Disruption of Sex Pheromone Communication in the Rice Stem Borer Moth, *Chilo suppressalis* WALKER (Lepidoptera: Pyralidae), with Sex Pheromone Components and Their Analogues

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In the rice stem borer moth, *Chilo suppressalis* WALKER, the influence of the sex pheromone components, Z-11-hexadecenal (Z-11-HDAL) and Z-13-octadecenal (Z-13-ODAL), and their eight structurally related compounds on sexual communication between male and female was studied under field conditions. Relatively good inhibition of male attraction could be achieved with most of the synthetic test compounds except acetates. Among test compounds, Z-11-HDAL, major component of the sex pheromone, and Z-5-hexadecene (Z-5-HD) were most effective as inhibitors on male attraction. These compounds have been shown to cause over 90 percent reduction of males caught in the traps baited with virgin females. In the mating suppression test, all of the four test compounds, sex pheromone components and hydrocarbons, significantly suppressed the mating. Z-11-HDAL and Z-5-HD had a highly suppressive effect on mating. It was also shown that the disruptive effect of Z-11-HDAL on male attraction was apparently higher than Z-5-HD although the release rate of Z-11-HDAL was lower than Z-5-HD in comparative field test. Nevertheless, it seems that Z-5-HD is more suitable for practical use than Z-11-HDAL, because of chemical stability and simplicity in the synthesis. Z-5-HD decreased injury to the plants when it was treated in wide areas.

INTRODUCTION

The rice stem borer, *Chilo suppressalis* WALKER, is one of the most serious pests of the rice plant throughout the Far-East and South-East Asia. Larval stem boring results in extensive damage and crop loss. A large quantity of insecticides is used
annually for control in these areas, but the use of insecticides can lead to insect resistance and environmental problems. Identification and synthesis of the female sex pheromone complex was undertaken in the hope that the synthetic pheromones would be of value for population monitoring and for direct control by mating disruption techniques. The female sex pheromone of this insect was identified as two olefinic aldehydes, Z-11-hexadecenal (Z-11-HDAL) and Z-13-octadecenal (Z-13-ODAL) (Nesbitt et al., 1975; Ohta et al., 1976). It was confirmed that the synthetic Z-11-HDAL and Z-13-ODAL were attractive when they were mixed together in ratios of 1:1 to 20:1, especially 3:1, 5:1 and 7:1 being most effective (Tatsuki et al., 1977).

As reviewed by Roelofs and Cardé (1977), the sex pheromone consists of multiple components in many lepidopterous species, and each of the pheromone components have been used as "attraction disruptants", as well as the sex pheromones and their analogues. The authors also confirmed that the pheromone components of Chilo suppressalis and their twelve structurally related compounds disrupted male attraction when the compounds were set with virgin females in the traps (Kanno et al., 1978). In Spodoptera frugiperda (Mitchell et al., 1974) and Cydia molesta (Rothschild, 1974), however, it was observed that some kinds of non-pheromone chemicals inhibited male attraction to the traps baited with virgin females when they were dispensed simultaneously from the traps did not block pheromone communication when they were evaporated into the atmosphere surrounding the traps.

In the present study, therefore, the authors investigated the disruptive effect of atmospheric permeation with the sex pheromone components and closely related compounds which were selected in the previous screening test. In particular, the comparative test on the effects of the pheromone components and the hydrocarbons which had a high effect in the previous test was conducted in detail. Furthermore, we also attempted a preliminary test to see whether the crop damage by the larvae of the next generation could be decreased with Z-5-hexadecene (Z-5-HD).

MATERIALS AND METHODS

Insect used. The female moths used in this study as the attractive source and the tethered females were reared successively in bottles (750 cm³ in volume) at 25°C, 16 hr illumination. Rice seeds, variety Todorokiwase, were germinated in the bottles and served as food. Pupae were collected from the rearing bottles, and separated by sex and held in different cages to prevent mating after moth emergence.

