## Antimicrobial and Antibiofilm Properties of Fatty Acids Against Streptococcus Mutans

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Abstract : Planktonic bacteria can form biofilms which are microbial aggregates embedded within a matrix of extracellular polymeric substances (EPS). They can be found attached to abiotic or biotic surfaces. Biofilms are responsible for oral diseases such as dental caries, gingivitis and the progression of periodontal disease. Biofilms can resist 500 to 1000 times the concentration of biocides and antibiotics used to kill planktonic bacteria. Biofilm development on oral surfaces involves four stages, initial attachment, early development, maturation and dispersal of planktonic cells. The Minimum Inhibitory Concentration (MIC) was determined using a range of saturated and unsaturated fatty acids using the resazurin assay, followed by serial dilution and spot plating on BHI agar plates to establish the Minimum Bactericidal Concentration (MBC). Log reduction of bacteria was also evaluated for each fatty acid. The Minimum Biofilm Inhibition Concentration (MBIC) was determined using crystal violet assay in 96 well plates on forming and pre-formed S. mutans biofilms using BHI supplemented with 1% sucrose. Saturated medium-chain fatty acids Octanoic (C8.0), Decanoic (C10.0) and Undecanoic acid (C11.0) do not display strong antibiofilm properties; however, Lauric (C12.0) and Myristic (C14.0) display moderate antibiofilm properties with 97.83% and 97.5% biofilm inhibition with 1000 µM respectively. Monounsaturated, Oleic acid (C18.1) and polyunsaturated large chain fatty acids, Linoleic acid (C18.2) display potent antibiofilm properties with biofilm inhibition of 99.73% at 125 µM and 100% at 65.5 μM, respectively. Long-chain polyunsaturated Omega-3 fatty acids α-Linoleic (C18.3), Eicosapentaenoic Acid (EPA) (C20.5), Docosahexaenoic Acid (DHA) (C22.6) have displayed strong antibiofilm efficacy from concentrations ranging from 31.25-250µg/ml. DHA is the most promising antibiofilm agent with an MBIC of 99.73% with 15.625µg/ml. This may be due to the presence of six double bonds and the structural orientation of the fatty acid. To conclude, fatty acids displaying the most antimicrobial activity appear to be medium or long-chain unsaturated fatty acids containing one or more double bonds. Most promising agents include Omega-3-fatty acids Linoleic,  $\alpha$ -Linoleic, EPA and DHA, as well as Omega-9 fatty acid Oleic acid. These results indicate that fatty acids have the potential to be used as antimicrobials and antibiofilm agents against S. mutans. Future work involves further screening of the most potent fatty acids against a range of bacteria, including Gram-positive and Gram-negative oral pathogens. Future work will involve incorporating the most effective fatty acids onto dental implant devices to prevent biofilm formation.

Keywords : antibiofilm, biofilm, fatty acids, S. mutans

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