Investigating insecticidal and repellent activity of *Momordica charantia*, *Calotropis gigantea* and *Cordia curassavica* against *Oebalus poecilus*

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Abstract

*Oebalus poecilus* (paddy bug) is a destructive rice pest in Guyana that is currently controlled with synthetic chemical pesticides. Increasing resistance rates and farmer's overuse of pesticides have resulted in economic and ecological issues for Guyanese rice farmers. Amid growing economic and ecological concerns, this experiment aimed to investigate the use of alternative botanical extracts from the leaves of *Momordica charantia* (Baan-Carailli), *Calotropis gigantea* (Madar-Flower) and *Cordia curassavica* (Blacksage) to control paddy bug infestation by testing their efficacy as insecticides on *O. poecilus*. A completely randomized experimental design and a hot water extraction method was used to prepare the leaf extracts used in this experiment. Three different concentrations (6%, 12% and 24%) of each extract were tested on the adult stage of the bugs for mortality and repellent activity. The results, after 72 hours, were recorded and revealed that all extracts demonstrated insecticidal properties. No toxicity as a systemic insecticide on contact with bugs was observed, thus implying no repellent effect. At 72 hours, the average mortalities recorded for the different extracts were *Momordica charantia* (98.62%), *Cordia curassavica* (90.25%) and *Calotropis gigantea* (100%). Extracts performed better at 6% and 24% when compared. Leaf extracts from *Calotropis gigantea* at 6% and 24% were most effective when compared to the other extracts used in this experiment.

Keywords: *Oebalus poecilus*; Leaf extracts; Insecticide; Repellant

1. Introduction

*Oryza sativa* (rice) is a staple in the diet of many individuals, and a large percentage of the world’s population depends on rice cultivation for their livelihood [1]. In Guyana, the rice industry is an important agricultural crop that makes a significant contribution to the country's economy.

The paddy bug (*Oebalus poecilus*), a late-season rice pest, is one of the most destructive rice pests currently in Guyana, and the Integrated Pest Management (IPM) system is used to control it since one method of control has proven to be ineffective to completely control this particular pest. With both adults and nymphs feeding on the paddy grains, the damage they wreak has a negative effect on the harvested paddy [2] [3] [4] [5]. Infested panicles lead to wind grains, malformation, and discoloration of grains, resulting in lower yields, reduced quality, and brittleness, which increase the breakage on milling [6].

Currently, various chemicals are used to control this pest in Guyana but significant damage is still experienced. Farmers have complained that even with the intense use of chemicals, the problem still exists and they are seeking new methods of control, especially given the increasing awareness of the ecological and environmental implications of the intense use of chemical pesticides [7] [8].
This research investigated the rate of mortality of *O. poecilus* when treated with hot water leaf extracts from *Momordica charantia* (Baan-Carailli), *Calotropis gigantea* (Madar-Flower) and *Cordia curassavica* (Blacksage) and sought to determine if there was any repellent effect of the extracts on *O. poecilus*.

This study has the potential to add to the body of knowledge that already exists regarding the search for alternative botanical, ecologically and environmentally safe pesticides that may be used to control paddy bugs. The results may also be useful in determining if extracts from the plants used in the experiment have insecticidal properties against the paddy bug.

2. Material and methods

This study was carried out on samples obtained from the Upper Corentyne, East Berbice, Guyana. A Completely Randomized Design (CRD) was utilized for this study [9]. Crude extracts were prepared at the University of Guyana Johns Science Centre where the experiment was conducted.

2.1. Plant and test materials

The present study utilized leaf samples from *Momordica charantia*, *Calotropis gigantea* and *Cordia curassavica* and the test organism was the adult stage of *O. poecilus*. All specimens were collected from Bushlot Village, Upper Corentyne, East Berbice, Guyana. The extract from each plant was prepared and used at three concentrations, 6%, 12% and 24% (Table 1). A control, Treatment (T10) was used in the experiment.

