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Research Article

Pesticidal Properties on the Leaf Extracts of *Strychnos-Nux-Vomica* Plant

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Abstract

Strychnos-nux-vomica which belongs to the family *loganiacea* is a medium-sized tree. Other names of *Strychnos-nux-vomica* are Kanjiram, Kuchla, Kupilu. In India, the quality/toxicity of traditional medical crude and processed *Strychnos* seeds can be controlled by examining the toxic alkaloids using established HPLC methods and/or HPLC-UV methods. *Strychnos nux-vomica* is also used in homeopathy. *Strychnos* has not been proven effective for the treatment of any illness. Since the seeds contain strychnine poison, conventional doctors do not recommend it as a medicine. In the present study reveals that, plant extracts of *Strychnos-nux-vomica* leaves in ethyl acetate solvent is highly toxic against adults of *Sitophilus oryzae*. Higher doses and exposure time are required to achieve 100% mortality for the adults of *Sitophilus oryzae*. The ethyl acetate extract of *Strychnos-nux-vomica* could be used as a potential grain protectant against *Sitophilus oryzae*. The use of botanical materials as insecticides will benefit our agricultural sector. They are not only of low cost, but have no environmental impact in term of insecticidal hazard. Therefore, the findings of the current experiments strongly support the use and exploration of botanicals in pest management practices.

Key words: *Strychnos-nux-vomica*, *Sitophilus oryzae*, Disc method

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1. Introduction

Strychnos-nux-vomica which belongs to the family *loganiacea* is a medium-sized tree. Other names of *Strychnos-nux-vomica* are Kanjiram, Kuchla, Kupilu. The wood is dense, hard white, and close-grained. The branches are irregular and are covered with a smooth ashen bark [1]. The leaves have an opposite decussate arrangement, short stalked, are oval shaped, also have a shiny coat and are smooth on both sides. The leaves are about 4 inches (10 cm) long and 3 inches (7.6 cm) wide. The

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flowers are small with a pale green colour with a funnel shape. They bloom in the cold season and have a foul smell. The fruit are about the size of a large apple with a smooth and hard shell which when ripened is a mild shade orange colour.

In India, the quality/toxicity of traditional medical crude and processed *Strychnos* seeds can be controlled by examining the toxic alkaloids using established HPLC methods and/or HPLC-UV methods. *Strychnos nux-vomica* is also used in homeopathy. *Strychnos* has not been proven effective for the treatment of any illness. Since the seeds contain strychnine poison, conventional doctors do not recommend it as a medicine. It is on the Commission E list of unapproved herbs, because it is not recommended for use and has not been proven to be safe or effective. There is also no clinical trial evidence of *Strychnos* supporting it being a viable cancer treatment.

Prevention of food losses during postharvest storage is of paramount economic importance [2]. The major pests of stored grain are the primary pest those which are capable of penetrating and infesting intact kernel of grain. High moisture weeds, seeds and grains were damaged by primary pests. The important primary pests are *Sitophilus oryzae* commonly called the rice weevil. It is the most common and the most destructive pest of stored grains [3].

The chemical compounds present in *Strychnos nux-vomica* plant are likely to be biologically active even at lower concentrations. No work had been reported on the extract of the leaves of *Strychnos-nux-vomica* collected from Thrissur district, Kerala. The present work reports the pesticidal activity of three different extracts from *Strychnos nux-vomica* leaves against *Sitophilus oryzae*.

2. Materials and Methods

Plant material

Leaves of *Strychnos-nux-vomica* plant were collected from different areas of Thrissur district. The leaves of plant were collected in the middle of January 2015. Leaves was collected from the plant were shade dried. After drying the plant materials were powdered. Powders of the leaves were used for extraction in different solvents. Fifty gram of powder was used for extraction.

