EFFICACY OF DIFFERENT PARTS OF CLOVE (*Syzygium aromaticum*) FOR THE MANAGEMENT OF RICE WEEVIL (*Sitophilus oryzae* L.)

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INTRODUCTION

The staple food of Sri Lankans; rice, subjected to 4-6% of loss during storage due to insect infestations. The rice weevil, (*Sitophilus oryzae* L.) is one of the most destructive primary pests among them (Kumari *et al,* 2020). Primarily, these pests are controlled by synthetic insecticides such as phosphine and pirimiphos-methyl. Though the use of such synthetic insecticides is efficient, it influenced negatively on the eco-system due to long half-life and human health risks, etc. (Chowański *et al*, 2014). Therefore, there is an urgent need of developing alternatives along with the emerging trend in the world.

Among many locally available botanicals which have insecticidal properties, there is very little research on insecticidal potential on clove (*Syzygium aromaticum*). Furthermore, there is no evidence of the insecticidal efficacy of clove stems and leaves. Most importantly, the clove stems are underutilized, low-cost plant materials in Sri Lanka and the clove leaves are a waste during the harvesting season. Therefore, the main objective of this study was to evaluate the repellent activity and efficacy of extracts (Ethanolic, Hexane and Ethyl acetate) and powders of clove bud, stem and leaf against the rice weevil.

METHODOLOGY

Plant material collection and preparation of extracts

Clove (*Syzygium aromaticum*) buds (budding stage 3) (Alfikri *et al*, 2020) and stems (harvested with buds of budding stage 3/ dark green), leaves (matured/maximum chlorophyll content/dark green color), were collected from Kandy area. Then the plant parts were air dried for about 5 days and crushed into a fine powder using an electrical blender. Powders were packed in airtight containers until they were used.

Three extracts; ethanolic, hexane and ethyl acetate were prepared from each plant part separately. A 30.0 g of fine powder in 300 ml of each solvent was stirred using a magnetic stirrer for 30 min and filtered after standing for 24 hours using a Whatman No.1 filter paper (Kamruzzaman & Shahjahan, 2005). This extract was preserved in glass bottles under refrigerated condition (4°C) until they were used. The bioassay was performed using three concentrations 25%, 50% and 75% of each extract.

Rearing of insect culture

Rice weevils (*Sitophilus oryzae)* were collected from infested rice grains in a local store. They were reared on un-infested rice grains in a glass jar under 28 ± 2°C condition. Adult weevils of 10-14 days old were used for the bioassays.

Contact repellent bioassay for extracts

Repellent bioassay was performed using the filter paper impregnation method (Jahan *et al*, 2019). Whatman No.1 filter paper was cut into two halves and 1 ml of each concentration of each extract was applied to one half while the other half (control) was treated with the respective solvent. Then the filter paper halves were air dried, for few minutes and attached together. It was placed in a glass petri plate (90 mm) and 10 weevils were released at the centre. The covered petri plates were kept in dark and the number of weevils settled on each half after 24 hours were counted. Then the data were expressed as percentage repellency (PR) using the below formulae (Gitahi *et al,* 2021). Each test was performed with three replicates.

Percentage repellency (PR)= $\frac{(Nc - Nt)}{(Nc + Nt)} \times 100$

Nc = No. of insects settled in the control half   *Nt* = No. of insects settled in the treated half

Contact and fumigation repellent bioassay for plant powders

Contact and fumigation repellent bioassay for powder was performed using modified cup bioassay method (Perera & Karunaratne, 2015), with slight modifications using 200, 400 and 600 mg of each plant powder mixed with 30.0 g of rice in a plastic container. A control was made without adding powder. These containers were perforated at ¼ of the height from the top allowing weevils to escape due to any repellent effect. Then, 20 adult weevils were added to the container and it was placed in a large container to trap escaped weevils.
A similar apparatus was used as the above test with some modifications for the fumigation test. The bottom of a similar perforated plastic bottle as above was removed and replaced with a nylon cloth. It was attached to another plastic container at the bottom to add powder. After adding powder, 30.0 g of rice was placed on the top container. Then, 20 adult weevils were added and the container was closed using a muslin cloth. The whole apparatus was kept in a large container to trap the escaped weevils. Then the escaped weevils were counted and the repellent percentage was calculated.

Repellent classes based on percent repellent effect (Das *et al.*, 2015)

Class 0 = 0%–0.1%
Class I = 0.1%–20%,
Class II = 20.1%–40%,
Class III = 40.1%–60%,
Class IV = 60.1%–80%,
Class V = 80.1%–100%.

