no significant differences between non-Bt and Bt rice (LSD test: $P \leq 0.05$ each comparison; Fig. 3).

Dominance Distribution. The arthropods were separated into five guilds: phytophages, parasitoids, predators, detritivores, and others. The Phytophagous subcommunity includes Cicadellidae, Delphacidae, Aphididae, Thripidae, Itonididae, Trypetidae, Agromyzidae, Drosophilidae, Ephyridae, Chloropidae, Pyrali-
Table 2. Dominance distribution of arthropod subcommunities in Bt (B1, B6, TT9-3, TT9-4, and KMD1) and non-Bt rice lines

<table>
<thead>
<tr>
<th>Sites</th>
<th>Year</th>
<th>Line</th>
<th>Phytophagous</th>
<th>Parasitoids</th>
<th>Predators</th>
<th>Detritivores</th>
<th>Others</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hangzhou</td>
<td>2003</td>
<td>B1</td>
<td>0.153 ± 0.126A</td>
<td>0.244 ± 0.069A</td>
<td>0.049 ± 0.038A</td>
<td>0.313 ± 0.061A</td>
<td>0.240 ± 0.155A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B6</td>
<td>0.180 ± 0.143A</td>
<td>0.149 ± 0.073A</td>
<td>0.028 ± 0.005A</td>
<td>0.396 ± 0.150A</td>
<td>0.247 ± 0.181A</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>935</td>
<td>0.171 ± 0.135A</td>
<td>0.228 ± 0.052A</td>
<td>0.019 ± 0.009A</td>
<td>0.312 ± 0.175A</td>
<td>0.270 ± 0.259A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B1</td>
<td>0.370 ± 0.122A</td>
<td>0.049 ± 0.103A</td>
<td>0.125 ± 0.070A</td>
<td>0.182 ± 0.064A</td>
<td>0.270 ± 0.187A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B6</td>
<td>0.403 ± 0.122A</td>
<td>0.028 ± 0.019A</td>
<td>0.075 ± 0.053A</td>
<td>0.185 ± 0.059A</td>
<td>0.310 ± 0.104A</td>
</tr>
<tr>
<td></td>
<td>2005</td>
<td>935</td>
<td>0.465 ± 0.089A</td>
<td>0.052 ± 0.037A</td>
<td>0.043 ± 0.035A</td>
<td>0.124 ± 0.079A</td>
<td>0.316 ± 0.131A</td>
</tr>
<tr>
<td>Jiaode</td>
<td>2003</td>
<td>TT9-3</td>
<td>0.817 ± 0.101A</td>
<td>0.016 ± 0.009A</td>
<td>0.178 ± 0.092A</td>
<td>0.022 ± 0.034A</td>
<td>0.020 ± 0.013A</td>
</tr>
<tr>
<td></td>
<td>2004</td>
<td>TT9-3</td>
<td>0.758 ± 0.152A</td>
<td>0.021 ± 0.019A</td>
<td>0.178 ± 0.092A</td>
<td>0.022 ± 0.034A</td>
<td>0.020 ± 0.013A</td>
</tr>
<tr>
<td></td>
<td>2005</td>
<td>TT9-3</td>
<td>0.672 ± 0.253A</td>
<td>0.022 ± 0.013A</td>
<td>0.225 ± 0.220A</td>
<td>0.036 ± 0.032A</td>
<td>0.450 ± 0.006A</td>
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<td></td>
<td></td>
<td>TT9-4</td>
<td>0.559 ± 0.279A</td>
<td>0.037 ± 0.025A</td>
<td>0.227 ± 0.269A</td>
<td>0.025 ± 0.019A</td>
<td>0.152 ± 0.192A</td>
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<td></td>
<td></td>
<td>IR72</td>
<td>0.572 ± 0.217A</td>
<td>0.034 ± 0.019A</td>
<td>0.101 ± 0.062A</td>
<td>0.098 ± 0.151A</td>
<td>0.203 ± 0.063A</td>
</tr>
<tr>
<td>Fuyang</td>
<td>2004</td>
<td>KMD1</td>
<td>0.485 ± 0.385A</td>
<td>0.031 ± 0.027A</td>
<td>0.039 ± 0.027A</td>
<td>0.023 ± 0.012A</td>
<td>0.442 ± 0.386A</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Xinshui1</td>
<td>0.842 ± 0.321A</td>
<td>0.024 ± 0.016A</td>
<td>0.063 ± 0.046A</td>
<td>0.042 ± 0.014A</td>
<td>0.459 ± 0.347A</td>
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<tr>
<td></td>
<td>2005</td>
<td>KMD1</td>
<td>0.852 ± 0.112A</td>
<td>0.003 ± 0.002A</td>
<td>0.055 ± 0.049A</td>
<td>0.017 ± 0.012A</td>
<td>0.073 ± 0.069A</td>
</tr>
</tbody>
</table>

Means (± SE) within column followed by the same letter are not significantly different based on ANOVA (Least significant difference test, P ≤ 0.05).