Method of extraction

Extraction: The leaves collected were shade dried. Coarsely powdered 50g of the plant material was extracted with 500 mL of petroleum ether, ethyl acetate and ethanol. The extraction was carried out in a round bottom flask by boiling the material in the solvents with a water condenser. Refluxed the material until the solvent started to boil and the hot content was left standing overnight. Then filtered and collected the extract and added fresh solvent to residue. The process is repeated three times to complete the extraction. The combined extract collected was reduced to 20 ml.

Crude extract dilution

From each crude extract 15mg, 30mg and 50mg extracts were accurately weighed and serially dissolved in 1ml of acetone. Then it was made upto 100ml by using distilled water. Thus from each extract 150ppm, 300ppm, and 500ppm concentration of test solutions were prepared. Test solutions of three different concentrations of each of the nine extracts were used for testing the *Sitophilus oryzae* in three time periods(24hr,48hr,72hr) [4].

Treatment- pesticidal activity

Lab studies have been carried out to ascertain pesticidal property of the plant extract *(Strychnos-nux-vomica)* against *Sitophilus oryzae.*

Disc method

The methodology used for contact assay with impregnated filter paper was proposed [5]. Sheets of filter paper (diameter 3cm) were impregnated with test solution (500ppm, 300ppm, 150ppm) .One ml of each extract solution was spread with the help of pipette over the filter paper and placed into petri dishes, while control sheets were impregnated by without any plant extract. The filter paper discs impregnated with extracts were air dried until complete evaporation. Then they were placed into and 25 adults of *Sitophilus oryzae* were released in each treatment and control dish, with three replicates per treatment and control plate. The plates were wrapped in plastic film to prevent escape of pest. Small holes were made on the plastic film for aeration. Mortality counts were taken after 24hr, 48hr, 72hr intervals of treatment and expressed as percentages of mortality. Dead insects were counted, death being ascertained by the complete absence of movement.

Data analysis

The pests *Sitophilus oryzae* were collected. Only adults were taken for bioassay. Plant leaves were sorted and identified to species level. Plant extracts screened for pesticidal activity was made. Data were grouped in Table1. Pest mortality was measured in percentage and if the control mortality was ranged between 5-20%, it was corrected using Abbotts formula [6].

Percentage of pest mortality can be calculated by using the formula

$$Percentage\ \ mortality = \frac{Number\ of\ \ dead\ \ pest}{Total\ \ number\ of\ \ \ pest} \times 100$$

Standard deviations were calculated for three replicates and recorded for 24hr, 48hr and 72hr.

Graphs were based on time intervals (24hr, 48hr, and 72hr). Microsoft Excel package was used to prepare percentage mortality graphs against different concentrations of plant extract. Each graph compares the percentage of mortality of three solvents (petroleum ether, ethyl acetate, ethanol).

3. Results and Discussion

Pesticidal activity of the plant extract (*Strychnos-nux-vomica*) in three different solvents (Ethanol, Petroleum ether, Ethyl acetate) was tested against adults of *Sitophilus oryzae*, at different concentrations (150ppm, 300ppm, 500ppm). The experiment showed a relation of pest mortality level among 3 time intervals (24hr, 48hr, and 72hr). The results indicate variation in mortality among the plant extracts tested against the selected pest. The summary of results, in which the percentage of mortality of adults of *Sitophilus oryzae* caused by different plant extracts are represented in Table1.

