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A new in situ model to study erosive enamel wear, a clinical pilot study



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ABSTRACT

Objectives: To develop an in situ model for erosive wear research which allows for more clinically relevant exposure parameters than other in situ models and to show tooth site-specific erosive wear effect of an acid challenge of orange juice on enamel. *Methods:* This pilot study included 6 edentulous volunteers wearing full dentures with 13 embedded enamel samples The study consisted of two control runs: habitual diet only for 30 days, and two experimental runs: habitual diet plus 125 ml orange juice four times per day (consumed over 4 m). In the first experimental run subjects were instructed to take the drink in their mouth and promptly swallow it. In the second experimental run subjects take the drink in their mouth and hold each 25 ml for 30 s, moving it around their mouth before swallowing. Sample enamel surface loss was measured using non-contact surface profilometry. *Results:* Drinking of orange juice additional to the habitual diet, caused significant but low increased erosive wear at buccal, palatal and lingual specimens. Significantly higher levels of tissue loss were found

erosive wear at buccal, palatal and lingual specimens. Significantly higher levels of tissue loss were found on all surfaces in the swishing experiment but molars (especially occlusal specimens in mandibular molars), palatal specimens sited in upper anterior teeth and lingual specimens in lower anterior teeth were most affected.

Conclusion: The model showed clinically relevant patterns of erosive tooth wear as seen clinically in erosive wear patients.

Clinical significance: The model could have many applications to study clinically erosive wear for specific diets or consumption patterns and to test preventive measures.

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1. Introduction

In situ models have been used extensively in dental erosive wear research to study the potential erosive effect of acidic beverages, mainly soft drinks, but also fruit juices [1–6]. Most, however, have considerable limitations in modeling the clinical situation. Appliances with the samples are usually worn only part-time, not during normal eating and drinking or during the night. Acid exposures are usually short and/or performed extra-orally, so as not to endanger the natural dentition of the volunteers. Also, in *in situ* studies volunteers wear an intra-oral appliance where enamel samples are placed either at the roof of the palate or in the buccal vestibule of the lower jaw. However, erosive-abrasive tooth wear can be observed on all tooth surfaces but is most common on

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occlusal and facial surfaces of maxillary and mandibular teeth and on palatal surfaces of the maxillary anterior teeth [7]. Ideally, sample placement would have been at those locations to mimic the clinical situation most closely.

The aim of the study was to develop an in situ model for erosive wear research which allows for more clinically relevant exposure circumstances than most in situ models and to demonstrate the site-specific erosive wear effect of an acid challenge of orange juice on enamel, as seen in erosive wear patients. Two hypotheses were proposed for the study: 1) daily drinking of orange juice in addition to a daily diet will show site-specific erosive wear as seen in erosive wear patients and 2) swishing of orange juice, compared to drinking, would cause progressive erosive wear distribution over tooth sites.

2. Materials en methods

This study consisted of a four phase, single center clinical trial involving subjects attending a prosthetic dental clinic in Arnhem, the Netherlands and who met the following inclusion criteria: aged

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between 40 and 75 years, edentulous in the upper and lower jaw and wearing full dentures, healthy with no relevant medical status, which could interfere with the conduct of the study. All participants had to have an unstimulated and stimulated whole mouth saliva flow rate of >0.25 and >0.75 ml/m, respectively. Potential subjects who met any of the following criteria were excluded from participation: use of medication which could influence salivary flow, self reported or observed grinding of clenching habit, symptoms of gastro-oesophageal reflux (heartburn, regurgitation), vegetarian diet and abnormal drinking habits (e.g. swishing or holding of drinks before swallowing). Six subjects were included in the study. All volunteers wear a full denture in the upper jaw and an implant-supported full lower denture. As there was no information at all available on the rate of erosive enamel loss under the proposed circumstances, a sample size calculation was impossible.

The study protocol was approved by Radboud University's research committee (CEOM: NL: 47343.091.14). Subjects were given verbal and written information concerning the study and gave and signed consent to participate. An incentive of 150 Euros per each month of participation was given.

