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Application of Crude Aqueous Solution as a Larvicidal and Antibacterial Agent Extracted from Coconut Shells

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Abstract: ***Dry coconut shell pieces when subjected to extraction yields crude aqueous solution, which is further purified by distillation method. The larvicidal and antibacterial activity was studied for this crude aqueous solution. 3rd and 4th instar larvae of Culex quinquefasciatus were used as clinical subject. Escherichia coli, Staphylococcus aureus, Salmonella typhi and Streptococcus pyogenes were used for antibacterial study. 10% v/v sample of crude aqueous solution gave 100% mortality within 24 hours whereas Salmonella typhi and Staphylococcus aureus were found highly susceptible to crude aqueous solution as it gave 31mm and 24mm zone of inhibition respectively.***

Keywords: *Culex quinquefasciatus*, Larvicidal activity, Antibacterial activity, Crude Aqueous solution

1. Introduction

The species of Culex mosquito can transmit Lymphatic filariasis, Avian malaria, St. Louis encephalitis, Western equine encephalitis, Japanese encephalitis and West Nile fever.[1,2] It is one of the most common mosquito species causing public health problem in India both rural and urban particularly among low socio-economic communities. [3] From last one decade's number of cases of encephalitis has increase such an extent that now it has become as an epidemic prone disease. Most of the cases were from Uttar Pradesh followed by West Bengal, Assam and Bihar.[4,5] The frequent and often predictable outbreaks of encephalitis in different parts of the country constitute a huge challenge to public health in India.

Microorganisms such as *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhi* and *Streptococcus pyogenes* are common micro-organism which are responsible for food borne diseases, Gastroenteritis, urinary tract infections, neonatal meningitis, Typhoid fever, Strep throat, scarlet fever, mastitis and necrotizing fasciitis.[6,7] Antibiotic resistance has been increased in bacteria last several decades.[8,9,10] The use of plant extracts with known antimicrobial properties, can be of great significance in therapeutic treatments.

Cocos nucifera plant species also known as 'Kalpvriksha' in Sanskrit is abundant in India and its each and every part being used by mankind since past many centuries. [11,12,13]It is used for food as an ingredient, Oil, Timber, Rope, Decorative materials, Toys, Fuel and Charcoal. [14,15,16]It is also used as Antifungal, antimicrobial agent for skin diseases and oral health issues, treatment of kidney problems, asthma, bronchitis, cold and cough. It is also proved to be an antioxidant and anti-cancer agent.[17,18,19,20]

Literature study and research has shown larvicidal activity of its crude oil.[21]While extraction of crude oil from coconut shell, amount of crude aqueous solution is much higher than crude oil i.e. approximately in 9:1 ratio in initial stage and purification of crude water from crude oil is also carried by an easy method such as distillation method.

Coconut is largely used in households, Hotel industries and oil refineries, where its karnel is used for the purpose and the shell left behind as a waste.

In the present research communication larvicidal and antibacterial activity of crude aqueous solution is described.

2. Materials & Method

1) **Preparation of Crude aqueous solution:** 1kg of dried & cleaned coconut shell pieces were subjected to bioactive crude oil extraction by heating it in earthen pot using patented method.[22]during the extraction initially large quantity of crude aqueous solution obtained which on concentration yields dark brown viscous oil. The crude aqueous solution then purified from crude oil by using distillation method and was used for study.
2) **Larvicidal Test Preparation:**
   Bioassay was conducted in 500ml glass beakers containing 250 ml of de-chlorinated water for each species separately. The samples were prepared ranged from 1% v/v to 10% v/v of crude water. The test solution with four replicates was prepared according to the guidelines of WHO (2005).[23] A batch of 3rd and early 4th instar larvae (10 in no.) of *Culex quinquefasciatus* were collected from the field and introduced in each test solution and in an appropriate control solution. The results were observed at 24hour.
3) **Microbial Test Preparation**
   a) **Test microorganisms:** The antibacterial potency of crude water extract was evaluated using four bacterial strains causing food poisoning diseases. Two Gram Negative strain *(Escherichia coli* & *Salmonella typhi*) and Two Gram Positive strains (*Staphylococcus aureus* & *Streptococcus pyogenes*).
   b) **Screening of Antibacterial Activity:** The evaluation of antibacterial activity is done by Ditch Plate Method. In the following method Nutrient Agar was melted and poured into a 14 cm Petri dish. After

solidifying, three parallel strips of agar 1 cm. in width were cut out. Organisms to be tested were inoculated in parallel streaks 1.5-2 cm. apart at right angles to the ditches. The plates were incubated at 37°C for 24 hours and the zone of inhibition was measured.[24,25,26] The test was performed in 3 replicates along with a control test.

3. Results & Discussion

Larvicidal Activity

100% mortality rate were observed in 10%v/v sample and 80% in 5% v/v sample of crude water within 24hoursagainst *Culex quinquefasciatus* and it decreases as concentration of crude aqueous solution decreases. It takes more time to kill all targeted larvae to lower concentration sample unit. Also prolongation of larval growth has extended reasonably. In which development of mosquito larvae altered by presence of crude aqueous solution which further leads to larval death.