Statistical analysis
All the data were analyzed by General Linear Model and means were compared by Tukey's multiple comparison test using Minitab 19 software.

RESULTS AND DISCUSSION

As there were no previous studies regarding the repellent effect of clove bud, stem and leaf powder and their ethanolic, hexane and ethyl acetate extract against the rice weevil, this study clearly showed the efficacy and potential of using these natural botanicals to control rice weevil.

The contact repellent activity of bud, stem and leaf at different concentrations (200,400,600 mg) at 1,2,3,6,12,24 hours after treatment were statistically significant ($p<0.05$). The highest repellent activity (93.33±1.67) was found in bud powder at 400 and 600 mg concentrations after 24 hours of treatment (Figure 01). A 600 mg of stem powder showed the second highest repellent activity (81.67±1.67) after 24 hours and the least repellent effect was found in the leaf powder. Further, bud powder showed more than 50% of repellent activity even after 2 hours of treatment while stem powder had the same effect after 6 hours of treatment. However, the leaf powder showed the lowest activity. According to the classification, bud powder (class IV) and stem powder (class III) were in highest repellent classes.

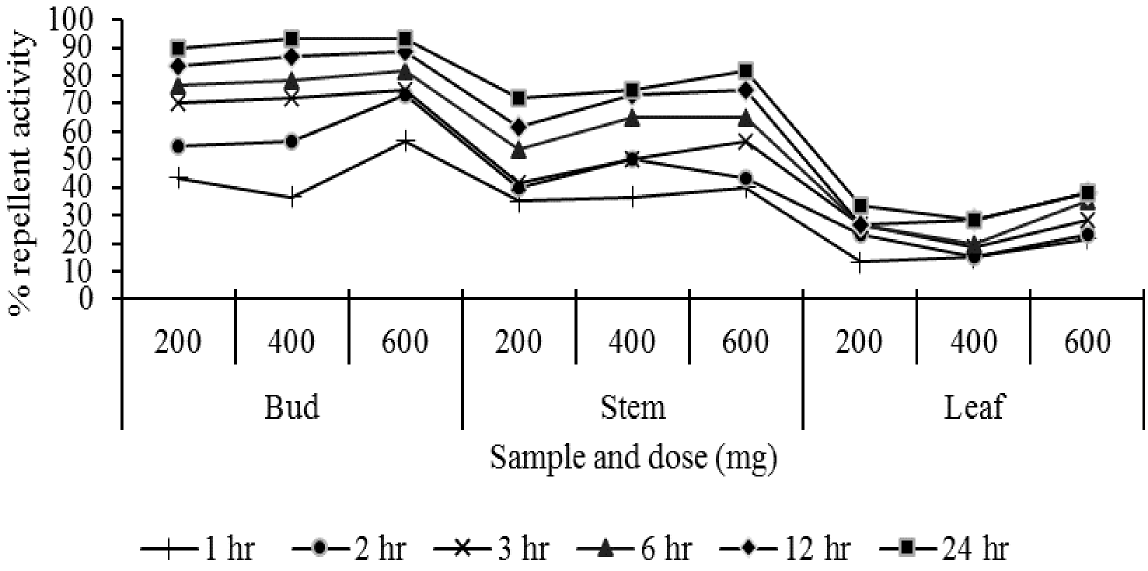

Figure 01: Contact repellent activity of powders of different clove parts

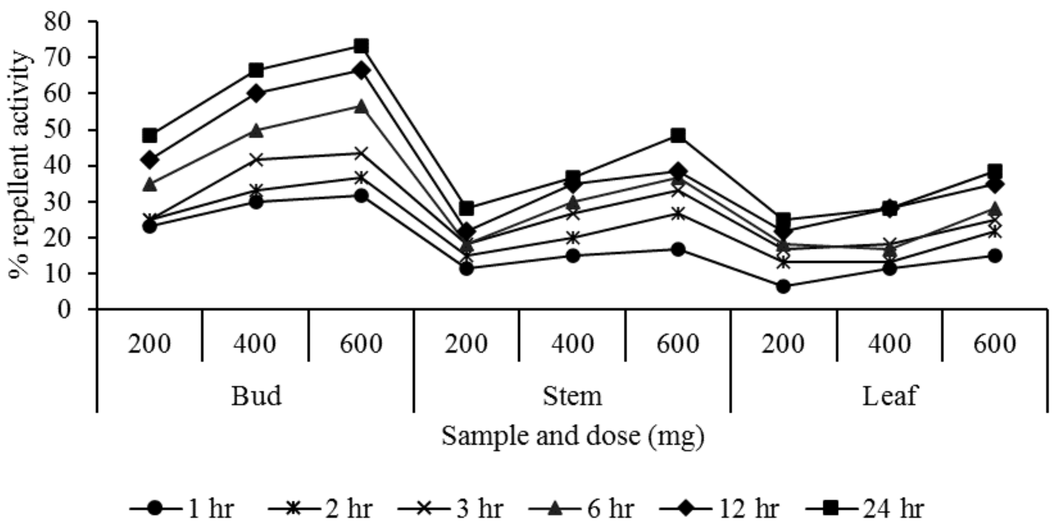

Figure 02: Fumigant repellent activity of powders of different clove parts

Similarly, the bud powder at 600 mg concentration showed the highest fumigation repellent effect (73.33±1.67) after 24 hours of treatment (Figure 2). The bud was classified under the repellent class III while stem powder and leaf powder were classified under class II and I respectively showing lower fumigation repellent activity.

There was a significant difference among samples, extracts and their concentrations (25,50,75%) for repellent activity of rice weevil after 24 hours of treatment (Figure 3). Moreover, there was a significant difference between bud (58.39±4.15) and other two plant part extracts, but there was no significant difference between stem (41.36±2.84) and leaves (33.58±2.13).

