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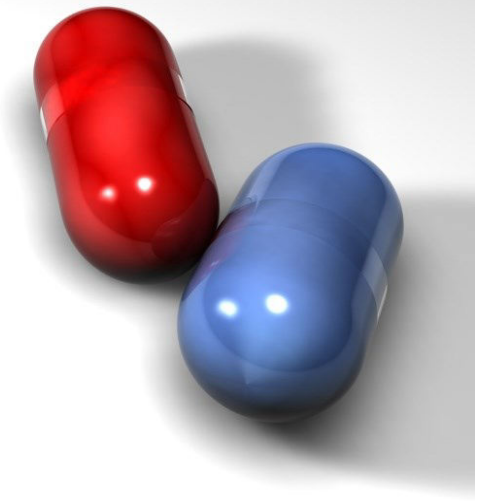

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Letter to the Editor

Effect of neem oil on some pathogenic bacteria

Sir,

Herbs are used for thousands of centuries by many cultures for their medicinal values. Herbal treatment is very popular because it is easily available, cheap and less toxic. *Azadirachta indica* (neem) is a herbal plant widely distributed in our subcontinent during all seasons. Each part of neem tree has some medicinal property. Neem leave, bark extracts and neem oil are commonly used for therapeutic purpose (Tewari, 1992). Neem oil suppresses several species of pathogenic bacteria such as *S. aureus* and *S. typhosa*, all strains of *M. tuberculosis* (Chaurasia and Jain, 1978; Rao et al., 1986). The growth of *S. paratyphi* and *V. cholerae* was inhibited (Rao, 2005). Efficacy of NIM-76, a spermicidal fraction from neem oil was investigated for its antimicrobial action against certain bacteria, fungi and poliovirus as compared to whole neem oil. This shows that NIM-76 has a potent broad spectrum antimicrobial activity (SaiRam, 2000). Available antimicrobial agents can control the infection but they are expensive and rapid emergence of anti-microbial resistance. Neem may be used for its easy availability and significant effect against bacteria. The neem tree is still regarded as 'village dispensary'.

Neem oil extraction was carried out with petroleum ether (bp 60-80°C). It was removed from filtrate by using ordinary distillation at 70°C, the trace amount of solvent remained in the oil was removed by putting the oil in a round bottom flask and placed on a water bath for 20 hours at 60-70°C in rotary vacuum evaporator. After extraction the neem oil was diluted with different amount of liquid paraffin to obtain different dilution of extracts. Individual bacterial strains in pure state (as single colony isolate) was obtained from the Microbiology Department of Dhaka Medical College. The growth of test organism in each dilution of neem oil was examined and compared with controls by matching their turbidity. The clear preparation was considered as no growth. The lowest dilution at which bacteria were inhibited as judged by lack of turbidity was considered as minimum inhibit-tory concentration (MIC). A dilution of neem oil was prepared at their different MIC and poured to the disc. In 'Agar disc diffusion method', the disc containing MIC of neem oil and 4th generation cephalosporin cefepime (30 µg/ disc) were placed on the plate with a sterile forcep and each of them was slightly pressed against agar surface. The diameter of zone of inhibition denoted the relative susceptibility to a particular antimicrobial agent which was detected by the formation of a clean zone around the disc. The diameter of zone of inhibition was measured in millimeter on the under surface of the Petri dish using a transparent scale.

Neem oil was prepared by steam distillation process and its effect against *S. aureus, S. typhi, E. coli* and *P. aeruginosa* was examined by detection of MIC by using 'broth dilution method' and by detection of bacterial

Table I

Diameter of zone of inhibition by different MICs of neem oil and cefepime against test bacteria

	Name of bacteria with average diameter of zone of inhibition							
	Staphylococcus aureus		*Salmonella typhi*		*Escherichia coli*		*Pseudomonas aeruginosa*	
	Disc potency	Zone of inhibition	Disc potency	Zone of inhibition	Disc potency	Zone of inhibition	Disc potency	Zone of inhibition
Neem oil	1:32 dilution	19 mm	1:16 dilution	17.5 mm	1:32 dilution	19.5 mm	1:8 dilution	17 mm
Cefepime	30 mg/ disc	30 mm	30 mg/ disc	26 mm	30 mg/ disc	23.5 mm	30 mg/ disc	21 mm

susceptibility by 'Agar disc diffusion method.' The MIC against *S. aureus, S. typhi, E. coli* and *P. aeruginosa* was at 1:32, 1:16, 1:32 and 1:8 dilution. The average diameter of zone of inhibition against *S. aureus* with neem oil was 19 mm whereas it was 30 mm with cefepime. *S. typhi, E. coli and P. aeruginosa* exhibited zone of inhibition (Table I). Among all test bacteria *S. aureus* had lowest MIC. *In vitro* antibacterial activity of neem oil showed 92% susceptibility against *P. aeruginosa, S. pyogenes, E. coli, Proteus* group and *K. aerugenes*. The MICs varying between ¼ to 1/64 dilution. Inhibitory zones of 13-30 mm were obtained with 65.5% strains while 26.5% strains showed zones of 8-12 mm.

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