Test compounds. All of the test compounds were synthesized at the Nissan Chemical Co., Ltd. in accordance with synthetic procedure described in Kanno et al. (1978). Purities of the test compounds were determined to be 65–70 percent except Z-5-HD, which was 95 percent pure in 1978, and more than 90 percent in 1979 by high-pressure liquid chromatography (LiChrosorb SI60 impregnated with 15 percent AgNO₃, 4 × 50 cm; 30:1 mixtures of n-hexane and ethyl acetate as developing solvent). The chemical structures of the test compounds are shown in Fig. 1.

Attraction disruption test. This test was conducted in the paddy field in Kubiki village, Niigata prefecture in June, 1978. The pheromone components and their eight structurally related compounds that showed relatively potent inhibition in the previous test were used. Twelve polyethylene capsules (BEEM No. 00 Reg Tip) each containing 100 mg of each compound were evenly spaced in two concentric circles.
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![Chemical structures of the test compounds.](image)

1 and 2: sex pheromone components. 3 and 4: geometrical isomers of 1,2. 5 and 6: EAG active (OHTA et al., 1976). 7 and 8: alcohols corresponding to 1, 2. 9 and 10: acetates of 7, 8.

of 1 and 2 m radius (Test A) or one circle of 5 m (Test B) with a virgin female baited trap at the center. As a control, the polyethylene capsules treated with solvent alone were used. The plots were placed at intervals of 200 m in two parallel straight lines, for duplication test, which were about 300 m apart. Both test A and B were run for five nights without renewing the capsules. The numbers of males caught in the traps each night were counted the next day.

**Mating suppression test.** This test was conducted at the same field as above mentioned in June, 1979. The pheromone components and the hydrocarbons, Z-5-HD and Z-5-octadecene (Z-5-OD), which had strong effect in the attraction disruption test were examined to determine whether they suppress mating or not. 10 mg of each compound was soaked in a polyethylene capsule, and then the capsule was heat-sealed to prevent the compound from leaking out rapidly. Twelve sealed capsules of each compound were set in a circle of 3 m radius at about 30 cm above the rice plants, and in a concentric 2 m circle fifteen virgin female each tethered with a cotton thread to a plastic rod were also evenly spaced. The moths were exchanged for new ones every day during the test period and the collected ones were then dissected to determine successful mating by observation on spermatophore. The plots were also designed at about 200 m intervals in duplication. The locations of compounds were changed every two nights to reduce the possibility of bias due to location.

**Evaluation of disruptive activity of Z-11-HDAL and Z-5-HD.**

This test was also carried out in Kubiki in June, 1979. 0.5 ml of the hexane solution with each amount of a compound, such as 0.1, 0.3, 1, 3 and 10 mg, was dispensed on a rubber septum. For the control, the solvent alone was treated on a septum. Six rubber septa treated with each amount of a compound were spaced in a circle of 1 m
radius with a virgin female baited trap at the center. The plots were separated from each other by a 200 m “buffer”, and was designed in duplication. The location of the compounds and of the control were also changed every two nights for re-randomization. This test was run for eight nights and the number of males caught in the traps each night were counted the next day.

Large scale communication disruption test with Z-5-HD.

This test was also conducted in the paddy field in Kubiki in the summer of 1979. For the test, a total of six experimental blocks were used, two of which were treated with the compound and the other four were untreated controls. Each block was nearly 2000 m² and separated from each other by 200 to 300 m. The two treated areas were ca. 600 m apart from each other. The compound was impregnated in synthetic rubber films (40 percent Z-5-HD in styrene-isoprene copolymer, 60 μm thickness), a side of which was covered with polyethylene films (25 μm thickness) as barriers. The laminated films were cut into 1×4 cm strips for use as dispensers. The dispensers were attached to fishing-lines at 1 m intervals and strung over the test field at 1.5 m intervals, so that the dispensers were suspended at 1×1.5 m intervals and 20 to 30 cm above the plants. Total numbers of the dispensers used in each treated area were 1221 for block I (2002 m²) and 1120 for block II (1999 m²), respectively. The treatment was continued from July 31 to August 25. This period almost coincided with the second moth flight; usually from late in July to late in August with the peak period around August 10. No insecticides were sprayed on the field during and after the experiment. We also placed the traps baited with virgin females in the tested fields to monitor the disruptive effect of Z-5-HD. The effect of Z-5-HD was evaluated early in September from the degree of the larval injuries in rice stubbles immediately after the harvest.

