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In vitro evaluation of the erosive potential of orange juice modified by food additives in enamel and dentine

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ABSTRACT

Objectives: To evaluate the erosive potential of orange juice modified by food additives in enamel and dentine.

Methods: Calcium lactate pentahydrate (CLP), xanthan gum (XG), sodium linear polyphosphate (LPP), sodium pyrophosphate tetrabasic (PP), sodium tripolyphosphate (STP) and some of their combinations were added to an orange juice. Pure orange juice and a calcium-modified juice were used as negative (C−) and positive (C+) controls, respectively. In phase 1, 15 modified orange juices were tested for erosive potential using pH-stat analysis. In phase 2, the additives alone and the combination with good results in phase 1 and in previous studies (CLP + LPP) were tested in an erosion–remineralization cycling model. In phase 3, the erosion and remineralization episodes were studied independently. Enamel was analysed by surface microhardness (SMH) and profilometry, whilst dentine by profilometry.

Results: In phase 1, reduction of the erosive potential was observed for all additives and their combinations, except XG alone. In phase 2, no detectable enamel loss was observed when CLP, LPP and CLP + LPP were added to the juice. XG, STP and PP had enamel loss similar to C− ($p > 0.05$). Amongst additives, the combination CLP + LPP showed the highest SMH values followed by CLP ($p < 0.05$). All the other groups presented SMH values similar to C− ($p > 0.05$). For dentine, only CLP + LPP lead to surface loss values lower than C− ($p < 0.05$). In phase 3, CLP, LPP and CLP + LPP seemed to protect against erosion; whilst none of the tested compounds seemed to interfere with the remineralization process.

Conclusions: CLP and LPP reduced erosion on enamel and this effect was enhanced by their combination. For dentine, only the combination CLP + LPP reduced erosion.

1. Introduction

The prevalence and incidence of dental erosion has increased over the last few decades,[1,2] and studies have related this fact to the increase of acidic soft drinks consumption worldwide.[3] Some important chemical aspects can modulate their potential to cause dental erosion, including pH,[4] titratable acidity,[5] type of acid,[6] buffer capacity,[7] chelating properties,[5] and concentration of calcium, phosphates and fluoride.[7] It is

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known that specific modifications on these parameters may lead to a reduction on the erosive potential of a given acidic beverage.[8]

A commonly investigated modification has been the use of additives, mostly salts containing calcium and/or phosphate ions.[7,9–12] They act based on the common ion effect, where the driving force for dental surface dissolution can be decreased by the saturated state of the drink with respect to the calcium and phosphate ions.[8] However, the addition of phosphates alone does not seem to be as effective as calcium.[13,14] The addition of food polymers has also been investigated and they have shown ability to reduce erosion due to their possible adsorption to the dental surfaces, leading to the formation of an acid-protective layer. This layer could reduce the exchange of H^+ and of calcium and phosphate ions between the hydroxyapatite and the solution.[15] The negative side of using food polymers could be that they also have mineralization-inhibiting properties, interfering with possible remineralization of the eroded dental substrate.[16]

In this study we aimed to investigate the modification of the erosive potential of an orange juice by the addition of salts of calcium and phosphate as well as of food polymers, either alone or in combination. Orange juice was chosen due to its acidic nature, well documented erosive potential[17–19] and widespread and worldwide consumption. The study hypothesis was that the additives, combined or alone, would be able to reduce dental erosion development, by either preventing the demineralization or enhancing the remineralization.