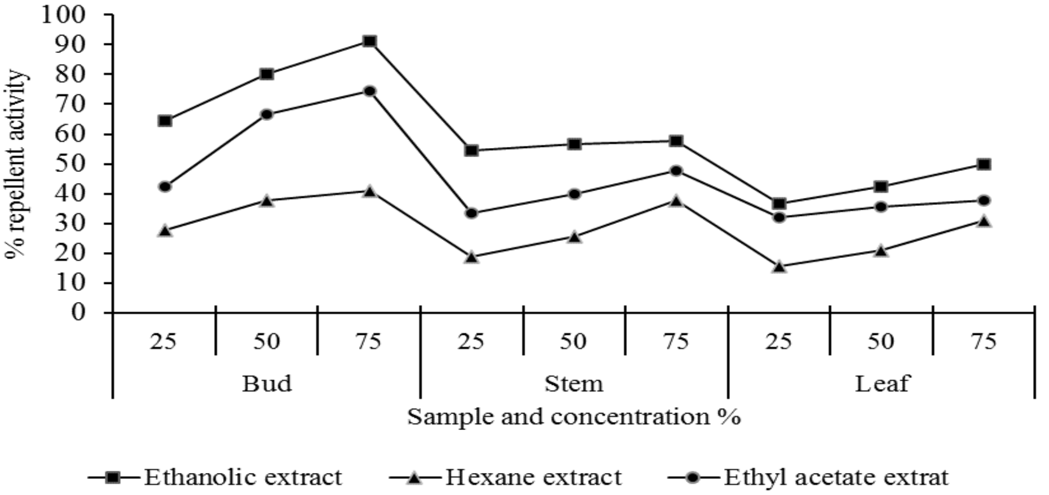

Figure 03: Contact repellent activity of different extract of clove parts after 24 hours of treatment

Among three extracts, the 75% ethanolic extract of bud showed the highest repellent activity (91.11±2.22), even the lowest concentration (25%) showed more than 50% of repellent effect (64.44±4.84). The repellent effects of stem and leaf extracts were not significantly different ($p>0.05$). The hexane extract of all the plant materials showed the lowest repellent effect while ethyl acetate extracts reported a medium effect. This may be attributed to the high amount of eugenol, methyl eugenol and iso-eugenol in ethanolic extract and natural aromatic compounds present in clove bud, stem and leaves (Abo-El-Saad *et al*, 2011).

Few studies have shown that the clove essential oil and bud itself can be used for the control of insect pests (Saad *et al*, 2017; Jairoce *et al.*, 2016). However, this study provided strong evidence on the repellent properties of clove stems and leaves as well. The clove stem reported the highest repellent effect followed by clove bud in all the bioassays demonstrating the potential of using effective raw materials for the control of rice weevil.

CONCLUSION

Both the clove bud and stem possess great efficacy in contact and fumigation repellency in powder and extract formulations. There was a significant difference between the repellency of bud and other two plant parts, but there was no significant difference between the repellency of stem and leaf. Considering the repellent effect and higher commercial value of clove buds, underutilized clove stems could be recommended as effective, eco-friendly and safe alternative for successful control of *S. oryzae.* Further studies are needed to identify the active agents and mode of action of the compounds available in the different parts for the commercialization purpose.

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