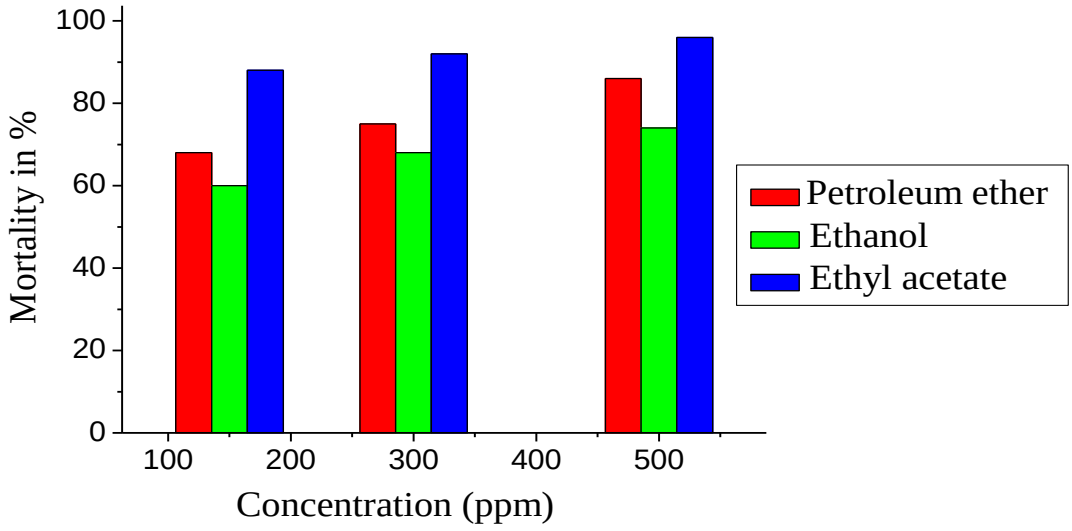
The results indicate that mortality values significantly increased depending on the increasing plant extract concentration and with time of exposure. As considering leaf extracts ethyl acetate medium *Strychnos-nux-vomica* is more effective than petroleum ether and ethanol. Studies were carried out with *Strychnos-nux-vomica* at the three concentrations in three different time interval. More over at 500 ppm concentration it gives 100% mortality.

The leaf extract of *Strychnos-nux-vomica* in three solvents (ethanol, petroleum ether, ethyl acetate) at three time intervals (24hr., 48hr., 72hr.,) also at different concentrations (150 ppm, 300 ppm, 500 ppm) the more effective one is ethyl acetate extract of *Strychnos- nux-vomica* leaves. And most significant result is produced after 72hr treatment i.e. 100% mortality.

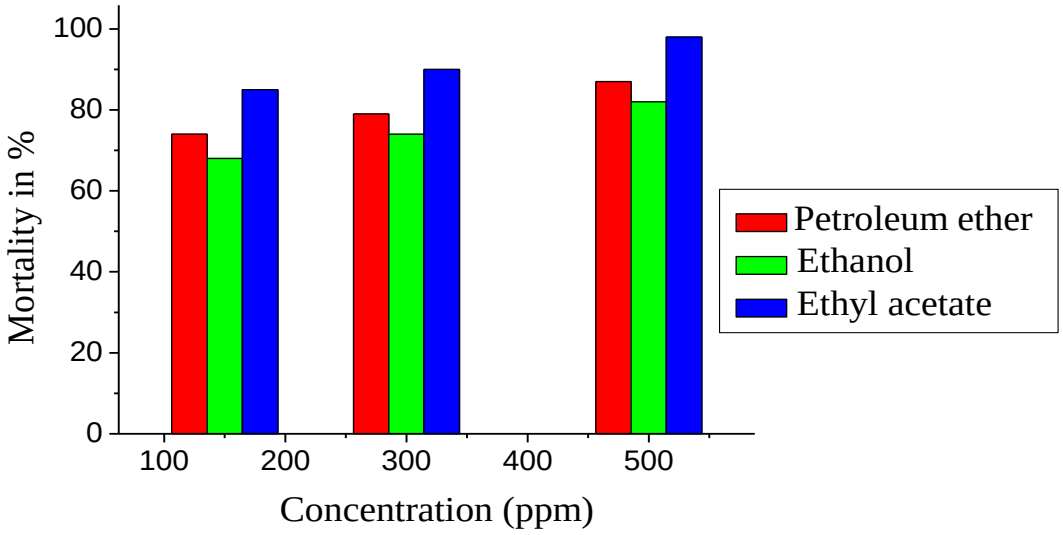
Table 1: Mortality percentage of *Sitophilus oryzae* treated with plant extracts of *Strychnos-nux-vomica* by disc method.

