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Can insoluble polysaccharide concentration in dental plaque, sugar exposure and cariogenic microorganisms predict early childhood caries? A follow-up study

T.M. Parisotto [a], *R. Stipp* [b], *L.K.A. Rodrigues* [c], *R.O. Mattos-Graner* [b], *L.S. Costa* [d], *M. Nobre-dos-Santos* [b,*]

[a] *Laboratory of Microbiology and Molecular Biology, Sao Francisco University Dental School, Bragança Paulista, SP, Brazil*
[b] *Department of Pediatric Dentistry, Piracicaba Dental School, University of Campinas, Brazil*
[c] *Department of Operative Dentistry, Faculty of Pharmacy, Dentistry and Nursing, Federal University of Ceará, Fortaleza, CE, Brazil*
[d] *Federal University of Rio de Janeiro State, Rio de Janeiro, RJ, Brazil*

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ABSTRACT

Background: Insoluble polysaccharide (IP) has been associated with caries prevalence in young children. However, the power of IP to predict ECC needs to be demonstrated.
Aims: To assess the relationships between early childhood caries (ECC) and extracellular insoluble polysaccharides (IP) in dental plaque, sugar exposure and cariogenic microorganisms.
Design: Visible plaque on maxillary incisors was recorded, followed by caries diagnosis in 65 preschoolers (3–4 years) at baseline and after 1 year. Plaque was collected for mutans streptococci (MS), total microorganism (TM) and lactobacilli (LB) enumerations in selective media, as well as for IP analysis, which was later assessed by colorimetry. Sugar/sucrose exposure was assessed by a diet chart.
Results: Positive correlations were found among the prevalence of caries and MS, TM, LB, solid sucrose and visible dental plaque. Additionally, children with IP concentrations in dental plaque higher than 2.36 μg/mg (odds ratio-OR = 6.8), with visible plaque on maxillary incisors (OR = 4.3), harbouring LB (OR = 13) and exposed to solid sugar more than twice/day (OR = 5) showed higher risk of developing caries ($p < 0.05$).
Conclusion: Extracellular insoluble polysaccharides, solid sugar/sucrose, visible dental plaque and cariogenic microorganisms could predict caries development, partially explaining the ECC pattern.

* *Corresponding author at*: Av. Limeira 901, Piracicaba, SP 13414-903, Brazil. Tel.: +55 19 21065290; fax: +55 19 21065218.
E-mail address: nobre@fop.unicamp.br (M. Nobre-dos-Santos).

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1. Introduction

Early childhood caries (ECC) is a multifactorial disease characterized by a process involving mineral transfer from the tooth to the surrounding environment in children younger than 6 years.[1] The dynamic process of dental caries is intimately related to dental plaque if left undisturbed, which covers tooth surfaces as a tightly adherent layer consisting of bacterial, inorganic and organic components; the so-called biofilm.[2]

With respect to the organic components/matrix, the role played by glucans should be highlighted. Glucosyltransferases, enzymes of bacteria such as *Streptococcus mutans* (*S. mutans*), synthesize glucans, which are extracellular polysaccharides formed from sucrose.[3] Depending on the predominant linkage type alpha 1–3 or alpha 1–6, the glucan or extracellular polysaccharide can be water-insoluble (termed mutan) or soluble (termed dextrans), respectively. The matrix formed mostly by the insoluble glucans, contributes to *S. mutans* tenacious adhesion and accumulation. The ability of *S. mutans* to adhere and accumulate on dental surfaces via glucan production is a very significant virulence factor, which could lead to an increased number of infected tooth sites.[4]

Mutans streptococci are the most common pathogens behind ECC, as demonstrated by many studies involving these bacteria.[5–7] However, recent evidences support that there is a wide diversity of species in ECC, including *Slackia exigua* and *Scardovia wiggsiae*, the latter being a candidate as a newly recognized caries pathogen.[8,9] Young children with ECC are usually colonized by mutans streptococci and often have inappropriate feeding practices, such as frequent consumption of carbohydrates and sweetened fluids.[5,10] Inappropriate feeding habits with a high frequency of sugar consumption provide sucrose, the specific substrate for glucan production. IP has been associated with ECC prevalence in young children. However, the power of IP to predict ECC needs to be demonstrated. Moreover, studying caries development after an identification of IP concentration in dental plaque, enable us to consider the child's response to this factor during the disease process. This way we are not assuming that a certain factor preceded caries development. Thus, the purpose of this study was to assess the relationships between ECC and dental plaque IP, sugar exposure and cariogenic microorganisms.

2. Material and methods

2.1. *Ethical considerations*

This study was independently reviewed and approved by the Ethical Committee in Research of Piracicaba Dental School/ UNICAMP (Protocols #015/2006 and #017/2008) and has been conducted in full accordance with ethical principles, including the World Medical Association Declaration of Helsinki. The preschools granted permission for the study and the children's parents signed a written positive consent form.

2.2. *Sample*

We based the sample power calculation (85%; $\alpha = 5\%$) on the study previously performed by our group[6] which used similar methodology and found 8.2 μg/mg of difference and a standard deviation of 19% in the extracellular insoluble polysaccharides concentration in dental plaque when caries free and pit and fissure caries children were compared. Since this was a 1-year longitudinal study, the calculated number (52) was increased by 27% to compensate for subject drop-out rate.

A comfort sample of 65 children, from both genders (girls – 51%, boys – 49%) and aged 3–4 year olds, took part in this study. These children were from low socioeconomic backgrounds and attended public preschools in the urban and fluoridated (0.5–0.8 ppm) area of Itatiba, state of São Paulo, Brazil. Children were excluded from the study if they refused to cooperate with the clinical examinations or if they had systemic diseases, severe fluorosis or dental hypoplasia. In the preschools, the children ate the same meals, stayed a minimum of 4 h/day and had their teeth brushed at least once a day with fluoride-containing dentifrice.

The children were submitted to clinical examinations for caries diagnosis; plaque collection for IP and microbiological analyses; dental plaque assessments of the maxillary incisors; and sugar exposure investigation. In order to determine changes in the prevalence of caries of the studied population, only the clinical examinations for caries diagnosis were repeated after 1 year. Thus, according to the changes (follow up scores – baseline scores) in the prevalence of caries, the children were assigned into 3 groups:

1. Caries arrestment (AR): children who had early carious lesions that arrested (decayed, missing, or filled surfaces-dmfs at baseline = 6.18, dmfs at follow up = 4.27, dmfs change = −1.9) (n = 11).
2. Caries free (CF): children who were always free of caries and never showed early carious lesions (ECL), cavitations or fillings (dmfs at baseline = 0, dmfs at follow up = 0, dmfs change = 0) (n = 19).
3. Caries active (CA): children with caries who continued to develop carious lesions, being it cavitations or ECL (dmfs at baseline = 10.26, dmfs at follow up = 14.86, dmfs increment = 4.6) (n = 35).

2.3. *Clinically visible dental plaque recording*

The presence or absence of clinically visible plaque on the maxillary incisors was recorded under artificial light, with the child lying on a table. This was performed before teeth cleaning for the clinical examinations.

2.4. *Clinical examination*

- Early childhood caries diagnosis was made according to the World Health Organization's criteria, with an additional measurement of early carious lesions. For further details on how active early carious lesions were diagnosed please refer to Parisotto et al.[7] Thus, in the study, active white chalky spot lesions were also considered caries.