Anti-Microbial Activity

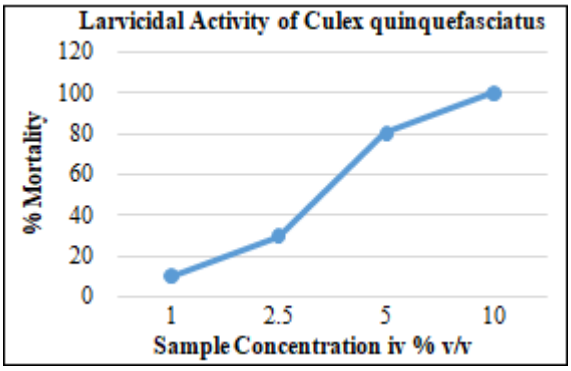
The study shows all four bacteria are susceptible to crude water in which *Salmonella typhi* and *Staphylococcus aureus* were found highly susceptible to crude water as they gave 31mm and 24mm zone of inhibition respectively. *Streptococcus pyogenes* and *Escherichia coli* are less susceptible in compared as they gave 20mm and 11mm zone of inhibition respectively.

4. Conclusion

Present study suggests the importance of crude water not only in the field of pesticides but also in antibacterial activity. By considering its huge abundant, simplicity in method of preparations and potential of larvicidal and antibacterial activity, this can be one of the cheapest alternate source to combat Culex mosquito species as well as skin and other oral diseases.

Table 1: % Mortality of *Culex quinquefasciatus* Larvae.

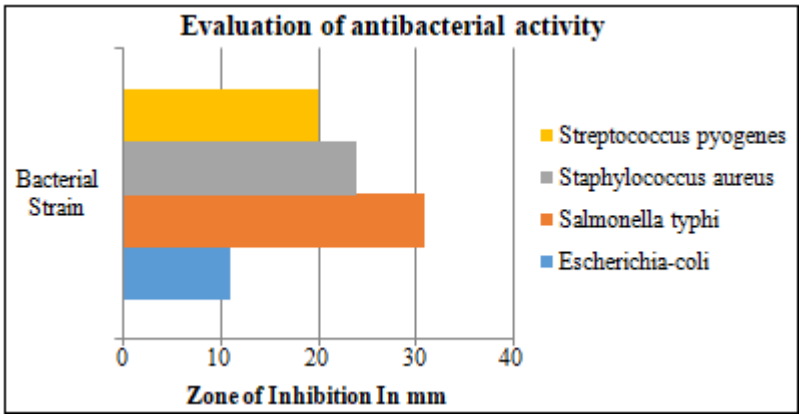
S. No.	Mosquito Species	Sample Concentration In % v/v	% Mortality
1	*Culex quinquefasciatus*	1	10
2		2.5	30
3		5	80
4		10	100



Graph 1: % Mortality of *Culex quinquefasciatus* Larvae

Table 2: The evaluation of antibacterial activity

Sr. No.	Bacterial Strain	Zone of Inhibition (In mm)	Mean ±SD (In mm)
1	*Escherichia coli*	11	1.4
2	*Salmonella typhi*	31	4.9
3	*Staphylococcus aureus*	24	2.9
4	*Streptococcus pyogenes*	20	2.9



Graph 2: The evaluation of antibacterial activity.

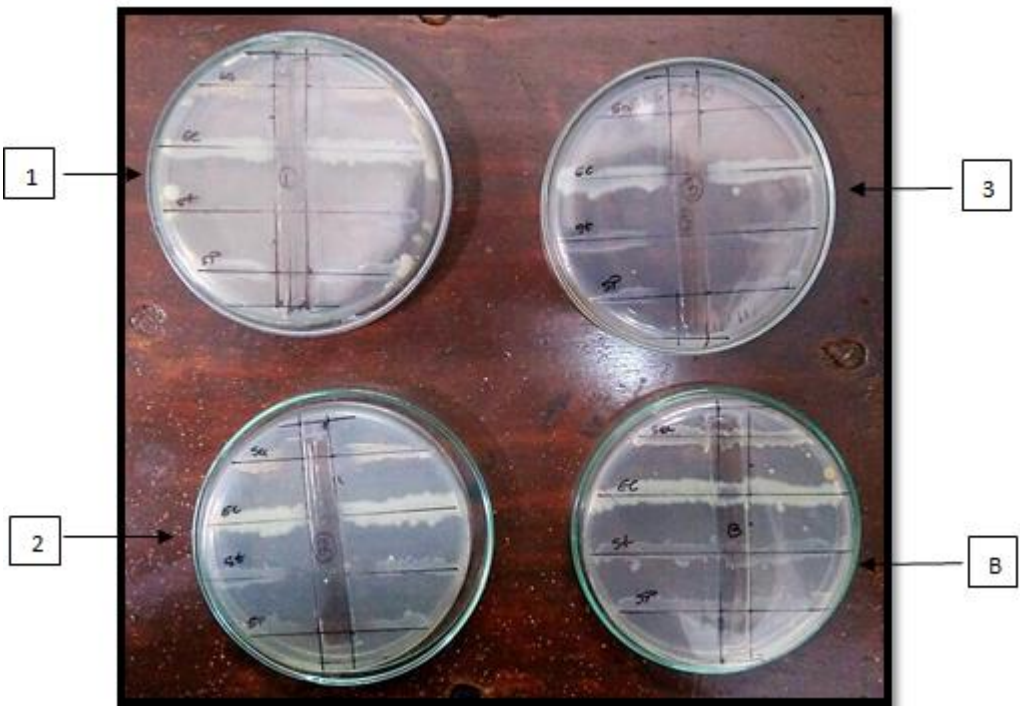


Image 1: The evaluation of antibacterial activity is done by Ditch Plate Method with three replicates (1, 2, 3) and a control test (B)

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