Measurement of the release rate of the test compounds from the polyethylene capsule, rubber septum and the synthetic film. The release rates of the compounds from the polyethylene capsule, rubber septum and the synthetic film were estimated by direct weighing using the micro balance in the laboratory.

RESULTS

Attraction disruption by air permeation with each compound released from polyethylene capsules

The results are shown in Table 1. In test A, relatively good inhibition of male attraction could be achieved with most of the compounds tested when dispensed in the same air currents that were carrying natural pheromone. The pheromone components, Z-5-HD and Z-11-hexadecenol (Z-11-HDOL) almost completely disrupted male attraction. E-isomers of the pheromone were also highly effective as the attraction disruptants. In test B, in which the dispensers were placed around the traps more sparsely than in test A, considerable inhibitory effect was also obtained by most of the compounds. However, Z-11-hexadecenyl acetate (Z-11-HDA) and Z-13-octadecenyl acetate (Z-13-ODA) which were very effective when set in the traps with virgin females in our previous test had little or no effect when these compounds were set around the traps.
Table 1. Effect of the Sex Pheromone Components and Their Related Compounds on Male Attraction to the Virgin Female Traps

<table>
<thead>
<tr>
<th>No.</th>
<th>Compound</th>
<th>Test A</th>
<th></th>
<th>Test B</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>No. of</td>
<td>Percentage of</td>
<td>No. of</td>
<td>Percentage of</td>
</tr>
<tr>
<td></td>
<td></td>
<td>males</td>
<td>disruption&lt;sup&gt;a&lt;/sup&gt;</td>
<td>males</td>
<td>disruption&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>1.</td>
<td>Z-11-hexadecenal</td>
<td>1</td>
<td>99.3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12</td>
<td>90.8&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>2.</td>
<td>Z-13-octadecenal</td>
<td>3</td>
<td>98.0&lt;sup&gt;a&lt;/sup&gt;</td>
<td>35</td>
<td>73.3&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>3.</td>
<td>E-11-hexadecenal</td>
<td>17</td>
<td>88.9&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>45</td>
<td>65.6&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>4.</td>
<td>E-13-octadecenal</td>
<td>15</td>
<td>90.2&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>15</td>
<td>88.5&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>5.</td>
<td>Z-5-hexadecene</td>
<td>1</td>
<td>99.3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>8</td>
<td>93.9&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>6.</td>
<td>Z-5-octadecene</td>
<td>50</td>
<td>67.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>39</td>
<td>70.2&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>7.</td>
<td>Z-11-hexadecenol</td>
<td>1</td>
<td>99.3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>28</td>
<td>78.6&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>8.</td>
<td>Z-13-octadecenol</td>
<td>42</td>
<td>72.5&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>37</td>
<td>71.8&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>9.</td>
<td>Z-11-hexadecenyl acetate</td>
<td>71</td>
<td>53.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>44</td>
<td>66.4&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>10.</td>
<td>Z-13-octadecenyl acetate</td>
<td>148</td>
<td>3.3&lt;sup&gt;d&lt;/sup&gt;</td>
<td>91</td>
<td>30.5&lt;sup&gt;bc&lt;/sup&gt;</td>
</tr>
<tr>
<td>11.</td>
<td>Control</td>
<td>153</td>
<td>—&lt;sup&gt;d&lt;/sup&gt;</td>
<td>131</td>
<td>—&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup> Total of two replicates for 5 nights.

<sup>b</sup> Percentage followed by the same letter within a column was not significantly different at the 5% level with Duncan’s multiple range test from original data.

