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LARVICIDAL EFFICACY OF THREE AROMATIC PLANTS AGAINST DENGUE VECTOR, *AEDES AEGYPTI*

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Aedes aegypti is the main carrier for viruses that cause dengue, dengue hemorrhagic and yellow fevers. Insecticide use to control this vector has led to the development of mosquito resistance, environmental pollution, and undesirable effects on non-target organisms.use of environment friendly and biodegradable natural insecticides of plant origin have received renewed attention as agents for vector control. This quality increased the demand of plant extracts. The present study was conducted to determine the effectiveness of leaf extracts of *Glyricidia maculate,Manihot esculanta, Glycosmis pentaphylla* against dengue and *chikunguniya* vector *Aedes aegypti.*

The larvicidal activity was determined against the early 4th instar larvae at concentrations of 500,1000 ppm. Larval mortality of *Aedes aegypti* was observed against petroleum ether, methanol and ethyl acetate extracts of *Gliricidia maculate, Manihot esculanta* and *Glycosmis pentaphylla* after 24h and 48 h. *Manihot esculanta* extract in ethyl acetate and *Glycosmis pentaphylla* extract in petroleum ether provided highest mortality rate of 69.9% and 69.6% respectively Petroleum ether extract of *Glycosmis pentaphylla* shows the least variation in mortality between 500ppm and 1000ppm at 24 hrs.

Keywords: Dengue, natural insecticides.

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The threat of insects and other pests such as mosquitoes, cockroaches, rodents,parasitic worms, pathogens and snails, has been well known and challenged by man. The Aedes aegypti mosquito is the main vector that transmits the viruses that cause dengue. The viruses are passed on to humans through the bites of an infective female Aedes mosquito, which mainly acquires the virus while feeding on the blood of an infected person. Yellow fever, Chikungunya, Viral haemorrhagic fevers, Dengue fever etc are the diseases caused by Aedes aegypti.

Plants are considered as a rich source of bioactive chemicals and they may be an alternative source of mosquito control agents. Man has struggled hard to eradicate mosquitoes by adopting various techniques. Since the discovery of DDT, control of mosquitoes has been mostly through synthetic chemicals like Malathion and synthetic pyrethroids, these non-biodegradable and hazardous chemicals have created environmental pollution and non-target organisms and man have been severely affected by these toxic pesticides This has necessitated the need for environmental friendly biodegradable methods for vector control. In recent years, the focus is mainly on the use of environmental friendly and biodegradable materials as pesticides. Several phytochemicals extracted from various parts of the plant have been found to be effective in vector control. Numerous workers have tested the efficacy of several plant extract using different mosquito species.

The current study tested the larvicidal activity of 15aromatic plants. Out of the selected, three plants which showed good larvicidal activity were used for standard extraction. The three plants were Glyricidia maculate, Manihot esculenta, Glycosmis pentaphylla. The selection was done by doing Crude Extraction Method. Crude extraction was followed by Standard Soxhlet Extraction using different solvents.

Materials and Method

Plant Material

The plant materials collected from different parts of kodungallur, identified the name and species with the help of Botany Department Christ College Irinjalakuda. The current study tested the larvicidal activity of 15aromatic plants. Out of the selected, three plants which showed good larvicidal activity were used for standard extraction. The three plants were Glyricidia maculate, Manihot esculenta, Glycosmis pentaphylla. The selection was done by doing Crude Extraction Method. Crude extraction was followed by Standard Soxhlet Extraction using different solvents.

Extraction

The dried leaves of Gliricidia maculate Manihot esculenta and Glycosmis pentaphylla leaves were mechanically grinded to get fine powder. 20g of dried powder were taken within a thimble made of filter paper. The solvent (250 ml of ethanol or petroleum ether or ethyl acetate) was added to a round bottom flask, which is attached to a Soxhlet extractor and condenser on an isomantle. The crushed plant material was loaded into the thimble, which is placed inside the Soxhlet extractor. The side arm is lagged with glass wool. The solvent was heated using the isomantle and began to evaporate, moving through the apparatus to the condenser. The condensate then drips into the reservoir containing the thimble. Once the level of solvent reaches the siphon it pours back into the flask and the cycle begins again. The process should run for a total of 24 hours. Once the process has finished, the ethanol should be evaporated, leaving a small yield of extracted plant material (about 2 to 3 ml) in the glass bottom flask. The mass of this remaining extract is measured and it stored in an air tight bottle for further experiment.

