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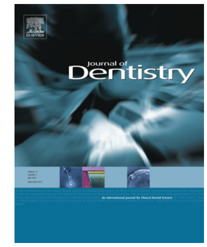

Calcium lactate pre-rinse increased fluoride protection against enamel erosion in a randomized controlled in situ trial

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ABSTRACT

Objectives: This in situ trial study was designed to evaluate whether calcium (Ca) pre-rinse would increase the fluoride (F) rinse protection against enamel erosion.

Methods: Fifteen volunteers participated in this split-mouth, 3-phase, crossover design study wearing a palatal appliance containing four sterilized bovine enamel slabs, for 10 days. In the 1st phase, five participants followed protocol A: daily rinse with a Ca lactate (CaL, 150 mmol/L, 1 min), followed by F (NaF 12 mmol/L, 1 min). Other five participants followed protocol B: daily rinse only with F, while the remainders followed protocol C: no rinse (negative control). Appliances were removed from the mouth and one side of the palatal appliance was exposed to a daily erosive challenge (0.05 M citric acid, 90 s); the other side served as control (deionized water – no erosion). In the 2nd phase volunteers were crossed over to other protocol and in the 3rd phase volunteers received the remaining protocol not yet assigned. Specimens were evaluated for surface loss using an optical profilometer.

Results: Repeated-measures three-way ANOVA ($p = 0.009$) and Tukey's test showed that CaL pre-rinse followed by NaF rinse significantly decreased surface loss of enamel when performed prior to an erosive challenge in comparison with the condition in which NaF only was used.

Conclusions: Pre-rinse with CaL may increase the protection exerted by NaF against erosive wear.

1. Introduction

The scientific foundation for dental erosion prevention and control rests on two major components: risk assessment and preventive measures.[1] The former refers to the understanding of the causes of erosive lesions, leading to the identification of chemical, biological and/or behavioural risk factors.[1,2] The latter relates to the efficacy of interventions for the management of erosive tissue loss.[1,3]

Although literature indicates incomplete evidence for the efficacy of measures currently used for erosion prevention,

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among the proposed approaches is the use of fluoridated products which protect tooth surfaces with mineral precipitates, especially of CaF_2-like mineral (KOH-soluble fluoride).[4,5] In fact, to a certain extent, such CaF_2-like mineral precipitates can mechanically protect the surface against erosive attacks.[1,6] However, these precipitates may be dissolved as the pH drops.[1,7–9]

To produce more acid resistant mineral precipitates, numerous studies have been devoted to assess the role of fluoride preparations containing polyvalent metal cations or organic compounds.[10–20] In addition, some papers deal with strategies to increase the fluoride bioavailability in the oral cavity. In this regard, it has already been shown that calcium (Ca) rinse used prior to fluoride rinse increases the formation of Ca-mediated deposits in saliva and may enhance the cariostatic effect of fluoride products.[21–23] In fact, Ca pre-rinse induced a 5.5 times increase in salivary fluoride over a NaF solution.[23] Although possibilities are that the same benefit may be exerted under erosive conditions, to the authors knowledge there is only one in vitro study that tested the protective effect of calcium and fluoride rinses on erosion.[24] In the quoted paper, a calcium lactate pre-rinse showed some promising results by enhancing the sodium fluoride protection against subsequent erosive challenges when dentine is under laboratory conditions that simulated reduced salivary flow.

Considering that acquired pellicle influences the retention of KOH-soluble fluoride *in situ*[15] and that saliva appears to be the reservoir of CaF_2-like mineral as a result of the use of Ca pre-rinse and fluoride rinse,[21,23] it seems important to move a step forward and test the potential of Ca rinse followed by NaF rinse in situ. To do so, this study was designed as a split-mouth, crossover trial to evaluate whether Ca rinse given before F rinse would increase enamel protection against erosive episodes. The null hypothesis tested was that there would be no significant difference in surface loss of enamel whether prior to erosive challenges Ca rinses followed by NaF rinses or NaF rinses only were performed.

