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Research Article

Larvicidal and antimicrobial activities of *Andrographis paniculata*

A.Chanthuru, M. Munees Prabhu and O.S. Aysha*

Dept of Microbiology, Mohamed Sathak College of Arts and science,
Sholinganallur, Chennai – 119, Tamil Nadu, India

*Corresponding author e-mail: *ayshamohamed786@gmail.com*

Abstract

Andrographis paniculata is an important ingredient in traditional Indian medicines. In the present paper deals the level of larvicidal activity of ethyl acetate extract of leaf and root of *Andrographis paniculata* at different concentrations *viz.*, 500, 1000 and 1500 ppm has been investigated. The results obtained show that this plant material exhibited significant activity and could be considered as potent natural larvicidal agent. The antibacterial activities of ethyl acetate extract of leaf and root, *A. paniculata* were screened against various pathogenic bacteria by 'agar well diffusion' method. The plant extracts showed various levels of activity on different test organisms among that ethyl acetate extract of *A. paniculata* stem was found to be the most potent extract.

Keywords: *Andrographis paniculata*, larvicidal activity, *Culex quinquefasciatus*, Antibacterial activity,

Introduction

Mosquitoes are the major vector for the transmission of malaria, dengue fever, yellow fever, filariasis, schistosomiasis and Japanese encephalitis (Das and Ansari, 2003). In India, malaria is one of the most important causes of direct or indirect infant, child and adult mortality with approximately two to three million new cases arising every year. Controlling of these vectors is being achieved for a long time, by using synthetic chemicals. But the chemicals may cause pollution problem and help to develop resistance in mosquito species (Das and Rajagopalan 1981). Hence, an alternative approach for mosquito control is the use of extracts of plant origin. The phyto-chemicals derived from plant sources can act as larvicides, insect growth regulators, repellents, ovipositional attractants and have deterrent actions as observed by many researchers (Yogananth *et al*., 2012) Antibiotics are sometimes associated with side effects (Cunha, 2001) whereas there are some advantages of using antimicrobial compounds of medicinal plants, such as often fewer side effects, better patient tolerance, relatively less expensive, acceptance due to long history of use and being renewable in nature (Vermani and Garg, 2002). All these data high lights the need for new alternative drug regimens. Studies on the natural plant products for their efficacy as larvicides and antimicrobials during the last decade have indicated them to be possible alternatives to synthetic chemical insecticides and antibiotics (Thomas *et al*., 2004 and Dharmagadda *et al*., 2005).

Andrographis paniculata also known commonly as "king of bitters or is a member of the plant family *Acanthaceae.* It is a plant that has been effectively used in traditional Asian medicines for centuries. Its perceived "blood purifying" property results in its use in diseases where blood "abnormalities" are considered causes of disease, such as skin eruptions, boils, scabies, and chronic undetermined fevers.The plant contains andrographolide, neo-andrographolide, deoxy- andrographolide and andrographiside. The leaves contain active like andrographolide, homo and rographolide andrographestrol and andrographone (Sharma *et al*., 1991).

Materials and Methods

Test organism

The larvae used to test for the larvicidal activity were obtained from colonies of *Culex quinquefasciatus* mosquitoes cultured and maintained in the laboratory at a temperature of 28°C ± 2°C and 80 - 90% relative humidity. The larvae were fed with mice feed and yeast powder in the ratio of 3:1. They were transferred to another clean bowl for three days at 24hrs interval and the water was aerated with the aid of an air pump.

Bioassay

Larvicidal activity of the mosquito *C. quinquefasciatus* was assessed by following the standard WHO method (1996). The ethyl acetate extracts of leaf and root of *Andrographis paniculata* for assayed against larvicidal activity was carried out at different concentrations ranging from 500, 1000 and 1500 ppm in distilled water. Ten second, third and fourth instar larvae of *C. quinquefasciatus* were collected separately and transferred gently to the test medium and simultaneously a control was maintained with ethanol-fresh tap water mixture. The larval mortality in both treated and control were recorded every 4 h, 8 h and 12 h. Dead larvae were identified when they failed to move after probing with a needle in the siphon or cervical region. The experiments were replicated three times and conducted under laboratory conditions at 25 - 30°C and 80 - 90% relative humidity.

Antimicrobial activity

Microorganisms tested

A total of eight bacterial cultures (*E.coli, Salmonella typhi, Salmonella paratyphi, Klebsilla penemonia, Staphylococcus aureus*) were used in this study. The bacterial strains were grown in nutrient broth at 37^0C and they were stored on nutrient agar slants for future use.