Larvicidal bioassay

The graded series of extracts were prepared using ethanol, petroleum ether and ethyl acetate as the solvent. Ten 4th instar larvae were released into 250 ml glass beaker containing 100 ml solution. Four replicates were setup for each concentration. Larvae were counted as dead when they were not coming to the surface for respiration or probe insensitive.Observation on mortality of the larvae was recorded after 24 hour of continuous exposure. The dead larvae in four replicates were combined and expressed as percentage of larval mortality for each concentration. In controls the larvae were exposed to the solvent.

Data analysis

Larval mortality was measured in percentage andif the control mortality wasranged between 5–20%, it was corrected using Abbott's formula [30].

$$corrected\ mortality = \frac{\%\ test\ \ mortality - \%\ control\ mortality}{100 - \%\, control\ mortality} \times 100$$

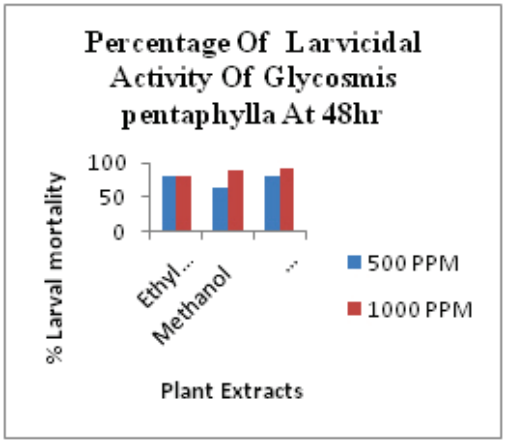
Standard deviations were calculated by the formula,

Standard Deviation $= \sqrt{\frac{\sum (x - \bar{x})^2}{n-1}}$

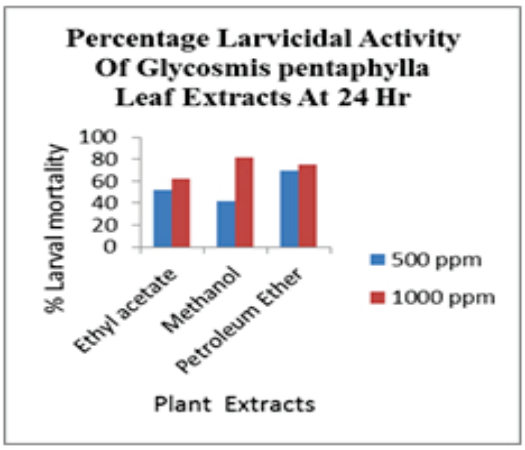
Results and Discussion

The relative larvicidal effects of three plants extracted in different solvents were noted. Among the three plants in three solvents, Manihot esculanta extract in ethyl acetate and Glycosmis pentaphylla extract in petroleum ether provided highest mortality rate of 69.9% and 69.6% respectively. Ethyl acetate extract of Glycosmis pentaphylla showed the same mortality percentages for both 500 and 1000 ppm. Manihot esculenta has comparatively equal mortality rates in methanol and petroleum ether extracts at 48hrs. At 24 hr Ethyl acetate extract of Manihot esculenta shows highest variation between 500ppm and 1000ppm

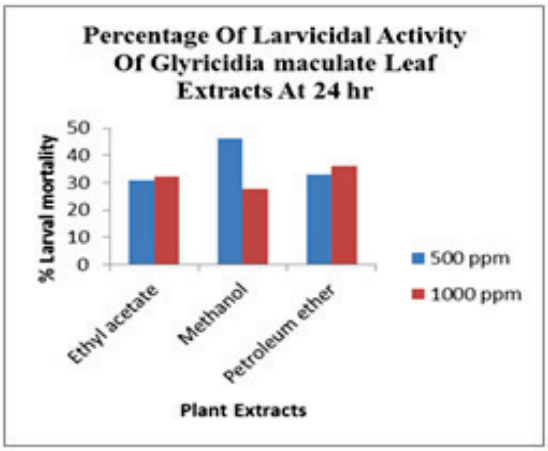
In general, it is observed that the methanolic extracts show highest variation in mortality between 500ppm and 1000ppm at 24 hrs. Mortality rate increases to more than 90% in 1000 ppm while it remains around 30%in 500 ppm. Different extracts of the same plant have also shown similarity in mortality percentages. Gliricidia maculate extract in ethyl acetate & petroleum ether and Manihot esculanta extract in ethyl acetate provided (30.72%, 33.03%, and 40%) the least mortality rate compared to the other extracts at lowest concentration and at least interval of time.In the present study, the methanolic extracts show highest variation in



