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Cashewnut Shell Oil as Effective Antibacterial Compound and its Effect in *Staphylococcus aureus*

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Abstract: ***Cashewnut shell oil has antibacterial and antifungal activity. It is a corrosive oil which has many medicinal applications. It is used against athletics foot in rural places. The purpose of this research is to analyze the Minimum Inhibitory Concentration of Cashewnut shell oil against Staphylococcus aureus. Cashewnut shell oil was extracted with Acetone, ethanol and petroleum ether were tested against Staphylococcus aureus for antibacterial activity.. The MIC ranges from 150 g/mL -0.15 g/mL of the extracts and it determined by using Broth dilution method. The MIC ranges from 150 g/mL -0.15 g/mL The Concentration of Cashewnut shell oil of about 9.4 µg/mL of* acetone*, 4.7 µg/mL of* ethanol and *9.4 µg/mL of* petroleum ether *showed good activity against* S. aureus*. The oil exhibit antibacterial activity against the pathogen even at low concentration. The present research thus suggests the value of the Shell oil in the treatment of various skin infections.***

Keywords: Minimum Inhibitory Concentration, Cashewnut Shell oil, broth dilution method

1. Introduction

Crude plant extracts were used traditionally in the form of infusion, decoction, tincture or herbal extract to treat various diseases, including infectious diseases. It contain phytochemicals such as phenolic acids, flavonoids, tannins, lignin, and other small compounds[(1,2)]. These compounds are necessary for various health-related effects such as antibacterial, antifungal, antimutagenic, anticarcinogenic, antithrombotic and vasodilatory activities [(3)]. A wide number of phytochemicals are present in plants that inhibit bacterial pathogens [(4)]. Herbal drugs were relatively safer and cheaper than the synthetic alternatives [(5)].

Cashew nut shell oil has approximately 90% anacardic acid ($C_{22}H_{32}O_3$) and 10% cardol ($C_{32}H_{27}O_4$) [(6,7)]. Anacardic acid from the CNSO is inhibitory to *Staphylococcus aureus* [(8)]. From the cashew nut shells, lower quantities of hydroxyl alkylphenols were obtained [(9)]. Anacardic acid showed more potent antibacterial activity except for molds[(10)]. CNSO ethanol extract showed high inhibitory zone against *Staphylococcus aureus*, *Enterococcus*, *Escherichia coli* and *Salmonella*[(11)].

Staphylococcus aureus become resistant to various commonly used antibiotics due to indiscriminate use of antibiotics. *Staphylococcus* resistance to penicillin is induced by penicillinase, an enzyme which breaks down the β-lactam ring of the penicillin. First report on penicillin resistant strain of *Staphylococcus* was published in 1945[(12)]. Penicillinase resistant penicillins are able to resist degradation by Staphylococcal penicillinase still used as first line treatment.

Antimicrobial resistances of bacterial pathogens play a vital role in the treatment of bacterial diseases. Determination of MIC of antibiotics for bacteria plays a major role in the determination of antibiotic resistance in bacteria. In this study, the Cashew nut Shell oil was extracted in acetone, ethanol and petroleum ether solvents. These three extracts were used to examine minimum inhibitory concentration by broth microdilution method against *S. aureus* which cause skin infection.

2. Materials and Methods

Cashewnut shell oil was extracted with acetone, ethanol and petroleum ether (1:3 v/v). The MIC of extracts of Cashewnut shell oil was determined by broth microdilution method as per the procedure described by Eloff, (1998) [(13)] and NCCLS (1997) [(14)]. The 12 test tubes were filled with 0.5mL of Mueller Hinton Broth medium. Sequentially, wells 2-11 were added with 0.5 ml of a mixture of culture medium and the oil extracts were serially diluted to a concentration sequence from 150µl/mL to 0.15µl/ mL. Test tube 1 served as growth control and test tube 12 served as antibiotic control. Tetracycline Hydrochloride (0.1mg/ml) was used as control. The test tubes were incubated for 24h at 37^0C. The resulting turbidity was observed by optical density readings at 660 nm with a Beckman DU-70 UV-Vis Spectrophotometer. The growth inhibition of each extract was determined by using the formula, Percentage of Inhibition = (OD of Control – OD of Test) / (OD of Control) X 100%

