

The Inhibiting Effect of Aqueous *Azadirachta indica* (Neem) Extract Upon Bacterial Properties Influencing *in vitro* Plaque Formation

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Abstract:

The purpose of this investigation was to examine the inhibitory effects of aqueous extracts derived from the bark-containing sticks (Neem stick) of *Azadirachta indica* upon bacterial aggregation, growth, adhesion to hydroxyapatite, and production of insoluble glucan, which may affect *in vitro* plaque formation. Neem stick extracts were screened for minimal bacterial growth inhibition (MIC) against a panel of streptococci by means of a broth dilution assay. Initial bacterial attachment was quantified by the measurement of the adhesion of ³H-labeled *Streptococcus sanguis* to saliva-conditioned synthetic hydroxyapatite. The effect of the Neem stick extract upon insoluble glucan synthesis was measured by the uptake of radiolabeled glucose from ¹⁴C-sucrose. Aggregating activity of the Neem stick extracts upon a panel of streptococci was also examined. No inhibition of bacterial growth was observed among the streptococcal strains tested in the presence of ≤ 320 $\mu\text{g/mL}$ of the Neem stick extract. The pre-treatment of *S. sanguis* with the Neem stick extract or the gallotannin-enriched extract from *Melaphis chinensis* at 250 $\mu\text{g/mL}$ resulted in a significant inhibition of the bacterial adhesion to saliva-conditioned hydroxyapatite. Pre-treatment of saliva-conditioned hydroxyapatite with the Neem stick or gallotannin-rich extract prior to exposure to bacteria yielded significant reductions in bacterial adhesion. The Neem stick extract and the gallotannin-enriched extract from *Melaphis chinensis* inhibited insoluble glucan synthesis. Incubation of oral streptococci with the Neem stick extract resulted in a microscopically observable bacterial aggregation. These data suggest that Neem stick extract can reduce the ability of some streptococci to colonize tooth surfaces.