

TITLE: PREVALENCE AND GENE EXPRESSION OF *LACTOBACILLUS CASEI* GROUP IN ACTIVE AND ARRESTED DENTINE CARIOUS IN CHILDREN WITH EARLY CHILDHOOD CARIES

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ABSTRACT:

Lactobacillus is part of the oral microbiota during the first years of children and it is believed to be pioneer in caries progress, especially in dentin. The *Lactobacillus casei* group, which is formed by *L. casei*, *L. paracasei*, and *L. rhamnosus*, is composed by bacteria strongly associated with the development of dental caries. This study aimed to investigate the prevalence of *L. casei* group and *L. paracasei* in active and arrested dentin carious lesions of children with early childhood caries (ECC) and to examine the expression profile of selected *L. casei* group genes. Dentine samples were collected from 17 active and 13 arrested carious lesions that were diagnosed in pre-school children aged 2-5 years. Total RNA was extracted from the dentine samples and reverse transcription-quantitative real-time PCR (RT-qPCR) analyses were performed for quantification of *L. casei* group and *L. paracasei* and for analyses of the expression of *L. casei* group genes associated with bacterial survival (*spxB*) and virulence (*spaC*, *spaE* and *wzB*). Expression of the tested genes was detected in both types of carious dentine. Our findings revealed that *L. casei* group is part of the viable microbial community in active and arrested dentine carious lesions. *L. casei* group, *spaC* and *wzB* genes were equally presents in active and arrested lesions ($p > 0.05$), while *L. paracasei* ($p = 0.05$) and *spaE* gene ($p = 0.02$) were more prevalent in active dentin lesions. The pili establish a primary contact in adhesive processes explaining the ability to persist in the host. This structure can be encode by *spaC* and *spaE* genes. Functionally, the *spaC* pili can binds to collagen protein and is credited as a key adhesive factor. The production of exopolysaccharides (EPS) is a key factor in the adherence of dental biofilm. *WzB* is a phosphotyrosine protein phosphatase that has been shown to regulate EPS biosynthesis. The *spxB* ($p = 0.02$) gene was expressed at higher level in arrested as compared to active lesions. *SpxB* encodes for pyruvate oxidase, the increase expression of the *spxB*, in arrested lesion, could be a way to produce energy and allow bacterial growth during unfavourable environmental conditions for microbial growth. In addition, the activity of pyruvate oxidase, which produces hydrogen peroxide, could be a mechanism for interspecies competition.

Keywords: Early Childhood Caries; Dentine Caries; Gene Expression; *Lactobacillus casei* group.

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