3. Results

In this study, the cashewnut shell oil was selected based on the abundant presence of phytochemical constituents and also for their traditional usage in the treatment of skin infections. The ethanolic extract exhibited stronger activity and a much broader spectrum of action than the acetone and petroleum ether extracts (Fig. 1). The lowest concentration (highest dilution) of the plant extract produced no visible growth (no turbidity) in the first 24 h and that was compared with the control tubes and considered as initial MIC.

The results in the Table-1 shows the average minimum inhibitory concentration (MIC) of the Cashewnut Shell oil extracts on *S. aureus* bacteria obtained using microdilution

method. To study the MIC of CNSO the concentration ranges used were from 150 µg/µl to 0.15 µg/µl. The MIC of CNSO that inhibits the growth of bacterial pathogen in the lowest concentration of ethanol extract was 4.7 µg/µl and for acetone and petroleum ether extract it was 2.4 µg/µl.

Table 8: MIC of Cashewnut Shell Oil against *Staphylococcus aureus*

S.no.	Dilution	Concentration	MIC of CNSO		
			Acetone	Ethanol	Petroleum ether
1	150 µg/µl	1:1	-	-	-
2	75 µg/µl	1:2	-	-	-
3	37.5 µg/µl	1:4	-	-	-
4	18.8 µg/µl	1:8	-	-	-
5	9.4 µg/µl	1:16	-	-	-
6	4.7 µg/µl	1:32	-	MIC	-
7	2.4 µg/µl	1:64	MIC	+	MIC
8	1.2 µg/µl	1:128	+	+	+
9	0.6 µg/µl	1:256	+	+	+
10	0.3 µg/µl	1:512	+	+	+
11	0.15 µg/µl	1:1024	+	+	+

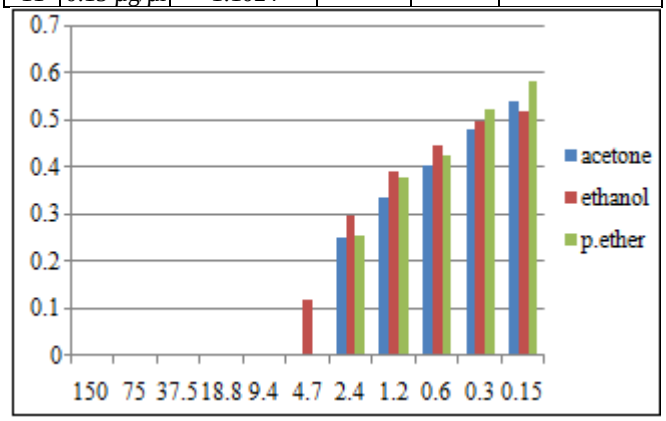

Figure 1: Estimation of MIC of three different solvents of Cashewnut shell oil

4. Discussion

Antimicrobial agents of plant origin have various therapeutic potential. These are highly effective in the treatment of infectious diseases at the same time they have less number of side effects that are often compared with synthetic antimicrobials (Rios and Recio) [17]. Hence, the need of the present research is to evaluate the role of antimicrobial agents of plant origin in the inhibition of the growth of *S. aureus*. The results demonstrate the effectiveness of the oil extracts against the pathogens tested. Isolation and purification of phytochemicals from the Cashewnut Shell may yield significant novel antimicrobials, as plant based antimicrobials have enormous therapeutic potential as they can serve the purpose without any side effects.

5. Conclusion

These results provide a rationalization for the use of these medicinal plants for the treatment and control of various infections in traditional medicine. Further evaluation can be conducted on the extracts to evaluate the safe limit of their dosage and consumption and formulation of the selected medicinal plants. It is necessary to identify the specific phytoconstituents their mode of action and their nontoxic nature in *in vivo* conditions.

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