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Differentiating fluoride reservoirs in dental biofilm: methodology development

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Abstract
Fluoride is found in dental biofilm bound to bacteria cells or or as precipitated minerals. The aim of this study was to test de dissolution rate of CaF2 in Fluoride or Calcium rich solutions. The results show that in 10 washes, CaF2 solubilizes partially in Ca or F-rich solutions.

Key words:
Dental biofilm, fluoride, calcium.

Introduction

Fluoride (F) is found in dental biofilm bound to bacteria cells or as precipitated minerals (e.g. calcium fluoride, CaF_2), and determining the amount of F in each pool is relevant and challenging. Aiming to develop a method to differentiate these reservoirs in F-rich biofilm samples, in this preliminary study we tested the dissolution rate of CaF_2 in calcium (Ca) or F-rich solutions.

Results and Discussion

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CaF_2 was precipitated in microcentrifuge tubes (n=18) from supersaturated 0.05 M PIPES buffer, pH 7.0, containing 10 mM F and 10 mM Ca. After centrifugation, supernatants were collected to determine Ca and F concentration. The CaF_2 precipitated (ppt) in the tubes (n=6/group) was: I – saved as control (no further extraction), II – extracted with PIPES buffer containing 5.0 mM Ca (mean concentration found in the supernatants) or III – extracted with PIPES buffer containing 0.88 mM F (means concentration found in the supernatants). Extractions consisted of five 30-s washes under vortex with 2 mL of the treatment solutions at 37ºC, followed by five 1-min washes. Between each wash, tubes were centrifuged and the supernatant collected to check F (in group II) or Ca (in group III) release, which were, respectively, 0.29±0.02 and 0.14±0.01 mM, indicating CaF_2 dissolution. After the 10 washes, CaF_2 remaining in the tubes were dissolved with 0.5 M HCl for 3 h at room temperature to determine the amount (mg) of CaF_2 not extracted by the washes made. The amount (mg) of CaF_2 not extracted in groups II (0.18±0.04) and III (0.13±0.03) differed ($p<0.01$, ANOVA and Tukey test) from that found in group I (0.38±0.01).

Conclusions

The results show that in 10 washes, CaF_2 dissolves partially, 52 and 66% in solutions containing saturating concentrations of either Ca or F, respectively.

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