2. Materials and methods

2.1. Study outline

This interventional study had a split-mouth, three-phase, crossover design with fifteen volunteers, who worn palatal devices loaded with bovine enamel slabs. The independent variables were *Rinsing* and *Erosion*. The former had three levels: CaL followed by NaF; NaF only; none (as control), while the latter had two levels: present and absent (as control).

According to a random allocation, at the first phase five volunteers were assigned to follow protocol A: rinse with a CaL solution, followed by NaF solution, while other five participants followed protocol B: rinse only with NaF solution and the remaining five volunteers followed protocol C: no rinse (negative control). In the second phase volunteers were crossed over to other protocol and in the third phase volunteers received the remaining protocol not yet assigned. In each phase, after had following one of the protocols, volunteers were instructed to immerse one side of the palatal appliance in a citric acid (HCa) solution while the other side was immersed in deionized (DI) water (negative control – no erosion), characterizing the split-mouth, crossover design mentioned above. Thus there existed six experimental conditions ($n = 15$, with duplicate samples), as follows: (1) CaL + NaF + HCa; (2) NaF + HCa; (3) no rinse + HCa; (4) CaL + NaF + DI water; (5) NaF + DI water; (6) no rinse + DI water. Sample size was determined based on previous experiments in our laboratory involving similar in situ studies so we could have at least 80% power to detect significant difference. Fig. 1 shows the CONSORT flow chart of the progress through the phases of the study. The dependent variable was surface loss, measured in μm, respectively.

2.2. Participants and ethical aspects

Fifteen participants (four men and 11 women) ranging from 21 to 32 years of age volunteered for this study after it was approved by the local Institutional Review Board (#2011/0148). The study was carried out by a principal investigator who was responsible for the observance of the protocol, and who, together with the other four investigators, performed the clinical and technical procedures.

All volunteers were students or staff working in the São Leopoldo Mandic Institute and Dental Research Center. Informed consent was obtained from all participants prior to their entry into this study. Volunteers were eligible if they exhibited good oral hygiene condition, no tooth wear lesions, no caries activity, no periodontal disease nor reflux disease and showed mean stimulated saliva flow rate ≥ 0.7 mL/min. Ineligible were subjects wearing removable orthodontic appliances and those taking medicines, as were those with systemic diseases. Smokers and patients suffering from alcoholism were not recruited. Women who were pregnant or breastfeeding were not included as well. Enrollment of the participants was made by the principal investigator.

2.3. Preparation of specimens

Sixty bovine incisors, stored in 0.1% aqueous thymol solution, with no coronal cracks or enamel malformations were used in this study. Teeth were scraped of any remaining soft tissues, polished with pumice slurry, and sectioned at the cemento-enamel junction, using a low-speed water-cooled diamond saw (Isomet 1000, Buehler Ltd., Lake Bluff, IL, USA). Each tooth was cut mesiodistally and buccolingually to obtain four enamel slabs measuring 3 mm × 3 mm × 2 mm. Sectioned pieces were mounted on acrylic rods with sticky wax and ground/polished using a Ecomet 250 grinder-polisher (Buehler Ltd., Lake Bluff, IL, USA) coupled with a Buehler Automet 250 head (Buehler Ltd., Lake Bluff, IL, USA). Slabs were wet flattened with aluminium oxide abrasive papers (600- and 1200-grit) and polished with a 0.3-μm alumina suspension Alfa Micropolish (Buehler Ltd., Lake Bluff, IL, USA). Slabs were then cleansed ultrasonically in DI water for 10 min to remove any residues of the polishing procedure.

2.4. Assessment of flatness and baseline microhardness of slabs and sterilization

Slabs were checked for flatness of the polished enamel surface using an optical profilometer (Proscan 2000, Scantron, Venture