Table 2. Effect of the Sex Pheromone Components and the Hydrocarbons on Mating Suppression of Tethered Females

<table>
<thead>
<tr>
<th>Compound</th>
<th>No. of females collected&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Percentage of mating&lt;sup&gt;b&lt;/sup&gt;</th>
<th>Percentage of mating suppression</th>
</tr>
</thead>
<tbody>
<tr>
<td>Z-11-hexadecenal</td>
<td>212</td>
<td>8.0&lt;sup&gt;a&lt;/sup&gt;</td>
<td>86.0</td>
</tr>
<tr>
<td>Z-13-octadecenal</td>
<td>209</td>
<td>26.3&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>54.1</td>
</tr>
<tr>
<td>Z-5-hexadecene</td>
<td>223</td>
<td>17.9&lt;sup&gt;b&lt;/sup&gt;</td>
<td>68.8</td>
</tr>
<tr>
<td>Z-5-octadecene</td>
<td>221</td>
<td>31.7&lt;sup&gt;c&lt;/sup&gt;</td>
<td>44.7</td>
</tr>
<tr>
<td>Control</td>
<td>218</td>
<td>57.3&lt;sup&gt;d&lt;/sup&gt;</td>
<td>—</td>
</tr>
</tbody>
</table>

<sup>a</sup> Total of two replicates for 8 nights.

<sup>b</sup> Percentage followed by the same letter was not significantly different at the 5% level with Duncan’s multiple range test from original data.

Mating suppression by air permeation with the sex pheromone components and the hydrocarbons released from rubber septa

To see if mating suppression could occur simultaneously with the inhibition of male attraction under conditions similar to those used in attraction disruption test, the pheromone components and the hydrocarbons, were tested. The effect of each compound on the successful mating is shown in Table 2. All of the four compounds tested significantly suppressed the mating, especially Z-11-HDAL and Z-5-HD had a highly suppressive effect on mating. Among them each C-16 compound was more effective than the corresponding C-18 compound in this case as well. This difference in the effectiveness might also reflect the difference in release rate. It was also shown that the release rate of each hydrocarbon was several times greater than that of the corresponding aldehyde as shown in Fig. 2.

Evaluation of disruptive activity of Z-11-HDAL and Z-5-HD by comparative field test

The disruptive activities of Z-11-HDAL and Z-5-HD were compared in a field
Fig. 2. Release rates of the compounds from the polyethylene capsule.

Fig. 3. Comparison of inhibitory activities between Z-11-hexadecenal and Z-5-hexadecene against male attraction to the virgin female traps.

test. Fig. 3. represents the result of this test. The disruptive effects of Z-11-HDAL and Z-5-HD on male attraction were affected by the amounts of compound in the rubber septum. The plot of 10 mg treatment of the Z-11-HDAL per septum disrupted perfectly. Also 10 mg plot of Z-5-HD exhibited high effectiveness on male attraction. However, these effects decreased gradually with decreasing amounts of the compound in the septum. It was also observed that the disruptive effect of Z-11-HDAL was ap-
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Table 3. Effect of Z-5-hexadecene on Injury in the Rice Plants by Larvae of Chilo suppressalis

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Block</th>
<th>Degree of injuries in stubbles&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Percentage of injured stubbles&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Z-5-hexadecene</td>
<td>I</td>
<td>14.2 a</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>12.2 a</td>
</tr>
<tr>
<td>Untreated</td>
<td>III</td>
<td>32.4 c</td>
</tr>
<tr>
<td></td>
<td>IV</td>
<td>25.8 b</td>
</tr>
<tr>
<td></td>
<td>V</td>
<td>25.2 b</td>
</tr>
<tr>
<td></td>
<td>VI</td>
<td>31.6 bc</td>
</tr>
</tbody>
</table>

<sup>a</sup> For evaluation, 500 stubbles were randomly sampled from each block.

<sup>b</sup> Percentages in each column followed by the same letter are not significantly different (P=0.05, \( \chi^2 \) test).