Well-in agar method

Anti-bacterial activity of plant extracts was tested by a modified well-in agar method (Sinclair and Dhingra, 1995). From the nutrient broth, the inoculum suspension was swabbed uniformly over the Muller Hilton Agar by using of sterile cotton swab. Subsequently, using a sterile borer, well of 0.5 cm diameter was made in the pathogen inoculated media. Different concentrations, i.e., 20, 40 and 60 µl of each extract were aseptically filled into the well. Later the plates were placed at room temperature for an hour to allow diffusion of extract into the agar. Then the plates were incubated for 24 h at 37^0C. The results were recorded by measuring the diameter of inhibition zone at the end of 24-48 h.

Statistical Analysis

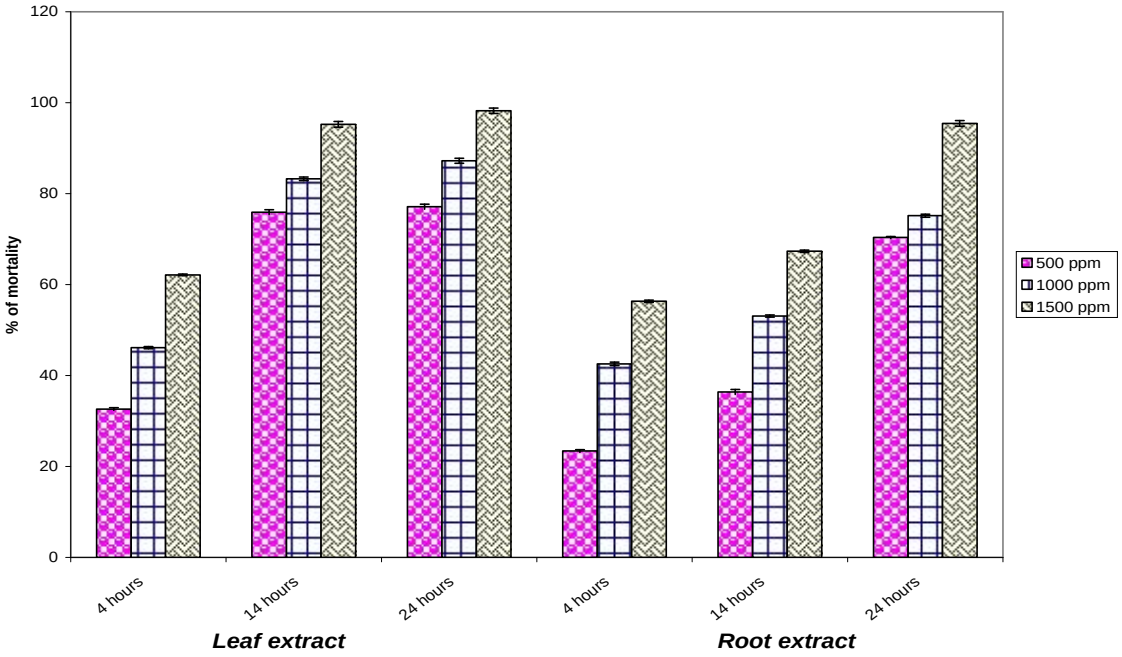
All the data were subjected to Duncan's multiple Range Test (DMRT). It was alone using the spss version 2007 software.

