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Interpersonal Variation of Salivary Microbiota Using Denaturing Gradient Gel Electrophoresis

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Abstract : The aim of this study was to characterize bacterial population and yeasts in saliva by Polymerase chain reaction followed by denaturing gradient gel electrophoresis (PCR-DGGE) and measure yeast levels by culture. PCR-DGGE was performed to identify oral bacteria and yeasts in 24 saliva samples. DNA was extracted and used to generate DNA amplicons of the V2–V3 hypervariable region of the bacterial 16S rDNA gene using PCR. Further universal primers targeting the large subunit rDNA gene (25S-28S) of fungi were used to amplify yeasts present in human saliva. Resulting PCR products were subjected to denaturing gradient gel electrophoresis using Universal mutation detection system. DGGE bands were extracted and sequenced using Sanger method. A potential relationship was evaluated between groups of bacteria identified by cluster analysis of DGGE fingerprints with the yeast levels and with their diversity. Significant interpersonal variation of salivary microbiome was observed. Cluster and principal component analysis of the bacterial DGGE patterns yielded three significant major clusters, and outliers. Seventeen of the 24 (71%) saliva samples were yeast positive going up to 10^3 cfu/mL. Predominately, C. albicans, and six other species of yeast were detected. The presence, amount and species of yeast showed no clear relationship to the bacterial clusters. Microbial community in saliva showed a significant variation between individuals. A lack of association between yeasts and the bacterial fingerprints in saliva suggests the significant ecological person-specific independence in highly complex oral biofilm systems under normal oral conditions.

Keywords : bacteria, denaturing gradient gel electrophoresis, oral biofilm, yeasts
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