Antibacterial activity of chewing stick extract (*Azadirachta indica*) against dental pathogens

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(Received: March 06, 2009; Accepted: April 12, 2009)

ABSTRACT

Neem (Azadirachta indica) is a large evergreen tree, in the Mahogany family. Neem contains large number of chemical compound that has a long history of use in both traditional Indian medicine and Ayurveda. To study the antibacterial activity of methanolic and distilled water extracts of the Indian neem sticks namely Azadirachta indica present study was investigated at Post Graduate Teaching Department of Microbiology, Rashtrasant Tukdoji Maharaj Nagpur University, Nagpur in the year 2007-08. The extract was reconstituted in methanol and distilled water in three different concentrations and further process is carried out by using these neem extract. Results from this study revealed that the antibacterial activities of the tested chewing sticks vary among themselves and are target-microbe specific. Major antibacterial activity was detected against Escherichia coli followed by Staphylococcus aureus at 100% concentration, by using methanolic extract. Lactobacillus species recorded least antibacterial activity at all concentrations of methanolic extract. Where as no zone was observed against Pseudomonas species in aqueous extract of sticks. Among the different concentrations of extract 100 percent extract gives maximum antibacterial activity as compare to 50 and 25 per cent in both methanolic and aqueous extract. The present study concluded that Methanolic extract of neem chewing sticks is more effective than the Antibiotics which are used during oral infection. The uses of neem chewing sticks were proved to be very effective treatment in maintaining oral hygiene among rural Asians. This indicates that, neem extract is effectively used to kill infectious oral pathogens.

Key words: Antibacterial activity, Azadirachta indica, methanolic and aqueous extract

INTRODUCTION

Neem (Azadirachta indica) is an aboriginal tree found in tropical and sub-tropical countries like Burma & India. Neem is a large evergreen tree Azadirachta indica, in the mahogany family Neem has been declared non-toxic to humans and every part of the tree is used as an active ingredient for different purposes. Neem is a compound that has a long history of use in both traditional Indian medicine and ayurveda.. In India neem is sometimes called "The Village Pharmacy". Over 100 pharmacologically active substances have been identified in this plant, and it has many traditional applications. Utilization of non-timber forest product (NTFP) is gaining importance in the tropical world because of their commercial importance to the host community. Chewing sticks are important NTFP

widely used for dental cleaning. The choice of chewing sticks to be used in most cases depends on its cleansing action of the teeth; the therapeutic value, or preferred taste or flavour. Neem is used to manufacture a number of natural medicines, which do not have any side effects other uses Antiseptic, Antiviral, Antifungal, Antiparasitic and Antiinflammatory. Azadirachtin is the important chemical compound present in almost all parts of neem tree but, high amount is found in seeds.

MATERIAL AND METHODS

The experiment was conducted at Post Graduate Teaching Department of Microbiology, R.T.M. Nagpur University, Nagpur. The experiment involved one of the type *Azadirachta indica* chewing sticks with two different concentrations of extracts (methanolic and aqueous) tests on four human pathogenic bacteria. The chewing sticks of *Neem tree* for the experiment were obtained from the Department of Botany, Nagpur University, campus Nagpur. All the test microorganisms used in the study were obtained from Medical lab of P.G.T. Department of microbiology, Nagpur except *Lactobacillus* species (Isolated from curd) and all the cultures were maintained on nutrient agar slants (Himedia, Mumbai) stored at 4°C until further use.

Extract preparation

Initially the stems of *Azadirachta indica* were well dried and then powdered. The fine powder obtained was dissolved in methanol (100%, 50%, and 25%) and the mixture was left as such for 3-5 days. Then the mixture was filtered through Whatt man No.1 filter paper and the extract was allowed to dry to get semi-solid mass. The extract was reconstituted in methanol and distilled water in three different concentrations (100%, 50%, 25%)) and used for further process. The aqueous extract was also prepared by procedure mentioned above. Isolation of test organism was done by preparing selective media for each clinical isolates and streak the organism on their selective media.

Antibacterial sensitivity test

The antibacterial sensitivity test was carried using antibiotic discs of Sparfloxacin, Erythromycin, Rifampicin, Clindamycin and Ofloxacin on test microorganisms (*Lactobacillus sp., Staphylococcus aureus, Pseudomonas aeruginosa* and *Escherichia coli*). The plates were incubated at 37°c for 14-19 hours for examination. The inhibition zone was measured and expressed in millimeter.

RESULTS AND DISCUSSION

The antibacterial activity of Methanolic extract of *Azadirachta indica* against dental pathogens was presented in Table 1. The methanolic stem extract of *Azadirachta indica* found to express remarkable antibacterial activity against all the test organisms tested. The highest zone of inhibition of 29mm was detected against *Escherichia coli, while* the lowest zone of inhibition of 10mm was recorded against *Lactobacillus species* in aqueous extract of chewing sticks whereas, no inhibition zone was observed against *Pseudomonas aeruginosa* in aqueous extract of chewing stick.