Results and Discussion

Larvicidal activity

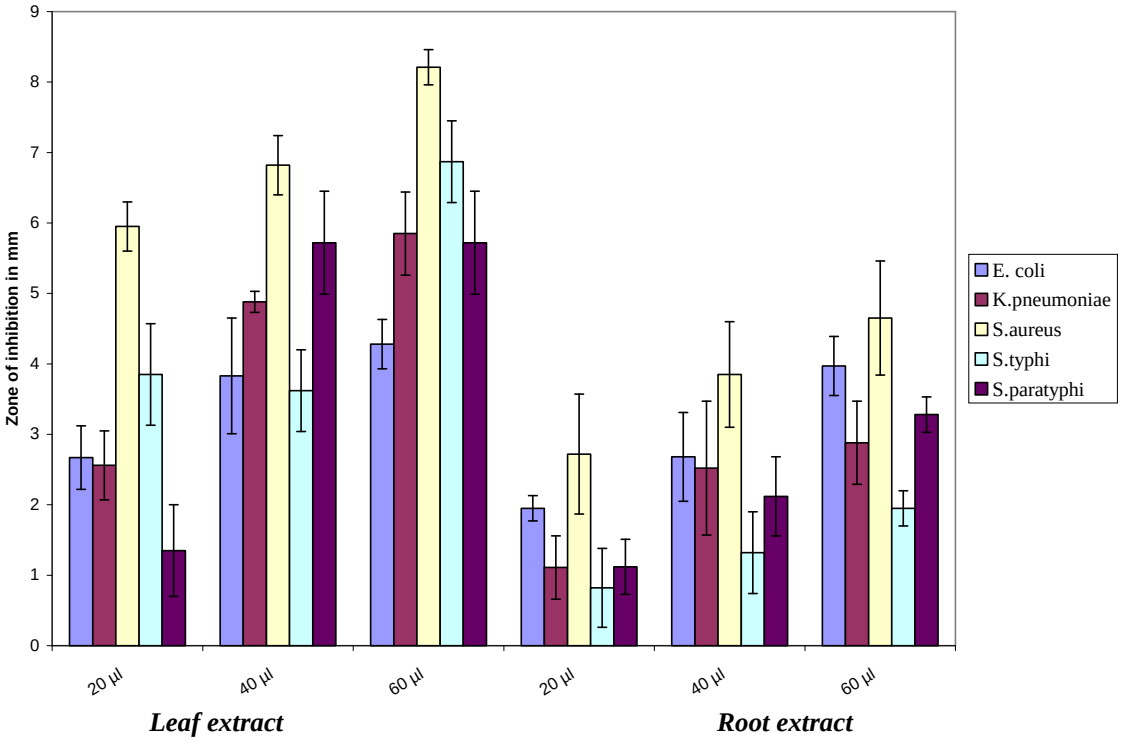
The larvicidal activity of ethyl acetate extracts of leaf and root of *A. paniculata* against *C.quinquefasciatus* mosquito larvae were given in Fig 1. The larvicidal activity of ethyl acetate extracts of leaf and root of *A. paniculata* showed 98.25% and 95.24 of death with the use of 1500 ppm concentrations, respectively, after 12 hrs.

Fig 1. Larvicidal activity of *Andrographis paniculata* against *Culex quinquefasciatus*



Statistical analysis data or expressed as Means ± Standard deviation

Fig 2. Antibacterial activity of *Andrographis paniculata* against bacterial pathogens



Statistical analysis data or expressed as Means ± Standard deviation

In 12 hrs, 1000 ppm concentration killed more than 75% of the larvae in both extracts. In 12 hrs, 500 ppm concentration killed more than 70% of the larvae in both extracts. Among the two extracts, the leaf extract of *A. paniculata* was found more lethal than root extracts. This work demonstrates the potency of *A. paniculata* in the control of mosquito larvae. The high mortality recorded for leaf extract might be attributed to deficiency of dissolved oxygen in the water. Senthil Nathan *et al*., (2005) reported that the plant allelochemicals may be quite useful in increasing the efficacy of biological control agents because plants produce a large variety of compounds that increase their resistance to insect attack. This result compared favourably with that from other species, for example, Elimam *et al*., (2009) reported that leaf extract of *Calotropis procera* has showed larvicidal activity against the mosquitoes *Anopheles arabiensis* and *C. quinquefasciatus*.

Antibacterial activity

The result of the anti-bacterial activity tests of *A. paniculata* leaf and root extracts are presented in Fig 2. The maximum zone of inhibition i.e., 8.21mm and 6.87mm were registered against *Staphylococcus aureus*, *Salmonella typhi* respectively. The ethyl acetate extracts of root exhibited minimum activity in some of the organisms, namely *Salmonella typhi* and *Klebsilla penemonia*. Among these two extracts, the leaf extracts showed the maximum antibacterial activity than root extracts. Agreement with the earlier findings methanolic extracts of Red rose petals gave the zone of inhibition of 25 mm, 26 mm, 27 mm against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* respectively (Hirulkar *et al*., 2010).

The results of the study also supports the traditional application of the plant and suggests that the plant extracts possess compounds with antibacterial properties that may be used as antibacterial and antifungal agents in novel drugs for the treatment of gastroenteritis, uretritis, dysentry, typhoid fever, allergy, anaemia and skin disorder.

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