Name of the plant (Solvent)	Conc. (ppm)	No. of insects used in the study	Mortality (%)		
			24(hr) (%)	48(hr) (%)	72(hr) (%)
Strychnos-nux-vomica (Petroleum ether)	500 300 150	25 25 25	86±0.08 75±0.05 68±0.07	87±0.07 79±0.06 74±0.08	87±0.05 78±0.08 73±0.09
Control	-	25	-	-	-
Strychnos-nux-vomica (Ethanol)	500 300 150	25 25 25	74±0.08 68±0.03 60±0.08	82±0.06 74±0.05 68±0.09	84±0.08 75±0.08 70±0.07
Control	-	25	-	-	-
Strychnos-nux-vomica (Ethyl acetate)	500 300 150	25 25 25	96±0.02 92±0.04 88±0.08	98±0.05 90±0.06 85±0.06	100±0.03 96±0.06 90±0.07
Control	-	25	-	-	-

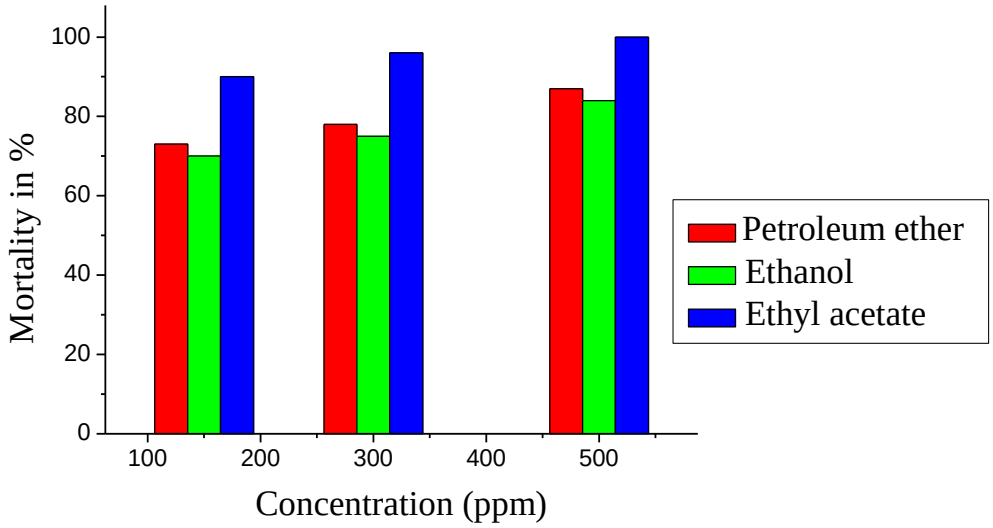
*values are mean percentage of three replication of experiment with 25 insects, ± SD



Graph1: Mortality percentage after 24hour treatment of *Sitophilus oryzae* with *Strychnos-nux-vomica.*



Graph 2: Mortality percentage after 48 hour treatment of *Sitophilus oryzae* with *Strychnos-nux-vomica*



Graph 3: Mortality percentage after 72 hour treatment of *Sitophilus oryzae* with *Strychnos-nux-vomica*

After 24 hour treatment higher mortality is in ethyl acetate extract and ethanol extract shows lesser mortality. Whereas petroleum ether extract shows medium mortality shown in Graph 1. The order of increase of mortality rate is: *Strychnos-nux-vomica* (ethyl acetate) > *Strychnos-nux-vomica* (petroleum ether) > *Strychnos-nux-vomica* (ethanol).

In Graph 2 for 48 hour treatment ethyl acetate extract shows higher mortality. Whereas ethanol and petroleum ether

extract shows similar mortality rates. The order of increase of mortality rate is: *Strychnos- nux-vomica* (ethyl acetate) > *Strychnos- nux-vomica* (petroleum ether) = *Strychnos- nux-vomica* (ethanol).