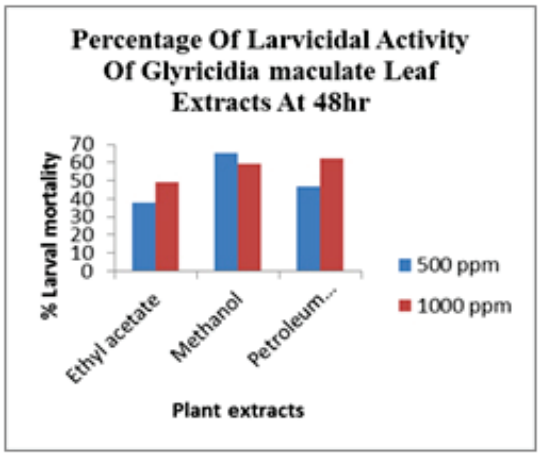
%Larvicidal Activity Of *Glycosmis pentaphylla*



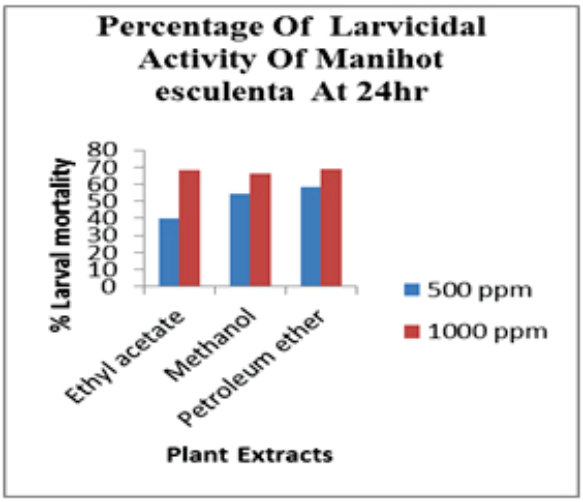
Leaf Extracts At 24 and 48hrs Against *Aedes aegypti.*



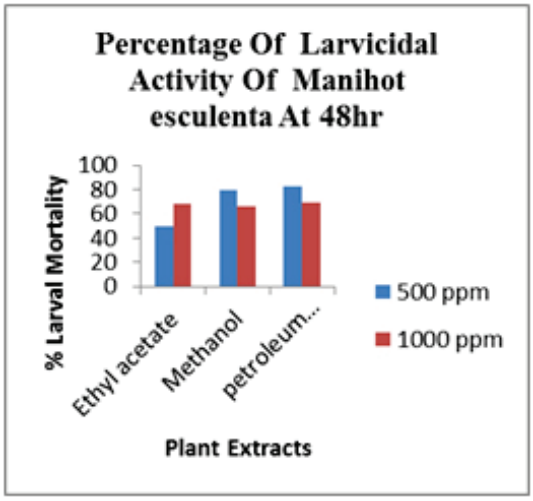
%Larvicidal Activity Of *Glyricidia maculate*



Leaf Extracts At 24 and 48hrs Against *Aedes aegypti.*



%Larvicidal Activity Of *Manihot esculenta*



Leaf Extracts At 24 and 48hrs Against *Aedes aegypti.*

mortality against Aedes aegypti between 500ppm and 1000ppm at 24 hrs. . Mortality rate increases to more than 90% in 1000 ppm while it remains around 30% in 500 ppm.

Recent studies on the larval and pupal mortality of Aedes aegypti after treatment of the isolated compound saponin from ethyl acetate extract of Achyranthes aspera was effective against the larvae of A. aegypti and Culexquinquefasciatus with LC50 value of 18.20 and 27.24 ppm, respectively [6].The neem for mulation, NeemAzal, produced an overall mortality or inhibition of emergence of 90% (EI90, when third-instar larvae were treated) at 0.046, 0.208, and 0.866 ppm in A. stephensi, C. quinquefasciatus, and A. aegypti, respectively.In the present study, the methanolic extracts show highest variation in mortality against Aedes aegypti between 500ppm and 1000ppm at 24 hrs. .

Mortality rate increases to more than 90% in 1000 ppm while it remains around 30% in 500 ppm. Ethyl acetate extract of Glycosmispentaphylla showed the same mortality percentages for both 500 and 1000 ppm.The results of this study will add to a great reduction in the application of synthetic insecticides, which in turn raise the opportunity for eco-friendly control of various vectors by botanical pesticides

Conclusion

The present study investigated the larvicidal activity of three traditionally important aromatic plants. Extracts from the leaves of Glycosmis pentaphylla, Manihot esculenta and Glyricidia maculate have shown potential larvicidal activity against Aedes aegypti. Application of these extracts could be very useful to eradicate the larvae of Aedes aegypt iand related vector borne diseases. The extracts in viable form can be applied to disturb breeding sites such as containers, ranging from watering cans, discarded plastic bags to ground depressions and blocked roof gutters. This would offer an eco-friendly and less expensive way to reduce the problem of the Aedes aegypti, especially that all of the examined plants are commonly available and used in India.

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