<table>
<thead>
<tr>
<th>Plant</th>
<th>Concentration of extract (%)</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Calotropis gigantea</em> (Madar-Flower)</td>
<td>6 %</td>
<td>T1</td>
</tr>
<tr>
<td></td>
<td>12 %</td>
<td>T2</td>
</tr>
<tr>
<td></td>
<td>24 %</td>
<td>T3</td>
</tr>
<tr>
<td><em>Momordica charantia</em> (Baan-Carailli)</td>
<td>6 %</td>
<td>T4</td>
</tr>
<tr>
<td></td>
<td>12 %</td>
<td>T5</td>
</tr>
<tr>
<td></td>
<td>24 %</td>
<td>T6</td>
</tr>
<tr>
<td><em>Cordia curassavica</em> (Blacksage)</td>
<td>6 %</td>
<td>T7</td>
</tr>
<tr>
<td></td>
<td>12 %</td>
<td>T8</td>
</tr>
<tr>
<td></td>
<td>24 %</td>
<td>T9</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>T10</td>
</tr>
</tbody>
</table>

2.2. Collection of leaf samples

The three test plants were identified by the taxonomist from the Division of Natural Sciences at the Johns Science Centre of the University of Guyana Berbice Campus. The samples were collected at Bushlot Village, Corentyne, using a stratified random sampling method. Mature leaves from ten (10) samples of each plant were randomly collected using a 1 m² quadrat.

2.3. Collection of paddy bugs and milk grains

The milk grains and paddy bugs were collected from the same rice field located at Bushlot Village, Corentyne, East Berbice, Guyana. The adult paddy bugs were collected using a sweep net and were transported in sterile plastic bottles with holes to prevent the bugs from dying from suffocation.

2.4. Leaf sample preparation and extraction

Sample preparation and extraction were carried out following the stipulated guidelines of the World Journal of Science and Research according to the method for phytochemical screening of plants [10]. The leaves from each test plant were air-dried for 2 to 3 days and then the dried leaves were ground into a powder, using an electronic blender. The leaf
powder was weighed into three different quantities: 15g, 30g, and 60g, using an electronic balance to make the 6%, 12% and 24% concentrations. The hot water extraction method was used to prepare the concentrations where, for example, to make the 6% concentration, 15 g of leaf powder was dissolved in a beaker filled with 1 litre of water which was then boiled until the volume decreased to 250 ml. The mixture was then filtered and the filtrate was poured into labelled plastic bottles and stored in the refrigerator until it was used. This process was a modified version that encompassed the general extraction procedures which included maceration, digestion, decoction, infusion and percolation [11] [12] [13].

2.5. Experimental Design

Petri dishes were washed, dried, sterilized and labelled using a permanent marker. Sixteen (16) milk grains were soaked in each concentration of extract for 3-4 minutes then placed in a labelled petri dish and eight (8) healthy bugs were placed in each petri dish. The control group had eight (8) bugs with sixteen (16) untreated milk grains. Each concentration for the experiment was set up in triplicate.

2.6. Observation of parameters of interest regarding mortality and repellant property

The mortality of bugs in each petri dish was recorded and along with the observation of any repellent activity. Results were taken every 30 minutes for three (3) hours, then every twelve (12) hours for three (3) days. Results for repellent activity were collected for four hours; between the hours of 5 AM and 7 AM and between the hours of 4 PM and 6 PM.

2.7. Data analysis

The data obtained from the experiment which was based on a CRD and with each treatment replicated three times, was recorded, analyzed, interpreted and discussed. A comparison with existing literature was done. Data were subjected to the CRD one-way ANOVA using STATISTIX 10 software package. Means were separated using Least Significance Difference (LSD) to determine whether there was a significant difference in the means of each treatment. The results were evaluated at a significance level of α = 5% or 0.05 or 5% probability level.

3. Results

3.1. Mean mortality rate for each treatment

The mean mortality rate of *O. poecilus* for each treatment with time is presented in Table 2.

