BIOEFFICACY OF SHIYALMUTRA LEAF EXTRACT AGAINST RICE WEEVIL

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ABSTRACT

Pest management in the field and storage condition by indigenous plant materials is becoming popular because of their non-toxic effect on environment. In this study aqueous leaf extract of shiyalmutra, *Blumea lacera* were assessed for its toxicity, repellency and residual effects against the stored grain pest, rice weevil (*Sitophillus oryzae*). This study results showed that the highest (63.25%) and lowest (21.23%) mortality rates of weevils were occurred by 5% and 1% extracts, respectively. Shiyalmutra leaf extracts at 5% concentration had highest repellency rate (63.3%) of the weevils at 3 hours after treatment. The thin layer chromatography (TLC) examination of aqueous extract showed three distinct compounds at Hexane: Ethyl acetate (10:1, v/v). These results indicated the value of shiyalmutra plant as a means of protection of rice in storage condition.

Key words: Aqueous extracts, chemical investigation, insecticidal effect, rice weevil

INTRODUCTION

Insect infestation in stored grains and other stored products is a world wide problem. There are about 200 insects and mites which attack stored grains and stored products (Maniruzzaman, 1981). Losses due to insect infestation are the most serious problem in grain storage, particularly in the cases of developing countries like Bangladesh. It is reported that about 5-8% of the food grains, seeds and different stored products are lost annually due to storage pests, and if the losses incurred on farms were included, it would amount to 10%. Insect pests also cause considerable losses to stored rice. The rice weevil is one of the most important pests of cereals. It causes loss to grain in storage, either directly through consumption of the grain or indirectly by producing 'hot spots' causing loss of moisture and thereby making grain more suitable for their consumption. Besides rice, the insect also infests maize, wheat and sorghum, oats, barely, cotton seed, linseed and cocoa.

Many preventive and effective control measures have been reported to minimize the loss of stored grains due to insect attack. Among these, many investigators suggested chemical insecticides for their control effectively. Synthetic or chemical pesticides which have been used for a long time for controlling insect pests have got many limitations and undesirable side effects (Fishwick, 1988). Increasing problems have dictated the need for effective, biodegradable pesticides with greater selectivity. This awareness has created a worldwide interest in the development of alternative strategies including the search for new types of insecticides and use of age-old traditional botanical pest control agents. Plants are a rich source of compounds having insecticidal activity such as potential of the neem tree (*Azadirachta indica*) for pest control and rural development (Ahmed and Grainger, 1986). Different types of naturally occurring bioorganic compounds have been isolated from plants. So, the present study was undertaken to assess the pesticide effect of shiyalmutra plant extract against rice weevil.

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MATERIALS AND METHODS

The present study was conducted in the Laboratory of the Department of Agricultural Chemistry, Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur.

Preparation of plant extract: Fresh leaves of shiyalmutra, collected from the campus of HSTU, were washed in running water and then dried in shade and finally dried in oven at 50-60 °C for 24 hours to gain constant weight. Hundred grams of leaf powder was then mixed with 1000 ml distilled water and left for 72 hours with an interval of shaking. After 72 hours it was then filtered. The filtrate was considered as 10% solution from which 1, 3 and 5% solution were prepared for insect bioassays.

Insect Bioassays: Insect bioassays such as direct toxicity, repellency, feeding deterrence and residual toxicity were observed in the laboratory under ambient conditions. Three to seven days old rice weevils were used for insect bioassey. The trial insects were supplied by the Department of Entomology, HSTU, Dinajpur.

Direct toxicity test: The direct toxicity test was conducted according to the method described by Talukder and Howse (1993). One μ L. solution of different concentrations (1, 3 and 5%) was applied to the dorsal surface of the thorax of each insect using a micro capillary tube. Before applying, insects were chilled to immobilize for a period of 10 minutes. Ten insects per replication were treated and each treatment was replicated 3 times. In the control same number of insects was treated with solvent (distilled water) only. After treatment, the insects were transferred into 9 cm diameter Petri-dishes (10 insects / Petri -dish) containing food. Insect mortalities were recorded at 24, 48 and 72 hours after treatment (HAT). Observed mortalities of the insects were corrected by Abbott's Formula (Abbott, 1987) and then analyzed by Analysis of Variance (ANOVA). The mean values were separated by DMRT.

Repellency test: Repellency test was conducted according to the method of Talukdar and Howse (1994). Nine cm diameter filter paper (Whatmann No. 40) was marked into two portions. With the help of a pipette 1 ml solution of each concentration of plant extract was applied to one half of the filter paper. The treated half disk was then air dried. Each filter paper was then placed in a Petri-dish and 10 insects were placed there. There were 3 replications for each concentration of plant extracts. Numbers of insects on each portion were counted at hourly intervals up to the 5th hour. The data were expressed as percentage repulsion (PR) by using the formula: PR (%) = (Nc-50)×2. Where, Nc = the percentage of insects present in the control half. Data (PR) were analyzed using analysis of variance (ANOVA) after arcsine percentage transformation.

