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Genotypic Identification of Oral Bacteria Using 16S rRNA in Children with and without Early Childhood Caries in Kelantan, Malaysia

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Abstract : Caries is the most common childhood disease which develops due to disturbances in the physiological equilibrium in the dental plaque resulting in demineralization of tooth structures. Plaque and dentine samples were collected from three different tooth surfaces representing caries progression (intact, over carious lesion and dentine) in children with early childhood caries (ECC, n=36). In caries free (CF) children, plaque samples were collected from sound tooth surfaces at baseline and after one year (n=12). The genomic DNA was extracted from all samples and subjected to 16S rRNA PCR amplification. The end products were cloned into pCR®2.1-TOPO® Vector. Five randomly selected positive clones collected from each surface were sent for sequencing. Identification of the bacterial clones was performed using BLAST against GenBank database. In the ECC group, the frequency of Lactobacillus sp. detected was significantly higher in the dentine surface ($p = 0.031$) than over the cavitated lesion. The highest frequency of bacteria detected in the intact surfaces was Fusobacterium nucleatum subsp. polymorphum (33.3%) while Streptococcus mutans was detected over the carious lesions and dentine surfaces at a frequency of 33.3% and 52.7% respectively. Fusobacterium nucleatum subsp. polymorphum was also found to be highest in the CF group (41.6%). Follow up at the end of one year showed that the frequency of Corynebacterium matruchotii detected was highest in those who remained caries free (16.6%), while Porphyromonas catoniae was highest in those who developed caries (25%). In conclusion, Streptococcus mutans and Porphyromonas catoniae are strongly associated with caries progression, while Lactobacillus sp. is restricted to deep carious lesions. Fusobacterium nucleatum subsp. polymorphum and Corynebacterium matruchotii may play a role in sustaining the healthy equilibrium in the dental plaque. These identified bacteria show promise as potential biomarkers in diagnosis which could help in the management of dental caries in children.

Keywords : early childhood caries, genotypic identification, oral bacteria, 16S rRNA
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