Effect of Neem Oil on Survival of *Nilaparvata lugens* (Homoptera: Delphacidae) and on Grassy Stunt and Ragged Stunt Virus Transmission

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**ABSTRACT** Neem (*Azadirachta indica* A. Juss) seed oil was found highly effective in reducing the survival of a planthopper, *Nilaparvata lugens* (Stål), and in suppressing transmission of grassy stunt and ragged stunt viral diseases of rice. Generally, insect survival and disease transmission decreased with increasing neem oil concentrations. After 3 days of exposure, the insect failed to transmit the viruses to plants sprayed with 50% neem oil, compared with control plants where virus transmission was successful.

*Insect damage* to plants results from direct feeding or from indirect transmission of pathogens during feeding. Therefore, antifeedants, which reduce or disturb feeding activities of insects by rendering plants unattractive or unpalatable, offer a novel approach in pest and vector management. Centuries before synthetic insecticides became available, farmers in India protected crops with natural repellents derived from the fruits and leaves of the neem tree, *Azadirachta indica* A. Juss (Saxena 1983). Mariappan and Saxena (1983, 1984) reported that neem seed oil adversely affected the survival of a leafhopper, *Nephotettix virescens* (Distant), and its transmission of semipersistent rice tungro virus. Laboratory and field trials showed that neem oil has potential as an antifeedant for the brown planthopper, *Nilaparvata lugens* (Stål), a serious rice pest and vector of persistent grassy stunt virus (GSV) and ragged stunt virus (RSV) (Saxena et al. 1981, 1984, Heyde et al. 1984). The incidence of RSV was reported to have been reduced significantly in rice fields sprayed periodically with 12% crude neem oil (Saxena et al. 1981). We tested the effect of different concentrations of neem oil on the survival of *N. lugens* and on its ability to transmit GSV and RSV diseases in neem oil-sprayed rice seedlings.

**Materials and Methods**

The crude oil expelled from decorticated neem seeds (1983 crop) was obtained from C. M. Ketkar, Advisor to the Neem cake Manurial Project, Poona, India. The oil was emulsified in water with 1.6% "Teepol" liquid detergent and tested at 3, 6, 12, 25, and 50% concentrations, as described by Mariappan and Saxena (1983). Hopper-susceptible Taichung Native 1 (TN1) rice seedlings (10 days old) were sprayed separately with various oil dilutions at a rate of 0.5 ml per 40 seedlings, using a quick spray atomizer (Pierce Chemical Co., Rockford, Ill. 61105), 3 h before exposure to the insect. Plants sprayed with 1.6% "Teepol" served as control. Each treated seedling was placed in a glass test tube (15 by 1.5 cm) covered with a polyvinyl cap and the tubes were arranged in racks by treatment. Second- and third-instar nymphs of *N. lugens* reared on virus-free 45-day-old TN1 plants in the insectary were transferred to GSV strain 2 (GSV 2) or RSV virus source plants to become viruliferous adults. Survival and disease transmission studies were conducted separately for grassy stunt and ragged stunt viruliferous *N. lugens*.

One newly emerged, viruliferous female was released into each test tube for inoculation access feeding. After 24 h, each surviving viruliferous insect was transferred to another freshly treated seedling while inoculated seedlings from each treatment were transplanted, for disease development, in an insect-proof screenhouse, in separate plots (50 by 50 cm) for each replication, arranged in a randomized complete block design. Successive inoculation access feeding of the survivors on treated plants was continued for 4 days. GSV 2 symptoms in inoculated seedlings were observed twice—after 12 days and after 3 weeks, RSV disease symptoms were observed after 30 days.

The experiment for each disease was replicated four times, using 40 viruliferous insects and 40 treated seedlings for each replication of 6 treatments. A total of 1,920 viruliferous insects and 4,784 treated seedlings, including those of successive feedings, were used in the two experiments. Data were subjected to analysis of variance and means were compared using Duncan’s (1951) multiple range test at the 5% level.

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Table 1. Survival of *N. lugens* and its ability to transmit GSV 2 on TN1 rice seedlings sprayed with neem oil (Int. Rice Res. Inst., 1984)

<table>
<thead>
<tr>
<th>Neem oil concn (%)</th>
<th>Survival (%)</th>
<th>GSV 2 infection (%)</th>
<th>Survival (%)</th>
<th>GSV 2 infection (%)</th>
<th>Survival (%)</th>
<th>GSV 2 infection (%)</th>
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<tr>
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<td>88a</td>
<td>11b</td>
<td>86ab</td>
<td>7ab</td>
<td>84ab</td>
<td>5ab</td>
</tr>
<tr>
<td>6</td>
<td>88a</td>
<td>9b</td>
<td>78ab</td>
<td>10ab</td>
<td>73bc</td>
<td>4abc</td>
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<tr>
<td>12</td>
<td>90a</td>
<td>7b</td>
<td>69b</td>
<td>3b</td>
<td>60c</td>
<td>1bc</td>
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<tr>
<td>25</td>
<td>82a</td>
<td>7b</td>
<td>70b</td>
<td>2b</td>
<td>9d</td>
<td>0c</td>
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<tr>
<td>50</td>
<td>61b</td>
<td>10b</td>
<td>31c</td>
<td>2b</td>
<td>9d</td>
<td>0c</td>
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<tr>
<td>0 (Control)*</td>
<td>94a</td>
<td>22a</td>
<td>94a</td>
<td>9a</td>
<td>91a</td>
<td>9a</td>
</tr>
</tbody>
</table>

All data are averages of four replications.