2.1. Design of full dentures and sample preparation

Before the start of the study, for each volunteer copies of subjects' full dentures were made by a dental technician. After enamel sample preparation, the samples were embedded in PMMA in metal holders and inserted into different (tooth) surfaces of the subjects' upper and lower denture.

2.2. Enamel specimen preparation

Sound human teeth (extracted third molars) were collected. One specimen was cut from the buccal surface of each tooth. Enamel specimens were cut with a diamond blade (Buehler diamond wafering blade nr.11-4244) with an average dimension of $3 \times 3 \times 2$ mm and were sterilized in ethylene oxide gas (Wimac Kliniekdiensten BV, Rotterdam, The Netherlands).

After sterilization the enamel specimens were embedded in stainless steel rings in PMMA (Auto Plast, Candulor AG, Wangen, Switzerland; see Fig. 1). After embedding, the specimens were ground with SiC paper with the ring serving as reference area. The enamel surface was polished as little as possible, just enough to create a flat surface to profile. Each specimen was inspected for surface defects with a stereomicroscope (magnification $10\times$) and rejected if cracked. The thickness of the enamel samples was between 500 and 800 micrometers after polishing.

In the subjects' upper and lower denture 6 and 7 enamel specimens, respectively, were inserted into occlusal, buccal, palatal and lingual tooth surfaces (Fig. 2). The specimen locations were in posterior sextants (upper and lower (pre)molar area of the full dentures), one enamel specimen in occlusal position, one specimen sited on the buccal surface of the upper molars (16–17 or 26–27, respectively) and one enamel specimen on the lingual surface of the lower molars (36–37 or 46–47) and one specimen located on the buccal surface of the 34 and 44. In anterior sextants two specimens (palatal surface of the 11 and 21) were positioned and one sample in the middle of the lingual surface of the 31 and 41.

During the study subjects had to wear their dentures for 24 h per day, except during cleaning. Twice a day (in the morning and in the evening) the appliances and specimens were cleaned by gently brushing (soft tooth brush) with fluoride toothpaste (Prodent Softmint^R).

2.3. Experimental design

The design consisted of 4 30-days runs including one control run before the start of experimental run 1 (drinking of orange juice) and one before the start of experimental run 2 (swishing of orange juice). In the control runs the subjects only followed their habitual diet. During the two experimental runs the volunteers additionally consumed 125 ml orange juice four times per day. After each run enamel surface loss measurements were performed. After control measurements the enamel samples were polished until they again had a flat surface. After the first control/ experimental run combination new enamel samples were inserted.

For 30 days, 125 ml orange juice (brand: Aldi^R; pH 3.7) 4 times a day was taken during 4 m at 09.00, 11.00, 13.00 and 15.00 h, at refrigerator temperature $(4-5 \,^{\circ}\text{C})$. Each subject was issued a measuring beaker and stopwatch and asked to drink in 25 ml mouthfuls. The 125 ml orange juice was consumed in 5 mouthfuls at 1 m intervals (at start, after 1, 2, 3 and 4 m respectively). In the first experimental run subjects were instructed to take 25 ml of the drink in their mouth and promptly swallow it. In the second experimental run subjects were instructed to take the drink in their mouth and hold each 25 ml mouthful for 30 s, moving it around their mouth in a rinsing motion before swallowing. Subjects were always allowed to spit out the drink instead of swallowing it, in order to prevent gastro-intestinal problems.

The subjects were asked to fill in a questionnaire about their diet. They had to report on the daily intake of dietary products during each run. Overall consumption of acidic food and beverages per day were calculated by counting the number of potential erosive foods and drinks. A pH <5,5 was used to consider the potential erosiveness of a product. Tables of the pH and/acidity of foods and food products from the FDA/CFSAN [8] were used, as well as tables from the website Gezondheidsnet [9]. For determining pHs of drinks (waters, juices, soft drinks, alcoholic drinks) several sources were used. [10,11] Milk-based products were considered non erosive irrespective of their pH.



Fig. 1. Enamel specimens embedded in stainless steel reference ring. The height mean difference was calculated from the reference areas A and B compared to enamel area C.

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