There was almost significant dominance of phytophagous subcommunities at each site during the 3 yr, except 2003 at Hangzhou. Parasitoids and detritivores displayed the second highest level of dominance among the subcommunities. There was no significant difference in subcommunity composi-
tion between the Bt rice and non-Bt rice (Tables 2 and 4; Fig. 4).

Discussion

The number of arthropod species found in this study was obviously lower than those of other studies from tropical area (Settle et al. 1996, Schoenly et al. 1998). This was probably caused by the different study locations and differences in sampling strategies. Our study area was located near the northern border of the subtropics where species biodiversity is believed much less than that in the tropics.

We found that plots of Bt rice and non-Bt rice did not differ significantly in diversity or dominance distribution of arthropods. Thus, the similarity of arthropod communities in Bt and control rice plots was apparently high. It was quite clear that Bt rice generally had no marked negative effects on the arthropod community in the paddy fields, a result similar to that of Liu et al. (2003) for cry1Ab/cry1Ac-carrying transgenic Bt rice and non-Bt rice.

Laboratory studies suggest that the effects of Bt crops on natural enemies may depend on whether the prey or host takes up the toxin or is affected by the toxin (Romeis et al. 2006). Accordingly, no impact is expected for natural enemies attacking phloem-feeding herbivores (Dutton et al. 2002, Obrycki et al. 2003). If the natural enemy is a specialist on an herbivore that has high susceptibility to the toxin, the natural enemy’s numbers are likely to decline in the field because of the absence of the host (Schuler et al. 2001, Andow and Hilbeck 2004). In our results, we did not find a difference in the abundance of parasitoids and predators among treatments (Table 2). This might be because of the target lepidopteran pests having a significantly lower dominance distribution among all arthropod phytophages, whereas nontarget pests like Cicadellidae and Delphacidae were more dominant. It has been suggested that Bt rice does not adversely affect nontarget planthopper and leafhopper populations and poses no potential risk of increasing their occurrence or damage (Chen et al. 2003, 2006). Therefore, the effects of Bt rice on the target pests and their

Fig. 4. Dominance distribution of sub-communities of arthropods in plots containing different lines of rice. (A–C) Hangzhou. (D–F) Jiande. (G–H) Fuyang. Bars are 95% CIs.
natural enemies may have been masked by the dominant nontarget pests. The effect of transgenic plants on arthropod communities has been examined in other crops. Men et al. (2003) indicated that Bt-cotton increased the diversity of arthropod communities and pest subcommunities; however, it decreased the diversities of natural enemy subcommunities. Sisterson et al. (2004) showed minor differences in the arthropod community between Bt and non-Bt cotton. A long-term study showed essentially no effects of Bt cotton on natural enemy function in Arizona cotton, and in a companion study, minor reductions in density of several predator taxa in Bt cotton were observed (Naranjo 2005). The abundance of canopy- and ground-dwelling predators across 3 yr in cotton fields was not negatively impacted by Bt cotton (Torres and Ruberson 2005). A slight difference was shown between the total community in un sprayed conventional and Bt cotton in Australia (Whitehouse et al. 2005). In China, the diversity of arthropod communities in transgenic cotton plots was similar to that in conventional cotton fields without spraying (Li et al. 2004). In corn, numbers of soil mites, Collembola, and nematodes were similar in soil planted with Bt corn and soil planted with its isolate (Al-deeb et al. 2003). Candolfi et al. (2004) observed no effects of Bt corn on the communities of soil-dwelling and nontarget plant-dwelling arthropods. Dively (2005) found that densities of most nontarget taxa exposed to the Bt corn showed no differences compared with the isogenic control. Bitzer et al. (2005) observed few differences in the abundance of individual Collemobola species in Bt and isolate corn, nor did the estimated species richness or the Shannon or Simpson diversity indices differ significantly between Bt and isolate corn at two locations over 3 yr, indicating no effect of Bt corn on springtail diversity. Furthermore, the use of insecticide to control the same pests more strongly impacts springtails and their predators than did the transgenic crop (Bitzer et al. 2005).

In conclusion, our findings suggest that the Bt rice generally has no marked negative effects on the arthropod community in paddy fields. We acknowledge several limitations to our study, in that we did not study arthropods living on the ground or in the soil and water, and our sample sizes were not large enough to enable rigorous comparisons between Bt and non-Bt rice for rare families of arthropods. The generality of our results can be enhanced by experimentally addressing these issues.

Acknowledgments

We thank S. K. Datta (International Rice Research Institute, Manila, Philippines) for providing two Bt rice lines (TT9-3 and TT9-4) and I. Altilosa (University of Ottawa, Ottawa, Canada) for providing the cry1Ab gene for developing two Bt rice lines (KMD1 and KMD2). We also thank two anonymous reviewers for valuable comments. This project was supported by ‘973’ Projects Grant (001CB109004) from the Ministry of Science and Technology of China and the Innovation Research Team Program of the Ministry of Education of China (IRT0355).

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Cnaphalocrocis medinalis (Guenée) under field conditions. Crop Prot. 22: 171–178.


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