* Control treatment was sprayed with 1.6% "Teepol" in water. Within a day, means in a column followed by the same letter are not significantly different (*P* = 0.05; Duncan's [1951] multiple range test).

Fig. 1. Effect of different concentrations of neem oil on the survival of rice grassy stunt viruliferous *N. lugens* females after exposure of (a) 1 day, (b) 2 days, and (c) 3 days, and of ragged stunt viruliferous females after exposure of (d) 2 days and (e) 3 days on neem oil-treated rice seedlings (Int. Rice Res. Inst., 1984).
Transmission of GSV 2 by the viruliferous *N. lugens* generally decreased with increasing neem oil concentrations (Fig. 2). One-day inoculation feeding on control seedlings infected 22% plants, which was significantly more than the percentage of infection at all neem oil concentrations (Table 1). After 2 days of feeding, 19% of the control seedlings were infected; significantly less infection occurred at 12% and higher neem oil concentrations. After 3 days of feeding, significantly less seedlings were infected at 25% neem oil concentration than the control. No infection occurred on 50% oil-sprayed plants. *N. lugens* Survival and RSV Transmission. Survival of *N. lugens* and RSV transmission decreased on neem oil-treated seedlings (Table 2, Fig. 1, 2).

### Table 2. Survival of *N. lugens* and its ability to transmit RSV on TN1 seedlings sprayed with neem oil (Int. Rice Res. Inst., 1984)

<table>
<thead>
<tr>
<th>Neem oil concn (%)</th>
<th>Survival (%)</th>
<th>RSV infection (%)</th>
<th>Survival (%)</th>
<th>RSV infection (%)</th>
<th>Survival (%)</th>
<th>RSV infection (%)</th>
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<td>84a</td>
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<tr>
<td>0 (Control)*</td>
<td>93a</td>
<td>22a</td>
<td>83a</td>
<td>13a</td>
<td>73a</td>
<td>20a</td>
</tr>
</tbody>
</table>

All data are averages of four replications.

* Control treatment was sprayed with 1.6% "Teepol" in water. Within a day, means in a column followed by the same letter are not significantly different (P = 0.05; Duncan's [1951] multiple range test).
One day after feeding, 57% females survived on the seedlings treated with 50% neem oil. This was significantly less than the survival percentage on the control (93%). Insect survival remained high after 2 (83%) and 3 (73%) days of feeding on control seedlings. However, insect survival in treatments involving 50% neem oil after 2 days of feeding and 25 and 50% neem oil after 3 days of feeding was significantly reduced compared with the control. Other treatments of low neem oil concentrations did not differ significantly from the control.

One-day inoculation access feeding infected 22% of the control seedlings; significantly less infection occurred at 12% or higher neem oil concentrations. After 2 days of feeding, 13% of the control seedlings were infected, which was significantly higher than on 25% neem oil-sprayed plants. No infection occurred in seedlings sprayed with 50% neem oil. After 3 days of feeding, significantly less seedlings were infected at all neem oil concentrations compared to control.

Discussion

The complexities of the control of *N. lugens* and of the viral diseases of rice plants it transmits have increased in recent years due to resistance to several insecticides and population resurgence with others in the fields (Heinrichs et al. 1979, Saxena et al. 1981). Planting hopper-resistant rice varieties as an alternative strategy has not been very successful, because of development and occurrence of *N. lugens* biotypes in several rice growing countries (Athwal and Pathak 1972, Mochida 1978, Kalode and Krishna 1979, Pathak and Khush 1979).

Various mineral oils are reported to reduce insect transmission of stylet-borne plant viruses (Bradley et al. 1962, Vanderwen 1968a, Bhargava and Khurana 1969, Dubey and Nene 1974). These mineral oils accumulate on the antical wall of the leaves and act as interfering agents in virus transmission (Simons and Beasley 1977). Although mineral oils protect crops from insect transmission of stylet-borne viruses, they do not inhibit the transmission of persistent viruses (Vanderwen 1968b). A possible reason is that the feeding behavior of the vector is not modified by oil treatment (Vanderwen 1973, Peters and Lebbink 1973).

In a review of the innovative methods of controlling insect-transmitted viral diseases, Simons (1981) recommended the use of oil formulations with naturally occurring antifeedants derived from plants. In our approach, we used neem oil not only as an interfering agent that reduces viral transmission, but also as an insect repellent with antifeedant properties. The insect repellent action of neem has been attributed to azadirachtin, a tetracyclic triterpenoid, and other related novel chemical compounds in neem seed derivatives (Jacobson 1981, Kraus et al. 1981, Morgan 1981). Saxena et al. (1981) reported that the feeding duration of *N. lugens* on neem oil-treated rice plants decreased by 0.93 min per h for every 1% increase in neem oil concentration, while search and avoidance of feeding sites increased correspondingly.

Recently, Heyde et al. (1984) and Saxena et al. (1984) found that the food intake by *N. lugens* was significantly reduced on rice plants sprayed with neem oil. Thus, observed adverse effects of neem oil on the survival of *N. lugens* and reduction in its ability to transmit persistent GSV 2 and RSV can be ascribed to reduced feeding.

Acknowledgment

We thank H. Hibino, virologist (Int. Rice Res. Inst.) for providing us with grassy stunt (strain 2) and ragged stunt virus source plants and Zenaida M. Flores for assistance in recording the number of infected plants.

References Cited


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