Residual toxicity test: Residual toxicity effect was evaluated by observing mortality of insects released in the treated rice grain at 3 intervals i.e. **7**, 14 and 21days after treatment (DAT). The experiment was conducted according to the method of Mishra *et al.* (1992) with some modifications. Rice seed (rearing media) was treated with different concentration of the extract and about 10 g of treated foods were then taken in each Petri-dish. For control, the foods were treated with solvent only. Three replications were made for each dose and also for control. Five pairs of newly emerged adult beetles from laboratory culture were released in each Petri-dish at 7, 14 and 21 DAT. The mortality of insects was recorded at 3 days after releasing of insects.

Isolation of crude compound from plant extract: An attempt was made to isolate the crude compound(s) from the treated plant extract by using chloroform and ethyl alcohol. For this purpose 100 g powder of shiyalmutra leaves was mixed with 250 ml chloroform / 200 ml ethyl alcohol separately. It was then kept for 72 hours with regular interval of shaking. After 72 hours it was filtered

by using Whatman filter paper No.1. The extract was collected and again mixed with 200 ml of chloroform / ethyl alcohol separately and kept for next 72 hours with regular interval of shaking and then filtered. The extracting processes were repeated for at least three times. The chloroform / ethyl alcohol extracts were combined together. The solvent was evaporated by using Thin Film Rotary Evaporator under reduced pressure. Crude compound obtained was stored in refrigerator at 0° C for further investigation.

Examination of crude compounds by Thin Layer Chromatography (TLC): Thin Layer Chromatography is an important technique to detect or identify the presence of the number of compounds or number of components present in a crude extract or crude compound in which R_f value of each component was calculated by using this formulae: TLC was carried on glass plates (slides) coated with silica gel G type 60 (BDH, England).

$R_{f} = \frac{\text{Distance traveled by the component}}{\text{Distance traveled by the solvent front}}$

For TLC plates, slurry was prepared by the slow addition with shaking 30 g of absorbent (silica gel) to 100 ml of chloroform in a wide-racked capped bottle. A pair of microscopic slides was held together and dipped into the slurry, slowly withdrawn and allowed to drain momentarily while held over the bottle. The slides were parted carefully and placed horizontally in a rack; it was then dried in sunlight or in oven at 30-40 $^{\circ}$ C for 10-15 minutes (Furniss *et al.*, 1989). The crude extract was dissolved in the appropriate solvent (chloroform) and the solution of the compounds was then spotted with thin glass capillary tube at one end of the plate. The plate was then placed vertically with the spotted end downward in a solvent tank.

RESULTS AND DISCUSSION

Toxicity effect: Figure 1 showes that shiyalmutra leaf extracts possessed significant effect on the mortality of rice weevil. Mortality percentages were directly proportional to the concentrations of extract and exposure time. Insect mortality indicated that 5% leaf extracts possessed the highest toxicity and the mortality gradually increased with the increase of exposure time at all concentrations. This study showed harmony with the report of Shahjahan and Amin (2000) who stated that water extract of akanda, biskatali and neem had direct toxic effect on rice weevil and higher concentration of plant extract revealed higher toxic effect.

Repellency effect: The repellency effect of shiyalmutra leaf extract on rice weevil has shown in figure 2. The 5% extract revealed the maximum repellent action ($63.3 \pm 11.5\%$) on rice weevil. The repellent action increased with the increasing concentration of the extract. This study result showed agreement with the findings of Amin et al. (2000) who observed the leaf extract of biskatali on lesser grain borer and reported that 4 % extract showed strong repellent effect.



Figure 1. Cumulative mortality rates of rice weevil after treatment of shiyalmutra leaf extract.

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Figure 2. Repellency rates of rice weevil after treatment of different concentrations of shiyalmutra leaf extract. Bars with no common letter are significantly different.

Residual toxicity effect: The efficacy of shiyalmutra leaf extract as protectant for rice grain is shown in figure 3. Progeny emergence rate was found highest in 1% leaf extract at 14 days. It was lowest in 5%. In 21 days, progeny emergence rate was found highest in 1% leaf extract at 14 days. It was found lowest in 5%. The results are statistically similar for 14 and 21 days of progeny emergency.



Figure 3. Effect of different concentrations of shiyalmutra leaf extract on the progeny production of rice weevil.



Figure 4. TLC of chloroform extract of shialmutra (Hexane:Ethylacetate (v/v; 10: 1).

Plant species	Hexane : Ethylacetate	Detected component	R _f value
Blumea lacera	10:1	$egin{array}{c} \mathbf{S}_3 \ \mathbf{S}_2 \ \mathbf{S}_1 \end{array}$	3.7 2.7 1.6

Table 1. Rf values detected components of shialmutra leaf extracts

Chemical investigation: The crude compounds were extracted from the powder of shiyalmutra leaves with different non-polar and polar solvents like chloroform and ethanol. The TLC of chloroform extracts of shiyalmutra (Figure 4) show two distinct compounds at hexane: ethyl acetate (10:1, v/v) suggesting that it contained three distinct compounds, designated as S_1 , S_2 and S_3 , respectively. Table 1 shows the higher R_f value indicating the most non-polar compound and lower R_f value indicating the most polar compound. Here the intensity of non-polar compound like S_1 was too much high comparison with S_2 and S_3 . Results on insect bioasseys indicate that leaves of shiyalmutra possessed toxic and repellent properties. The results are in agreement with the findings of Roy et al. (2005) who reported the toxic and repellent properties of shiyalmutra leaves against rice weevil. The biological activity of shiyalmutra encourages us the insecticidal activity for pest management.

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