2. Materials and methods

2.1. Experimental design

This study was carried out in 3 phases. In the first, five substances and their combinations (total of 15 formulations) were added to a commercially available orange juice and the erosive potential of these solutions was compared with the pH-stat as a screening method, tested in triplicate. In the second phase, six solutions were tested, comprising the 5 additives alone and the combination that showed the best protective action in phase 1, as well as positive and negative controls. In this phase both human enamel and root dentine specimens (*n* = 10) were tested, using an erosion–remineralization cycling model. In the third phase, we further investigated the mechanism of action of the additives by breaking down the cycling model in two independent tests: demineralization only and remineralization. Bovine enamel was the substrate tested (*n* = 5). A single factor, completely randomized experimental design was used for all the tests. The response variable for phase 1 was the volume (ml) of the titrant (0.1 N HCl). For phases 2 and 3, the response variables were surface loss (μm) measured by optical profilometry, and/or surface microhardness (SMH) determined by the Knoop hardness number.

2.2. Phase 1

In this phase, five food-approved substances were added alone or in combination to a commercial available orange juice (Minute Maid Original®, The Coca-Cola Company, Atlanta, GA, USA), creating the experimental groups showed in Table 1. The additives chosen for this study were: calcium lactate pentahydrate (CLP) (Fisher Scientific Pittsburgh, PA, USA); sodium polyphosphate with an average chain length of 25 phosphate units, linear structure (LPP) (Calgon 696, Thermos Inc., Cheshire, UK), which will be referred as 'sodium polyphosphate' during the paper; sodium tripolyphosphate (STP) (Sigma–Aldrich Co., St. Louis, MO, USA); sodium pyrophosphate tetrabasic (PP) (Sigma Aldrich Co., USA) and xanthan gum (XG) (Keltrol R; CP Kelco UK, Leatherhead, UK). The amounts used were based on previous publications.[10–12,15,20] The juice without additives was the negative control (C−) and a commercially available calcium-modified juice (Minute Maid Calcium®, The Coca-Cola Company, Atlanta, GA, USA), which has approximately 40 mmol/l of calcium[21] as calcium lactate,

Table 1 – Experimental groups and their codes, additives, concentrations used, means (SD) of the pH, volume of titrant (in ml) needed in the pH-stat method and hydroxyapatite dissolution (in mg).

Group code	Additives (g/l)					pH	Titrant volume	Hydroxyapatite
	CLP	XG	LPP	STP	PP	Mean (SD)	Mean (SD)	Dissolution mean (SD)
C−						3.83 (0.02)	1.23 (0.08)	15.50 (0.001)
CLP	3.1					3.83 (0.01)	0.46 (0.03)	5.80 (0.000)
XG		0.2				3.82 (0.02)	1.39 (0.01)	17.41 (0.000)
LPP			0.2			3.83 (0.00)	0.20 (0.01)	2.45 (0.000)
STP				0.2		3.82 (0.01)	0.73 (0.04)	9.15 (0.000)
PP					0.2	3.81 (0.01)	0.75 (0.08)	9.47 (0.001)
CLP + XG	3.1	0.2				3.82 (0.01)	0.63 (0.06)	7.87 (0.001)
CLP + LPP	3.1		0.2			3.83 (0.01)	0.03 (0.03)	0.43 (0.000)
CLP + STP	3.1			0.2		3.83 (0.00)	0.13 (0.10)	1.57 (0.001)
CLP + PP	3.1				0.2	3.82 (0.02)	0.16 (0.13)	2.03 (0.002)
XG + LPP			0.2			3.81 (0.00)	0.27 (0.02)	3.40 (0.000)
XG + STP				0.2		3.83 (0.01)	0.82 (0.01)	10.27 (0.000)
XG + PP					0.2	3.83 (0.01)	0.77 (0.01)	9.63 (0.000)
CLP + XG + LPP	3.1	0.2	0.2			3.83 (0.01)	0.04 (0.07)	0.52 (0.001)
CLP + XG + STP	3.1	0.2		0.2		3.82 (0.02)	0.07 (0.06)	0.88 (0.001)
CLP + XG + PP	3.1	0.2			0.2	3.83 (0.00)	0.01 (0.01)	0.09 (0.000)
C+						4.11 (0.01)	0.00 (0.00)	0.00 (0.000)