Parently higher than that of Z-5-HD although the release rate of Z-11-HD AL was lower than Z-5-HD, as shown in other tests described before.

Large scale communication disruption test with Z-5-HD

The practical usefulness of Z-5-HD as a disruptant was demonstrated through the results of several tests as described previously. Therefore, we attempted a large scale disruption test to see whether the crop damage by the larvae of the next generation could be decreased with Z-5-HD. The result is shown in Table 3. The data showed significantly less injuries to the plants in both of the Z-5-HD treated areas than those in all the control areas. This result suggested that the mating frequency in the treated areas was decreased by the treatment with this compound, despite using relatively small treatment areas. We also intended to monitor the disruptive effect by female baited traps during the test period, but we had to terminate the test because of extremely low male catches even in the control traps. Total amount of Z-5-HD that evaporated in the test areas during the test period was estimated at approximately 35 g (87.5 g/ha).

DISCUSSION

Recent research on insect sex pheromones has demonstrated that the sex pheromones in many insect species are composed of multiple components. In such cases, air permeation with an individual component can often disrupt the sexual communication between male and female. YUSHIMA et al. (1975) reported that the individual pheromone components in the armyworm, Spodoptera litura Fabricius, strongly inhibited the orientation behaviour of the male moth to the virgin female and that the minor component of the sex pheromone, Z-9, E-12-tetradecadienyl acetate, was more potent than the major component, Z-9, E-11-tetradecadienyl acetate. TAMAKI et al. (1975) also observed that male attraction was greatly repressed by the two individual components of the sex pheromone in the smaller tea tortrix (Adoxophyes sp.). In this case, however, the repressive effect of the minor component, Z-11-tetradecenyl acetate, was lower than that of the major one, Z-9-tetradecenyl acetate.

The effectiveness of related compounds of the sex pheromones as disruptants
has also been investigated on several insect species. In the pink bollworm, *Pectinophora gossypiella* Saunders, for example, McLaughlin et al. (1972) noted that hexalure, Z-7-hexadecenyl acetate, which resembles the sex pheromone, Z-7, Z-11 and Z-7, E-11-hexadecadienyl acetate, inhibited male attraction. Furthermore, Shorey et al. (1974) confirmed that the damage in cotton fields was decreased about 80 percent by application of a large quantity of hexalure.

Our present experiments also clarified that the sex pheromone and many pheromone-like compounds disrupt male attraction in *Chilo suppressalis*. The results of our experiments emphasize that all the test compounds having the same carbon chain length (C-16 and C-18) with a Z-olefinic linkage at the same position from the methyl terminal as the pheromones (see Fig. 1), showed potently disruptive effect. Among these compounds, Z-11-HDAL of the major pheromone component and Z-5-HD were very effective on male attraction and mating, and also clarified that the disruptive effect of Z-11-HDAL was apparently higher than Z-5-HD. Nevertheless, the usefulness of Z-5-HD was rather supported in this test for the following reason; Z-5-HD was shown to be ca. 10 to 30 times less effective with respect to the amounts treated than Z-11-HDAL and such extent of direct might be overcome by several advantages of Z-5-HD, such as chemical stability and simplicity in the synthesis.

For disruption of sex pheromone communication in *Chilo suppressalis*, other pheromone-related compounds, Z-9-tetradecenyl formate and Z-11-hexadecenyl formate, have been investigated as promising disruptants (Beevor et al., 1977; Beevor and Campion, 1979). Experiments would be conducted to determine how Z-5-HD have disruptive activity compared with that of the formates.

Moreover, it is interesting that the hydrocarbons like Z-5-HD will be applied to the sex pheromone communications of other species that utilize aldehydes as the pheromone components, such as *Heliothis spp.*, *Plutella xylostella*, etc.

Disruptive effects of mixtures of the pheromonal components or of the other related C_{16} and C_{18} compounds on male attraction and mating are also of interest. This is being examined in our laboratory.

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REFERENCES


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