**Table 2 Mean mortality rate of *O. poecilus* for each treatment with time**

<table>
<thead>
<tr>
<th>Time (Hours)</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
<th>T9</th>
<th>T10</th>
</tr>
</thead>
<tbody>
<tr>
<td>12</td>
<td>1.67</td>
<td>1.33</td>
<td>2.00</td>
<td>3.00</td>
<td>2.00</td>
<td>2.00</td>
<td>3.00</td>
<td>2.33</td>
<td>2.00</td>
<td>0.00</td>
</tr>
<tr>
<td>p-value</td>
<td>0.1894</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>24</td>
<td>4.67</td>
<td>5.00</td>
<td>5.67</td>
<td>7.00</td>
<td>6.33</td>
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<td>6.33</td>
<td>4.67</td>
<td>6.33</td>
<td>1.00</td>
</tr>
<tr>
<td>p-value</td>
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<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>36</td>
<td>6.67</td>
<td>6.67</td>
<td>6.67</td>
<td>7.33</td>
<td>6.67</td>
<td>7.00</td>
<td>7.00</td>
<td>5.33</td>
<td>7.33</td>
<td>1.00</td>
</tr>
<tr>
<td>p-value</td>
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<td></td>
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<td>48</td>
<td>8.00</td>
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<td>7.67</td>
<td>7.67</td>
<td>7.67</td>
<td>8.00</td>
<td>7.33</td>
<td>6.33</td>
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<tr>
<td>p-value</td>
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<td></td>
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<td></td>
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<td></td>
<td></td>
<td></td>
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<td>7.33</td>
<td>6.33</td>
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<td></td>
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<tr>
<td>72</td>
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<td>8.00</td>
<td>8.00</td>
<td>7.67</td>
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<td>7.33</td>
<td>6.33</td>
<td>8.00</td>
<td>1.00</td>
</tr>
<tr>
<td>p-value</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The mean mortality of paddy bugs under each treatment varied with time and treatments. The treatment after 12 hours did not show a significant difference (p-value = 0.1894) in the mean mortality rate. All treatments after 24 hours to 72 hours showed significant differences in mean mortality rates.

Treatments 4 and 7 had the highest mortality within the 12 hours while treatment 10 had no mortality. After 24 hours, the mean death of bugs under all the Treatments ranged from 1 to 7 and there was a significant difference (p-value = 0.0001) in the mean death of bugs among the Treatments. In 24 hours, Treatment 4 had the highest mortality, while treatment 10 had the lowest compared to the other treatments.
At thirty-six hours, there was a significant difference (p-value = 0.000), between the mean mortality of the treatments. This significant difference did not lie between treatment (1, 2, 3, 4, 5, 6, 7, and 9) but it lay between treatment (1 to 9) and treatment 10. There was also a significant difference (p-value = 0.000), between treatment (4, 6, 9) and treatment 8. At 36 hours, treatment 9 and treatment 4 had the highest mortality compared to the other treatments, with treatment 10 having the lowest mortality.

At forty-eight hours, there was a significant difference (p-value = 0.000) in the mean mortality of the treatments. The significant difference lay between treatment (1, 2, 3, 4, 5, 6, 7, 8, 9) and treatment 10 and between treatment (1, 2, 6, 9) and treatment 8. Treatment (1, 2, 3, 4, 6, and 9) had the highest mortality rate at this time compared to the other treatment, with treatment 10 having the lowest mortality.

After sixty hours, there was a significant difference (p-value = 0.000) in the mean mortality of the treatments. The significant difference was seen between treatment (1, 2, 3, 4, 5, 6, 7, 8, 9) and treatment 10 and between treatment (1, 2, 3, 4, 6, 9) and treatment 8. Treatment (1, 2, 3, 4, 6, and 9) had the highest mortality rate in sixty hours compared to the other treatment, with treatment 10 having the lowest mortality.