Organisms	Different concentrations of extract				
	100%	50%	25%		
Lactobacillus sp.	16mm	15mm	12mm		
S.aureus	27mm	23mm	19mm		
E.coli	29mm	23mm	21mm		
P.aeruginosa	19mm	17mm	12mm		

 Table 1: Antibacterial activity of Methanolic extract

 of Azadirachta indica against dental pathogens

 Table 2: Antibacterial activity of Aqueous extract (in distilled water) of Azadirachta indica against dental pathogens

Organisms	Different concentrations of extract						
	100%	50%	25%				
Lactobacillus spp.	15mm	12mm	10mm				
S.aureus	21mm	20mm	16mm				
E.coli	27mm	24mm	20mm				
Ps.aeruginosa	NZ	NZ	NZ				

In the present investigation, to find out the antibacterial activity of extract of chewing stick Azadirachta indica that is neem stick used for dental problems. Olurunke Taiws *et.al.* (1999) suggests that, the chewing sticks are very effective in controlling dental problems. Nimbin, Nimbidin, Nimbolin and Azadirachtin were the important chemical constituent of Azadirachta indica stem. The stem that is the chewing sticks is useful as the dental Analgesic, Carminative, Stimulant, Aromatic and Antiseptic. Kokate *et.al.*It is Stimulant, Antiinfection, Disinfectant and Insecticidal as well as good effective for human disorders like oral infection, skin infection and stomach problems as many, extract is very effectively and efficiently used for dental care.

From the study it has been concluded that the methanolic extract of neem chewing stick is more effective than the antibiotics used against dental pathogens. Though the aqueous extract of neem was effective against *Lactobacillus sp., Staphylococcus aureus* and *Escherichia coli,* no inhibitory action was recorded on *Pseudomonas aeruginosa.* And the medicinal plant extract and their products shows comparatively less side effect or allergic reactions in patients as compared to different antibiotics, therefore research is going on with these herbal plants. The finding provides scope for plant material that is Azadirachta indica stem (neem chewing sticks) for its medicinal uses.

Actions of different antibiotics were also observed. *Staphylococcus aureus* gives sensitive zones against all antibiotics (Ofloxacin, Sparfloxacin, Erythromycin, Rifampcin, and Clindamycin). Where as *Lactobacilus sp.* And *Psedumonas aeruginosa* were resistant. This indicates that the above used antibiotics were not effective against *Lactobacilus*

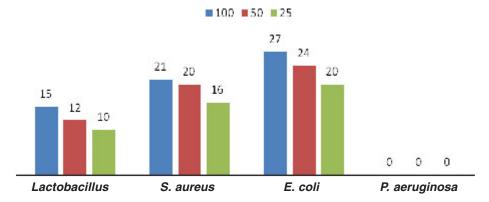
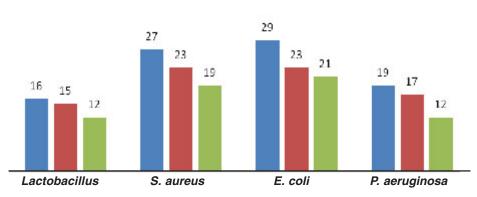


Fig. 1: Antibacterial activity of Aqueous extract (in distilled water) of Azadirachta indica against dental pathogens



■ 100 ■ 50 ■ 25

Fig. 2: Antibacterial activity of Methanolic extract of A. indica against dental pathogens

S. No.	Antibiotics	Staphylococcus aureus		Lactobacillus sp.		E.coli		Pseudomonas aeruginosa	
		Zones	Nature	Zones	Nature	Zones	Nature	Zones	Nature
1	Oflaxacin	23 mm	Sensitive	NZ	Resistant	29 mm	Sensitive	NZ	Resistant
2	Erythromycin	25 mm	Sensitive	NZ	Resistant	12 mm	Resistant	NZ	Resistant
3	Sparfloxacin	30 mm	Sensitive	NZ	Resistant	30 mm	Sensitive	NZ	Resistant
4	Clindamycin	24 mm	Sensitive	NZ	Resistant	10 mm	Sensitive	NZ	Resistant
5	Rifampicin	28 mm	Sensitive	NZ	Resistant	11 mm	Resistant	NZ	Resistant

 Table 3: Action of different antibiotics on Staphylococcus aureus,

 Lactobacillus sp., E.coli and Pseudomonas aeruginosa

and *Pseudomonus aeruginosa* and *E. coli* gives sensitive zones against Ofloxacin, Clindamycin and Sparfloxacin.

Thus, the present study concluded that, the methanolic extract of neem stem is more effective than the antibiotics which were used against dental pathogens. Extract in distill water is also effective against all test organism expect *Pseudomonas species*. So, it can be used directly in making useful products like medicines, ointments, creams, mouth fresheners etc. which are effective and safe to use. By using these herbal plants skin disorder and other diseases can be cured up to particular level.

Medicinal plants extract and other product shows comparatively less side effect or allergic reaction in patients as compared to different antibiotics therefore research is going on these herbal plants. The finding provides scope for plant material i.e. *Azadirachta Indica* stem for its medicinal uses.

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