At 72 hour treatment ethyl acetate extract attain 100% mortality. Whereas ethanol and petroleum ether extract shows almost similar results was obtained from Graph 3. The order of increase of mortality rate is: *Strychnos-nux-vomica* (ethylacetate) > *Strychnos-nux-vomica* (petroleum ether) = *Strychnos-nux-vomica* (ethanol).

From the analysis of above three graphs, it shows that ethyl acetate extract of *Strychnos- nux-vomica* produce higher pesticidal activity than petroleum ether and ethanol extract. Mortality value increases with concentration of extract and also with time of exposure.

The present work reveals the pesticidal effect of the leaf extracts of *Strychnos- nux-vomica* in three solvents (ethyl acetate, petroleum ether, ethanol) against adults of *Sitophilus oryzae*. The experiments reveal that ethyl acetate extract of *Strychnos- nux-vomica* offers higher percentage of mortality to *Sitophilus oryzae*. Meanwhile *Strychnos- nux-vomica* extract in ethanol shows the least mortality rate.

GC-MS analysis of *Strychnos- nux-vomica* from the earlier report show that highly poisonous alkaloids strychnine and brucine were present in high percentage. The higher mortality rate in ethyl acetate extract may be due to the presence of strychnine and brucine.

The toxicity of plant extract to stored product pest, *Sitophilus oryzae* is influenced by the chemical composition of the plant, time of exposure, plant parts used, method of extraction and concentration of the plant extract.

The results of the present study indicate that, by considering the percentage mortality as a main index, *Strychnos-nux-vomica* in ethyl acetate proved to be the most effective plant extract potential to be use as a grain protectant. These plants have a range of chemicals which can be isolated and used for pest control. The test plants being medicinal would yield environmentally sound chemicals having no harmful effects on the non target organisms also these pesticidal properties can be harnessed cheaply for use in agriculture and related fields.

Conclusion

The current study strongly recommends the need of exploring botanical derivatives as pest control agents. Even though there are differences in concentration of extracts, all solvent extracts could act as a toxic agent to prevent the proliferation of the test animal.

The toxicity of plant extract to stored product pest, *Sitophilus oryzae* is influenced by the chemical composition of the plant, time of exposure, plant parts used, method of extraction and concentration of the plant extract.

The studied plant extracts of *Strychnos-nux-vomica* leaves in ethyl acetate solvent is highly toxic against adults of *Sitophilus oryzae*. Higher doses and exposure time are required to achieve 100%mortality for the adults of *Sitophilus oryzae*. The ethyl acetate extract of *Strychnos-nux-vomica* could be used as a potential grain protectant against *Sitophilus oryzae*. The use of botanical materials as insecticides will benefit our agricultural sector. They are not only of low cost, but have no environmental impact in term of insecticidal hazard. Therefore, the findings of the current experiments strongly support the use and exploration of botanicals in pest management practices.

The results of pesticidal activity of a number of higher plants based on traditional knowledge strongly indicate that plants are endowed with pesticidal

property that can be harnessed cheaply for use in agriculture and related fields. The need to use plant-based products arises from the fact that the synthetic pesticides are harmful to humans, and the entire ecosystem due to high toxicity and persistence. Also, they are too expensive for the poor farmers in the developing countries of the world. On the other hand, plant- based products are cheap and bio-degradable and are therefore environmentally friendly. However, an agricultural programme that depends essentially on plant-based materials must be backed-up by a vigorous research programme into new plant sources.

Therefore, the leaf extract of *Strychnos-nux-vomica* in ethyl acetate solvent, a potentially useful agent to control *Sitophilus oryzae*, has to be better explored by doing detailed chemical analysis of the active compounds present in it and by understanding the mechanism involved in action.

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