After seventy-two hours, the p-value was also 0.000, which is indicative of a significant difference in the mean mortality of the treatments. A significant difference was noted between treatment (1, 2, 3, 4, 5, 6, 7, 8, 9) and treatment 10. Treatment (1, 2, 3, 4, 6, and 9) had the highest mortality in seventy-two hours compared to the other treatment, with treatment 10 having the lowest mortality.

### 3.2. Repellent activity for each treatment with time

![Figure 1](image-url) Repellent activity of treatments in the morning

<table>
<thead>
<tr>
<th>Time</th>
<th>T1</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
<th>T9</th>
<th>T10</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 mins</td>
<td>2.67</td>
<td>3.00</td>
<td>2.67</td>
<td>2.67</td>
<td>4.33</td>
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<td>3.67</td>
<td>4.00</td>
<td>3.67</td>
<td>0.7629</td>
</tr>
<tr>
<td>60 mins</td>
<td>2.67</td>
<td>3.33</td>
<td>2.67</td>
<td>2.67</td>
<td>2.33</td>
<td>2.67</td>
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<td>5.33</td>
<td>4.00</td>
<td>2.33</td>
<td>0.7691</td>
</tr>
<tr>
<td>90 mins</td>
<td>2.00</td>
<td>3.00</td>
<td>2.67</td>
<td>2.33</td>
<td>4.67</td>
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<td>2.33</td>
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<td>0.8711</td>
</tr>
<tr>
<td>120 mins</td>
<td>4.33</td>
<td>2.00</td>
<td>1.67</td>
<td>1.33</td>
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<td>2.33</td>
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<td>3.33</td>
<td>2.67</td>
<td>2.00</td>
<td>0.7348</td>
</tr>
<tr>
<td>12 hrs</td>
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<td>1.33</td>
<td>2.00</td>
<td>2.67</td>
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<td>0.5278</td>
</tr>
<tr>
<td>24 hrs</td>
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<td>1.00</td>
<td>0.67</td>
<td>1.33</td>
<td>1.67</td>
<td>1.33</td>
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<td>36 hrs</td>
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<td>1.00</td>
<td>2.00</td>
<td>1.67</td>
<td>2.67</td>
<td>0.5432</td>
</tr>
</tbody>
</table>
Repellent activity for each treatment with time is presented in Table 3, Figure 1 and Figure 2.

The p-values for all the timings are above α (0.05), which indicates that there was no significant difference between the ten treatments in terms of repellent activity, indicative that the extracts had no repellent effect on the paddy bugs.

4. Discussion

In this research, leaf extracts from *C. curassavica* (Blacksage), *M. charantia* (Baan-Carailli), and *C. gigantea* (Madar-Flower) were used to test mortality and repellent activity against *O. poecilus* (Paddy bug).

The use of these plants was based on reported information that they contained bioactive compounds [14] [15] [16] [17] that act as natural antifeedants and insecticides against grazing animals and insects that tend to attack plants [17]. [18] reported the presence of α-pinene, sabinene, (E)-caryophyllene, ar-curcumene, β-sesquiphellandrene, 7-cyclodecen-1-one, and ar-Turmerone as major compounds in the essential oil of *C. curassavica*. [16] reported the presence of bioactive compounds, alkaloids, phenols, tannins, flavonoids, phytosterols, terpenoids, and saponins in *C. gigantea* leaves. Also, [15] reported the presence of saponins, peptides, phenolics, flavonoids, triterpenes, carotenoids, alkaloids, and amino acids in *M. charantia*. Based on the results of research conducted by [14] [15] [18] [17] it is presumed that the toxicity against *O. poecilus* was due to the presence of these reported bioactive compounds which are known to have natural insecticidal properties.

According to [17], saponins are known to act as both antifeedants and insecticides against different life stages of insect pests, thus playing an important role in the natural defence mechanisms of some plants. The presence of saponins in both *C. gigantea* and *M. charantia* may have also played a role in the observed mortality of *O. poecilus* in this research.

The results obtained from the experiment (Table 2) seem to indicate that the botanical extracts of the Madar-Flower, Baan-Carailli and Black sage plant cannot be used as a contact insecticide. This may be deduced from the fact that based on the p-values, there was no significant difference in the mortality of *O. poecilus* between treatments (T1 to T9) and Treatment 10 (the control). If the extracts had the potential to be used as a contact pesticide, there might have been a significant difference in the mortality rate between treatments (T1 to T9) and Treatment 10 at 12 hours.

Based on the p-value (0.0001) for 24 hours, there is a significant difference in the mean mortality among the treatments. Among the other treatments, T4 (Baan-Carailli 6%) had the highest mortality; which may be indicative of the fact that it probably functioned better as an insecticide than the other concentrations of extracts. However, while T4 was the
quickest, treatments (T1, T2 and T3) which are the concentrations of Madar-Flower extracts, had the lowest mortality, indicating that these extracts performed slower than the other leaf extracts.

The results shown in (Table 2) reveal that extracts are effective from 24 to 48 hours after application. This is so because the mean mortalities for extracts at 48 hours are very close to the mean mortalities at 60 hours, and the mean mortalities at 60 hours are identical to the mean mortalities at 72 hours.

Although all of the plant extracts used were effective in terms of the mortality of the paddy bug, it was expected that at higher concentrations, the mean mortality will be higher compared to lower concentrations. This is so because higher concentrations ought to have a higher amount of the bioactive compounds than the lower concentrations.

In terms of mortality, there were fluctuations among concentrations used for all three extracts. At 72 hours, although all three concentrations of Madar-Flower had 100% mortality, the 6% and 12% concentrations performed faster than the 24%. For the Baan-Carailli extracts, the 6% and 24% concentrations performed better than the 12%. For the black sage extracts, although the 24% concentration performed better than the 6% and 12% concentrations, the 6% concentration performed better than the 12% concentration.

The fluctuations observed at 72 hours for the mortality of *O. poecilus* may be accounted for as a result of synergistic and antagonistic associations between the bioactive compounds present in each concentration. Whenever a compound exhibits a synergistic effect with another compound, they work together to produce a combined effect, which is greater than the sum of their individual effects. However, when a compound exhibits an antagonistic effect with another compound, they tend to be hostile to each other and one inhibits the other one from functioning as it should function [19].

It may also be assumed that since the 6% concentrations of the extracts performed better than the 12% extract, there may be more synergistic action at 6% than antagonistic actions. Regarding the extract from the Madar-Flower, the 24% concentration performed slower, and it may be assumed that there may greater antagonistic than synergistic action of the bioactive compounds that are present.

Regarding repellent activity, the results revealed that the botanical extracts may not be useful as a repellent against *O. poecilus*. As seen in Table 3, Figure 1 and Figure 2, there was no repellent activity recorded and no significant difference between the ten treatments.

5. Conclusion

From the study conducted, several conclusions may be drawn.

- The extracts from all three plants appear to possess insecticidal properties against paddy bug *O. poecilus*. However, these extracts did not appear to function as a contact insecticide.
- None of the extracts appeared to have repellent properties nor impact the feeding time of the paddy bug.
- Although extracts from the Madar-Flower appeared to perform the slowest, they appeared to be most effective.
- The 6% and 24% performed better than the 12% in most of the extracts, with the 12% extracts showing the lowest mortality and performing the slowest for two plant extracts, the Baan-Carailli and Black sage extracts.
- More research should be done to determine the active ingredients in the extracts and the effective concentrations that could be used by farmers to address the concerns of the ecosystem and environmental impacts of chemical pesticides.
- Given that all three of the test plants are abundant locally in Guyana and the method of preparation is simple, it will be useful to conduct other studies of this nature to fully explore the potential use of these plants in controlling the paddy bug.

Compliance with ethical standards

Acknowledgments

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Disclosure of conflict of interest
The authors declare there are no conflicts of interest.

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