

-  Ana Cristina Corso Scapinello<sup>1</sup>
-  Iana Lamadrid Aurélio<sup>2</sup>
-  Carine Weber Pires<sup>1</sup>

<sup>1</sup> Centro Universitário da Serra Gaúcha, Curso de Odontologia, Caxias do Sul, RS, Brasil.

<sup>2</sup> Universidade Federal de Santa Maria, Programa de Pós-graduação em Ciências Odontológicas, Santa Maria, RS, Brasil.

#### Correspondence

Carine Weber Pires

[cwpodonto@gmail.com](mailto:cwpodonto@gmail.com)

## Erosive potential of beverages consumed by Brazilian children: an in vitro study in bovine primary teeth

*Potencial erosivo de bebidas consumidas por crianças brasileiras: um estudo in vitro em dentes decíduos bovinos*

#### Abstract

**Objective:** The aim of this study was to verify the in vitro erosive potential of beverages frequently consumed by Brazilian children, including cola based soft drinks, regular and zero, industrialized box and powdered juices, chocolate milk drink and tap water. **Methods:** 42 sound primary bovine teeth were prepared and flat surfaces of enamel were obtained and distributed into six groups. The pH of each beverage was measured. The specimens were submitted to erosive challenge with the tested products for 28 consecutive days. The surface roughness was evaluated before and after erosion-remineralization cycling in profilometer. Data were analyzed by the paired T test (groups with normal distribution) and the Wilcoxon test (groups with non-normal distribution). The 5% level of significance was adopted for all analyzes. **Results:** In general, the surface roughness values of the primary bovine dental enamel increased from the initial values. For the groups water ( $p = 0.134$ ), chocolate ( $p = 0.283$ ) and juices

( $p = 0.091$ ), the roughness did not present a significant intragroup difference before and after erosive challenge. Both, regular cola soft drink ( $p = 0.043$ ) and sugar zero cola soft drink ( $p = 0.049$ ) showed significant erosive effect on deciduous bovine teeth. **Conclusions:** Regular cola soft drink and sugar zero cola soft drink significantly influence the roughness of deciduous bovine dental enamel, demonstrating a greater erosive potential among the beverages most consumed by Brazilian children.

**Keywords:** Dental Erosion. Diet. Tooth, Deciduous. Child.

### Resumo

**Objetivo:** Verificar o potencial erosivo in vitro de bebidas frequentemente consumidas por crianças brasileiras, como refrigerantes à base de cola regular e zero, sucos de caixa industrializados e em pó, bebida láctea achocolatada e água torneiral. **Métodos:** Foram preparados 42 dentes bovinos decíduos hígidos, de modo que superfícies planas de esmalte fossem obtidas, e distribuídos em seis grupos. O pH de cada bebida foi medido. Os espécimes foram submetidos a desafio erosivo com os produtos testados durante 28 dias consecutivos. A rugosidade superficial foi avaliada antes e depois da ciclagem erosão-remineralização em perfilômetro. Os dados foram analisados pelo teste t pareado (grupos com distribuição normal) e pelo teste de Wilcoxon (grupos com distribuição não normal). O nível de significância adotado para todas as análises foi de 5%. **Resultados:** De modo geral, os valores de rugosidade superficial do esmalte dental bovino decíduo aumentaram em relação aos valores iniciais. Para os grupos água ( $p=0,134$ ), achocolatado ( $p=0,283$ ) e sucos ( $p=0,091$ ), a rugosidade não apresentou diferença significativa intragrupo antes e após desafio erosivo. O refrigerante a base de cola convencional ( $p=0,043$ ) e o refrigerante a base de cola sem açúcar ( $p=0,049$ ) demonstraram efeito erosivo significativo em dentes bovinos decíduos. **Conclusões:** Os refrigerantes a base de cola convencional e sem açúcar influenciam significativamente na rugosidade do esmalte dental bovino decíduo, demonstrando maior potencial erosivo entre as bebidas mais consumidas por crianças brasileiras.

**Palavras-chave:** Erosão Dentária. Dieta. Dente Decíduo. Criança.

## INTRODUCTION

Biocorrosion or dental erosion is the gradual, progressive and irreversible loss of hard dental structure caused by frequent contact with acid substances without bacterial involvement, a characteristic that differentiates it from dental cavities.<sup>1,2</sup> The prevalence of dental erosion increases linearly with age, ranging from 5% to 28% in three-year-olds and from 6% to 35% in five-year-olds.<sup>3</sup> Thus, erosion in primary dentition can predict the same condition in permanent dentition.<sup>4</sup>

Deciduous teeth are less resistant to erosion than permanent teeth.<sup>5</sup> In addition, children have lower salivary flow rates and, consequently, lower saliva buffering capacity, which results in less neutralization and elimination of the acids that cause erosion, contributing to increased susceptibility to erosion.<sup>6</sup> As a result of erosion, these individuals may have hypersensitivity, loss of tooth contour, shortening of the upper incisors, difficulties in feeding, aesthetic impairment and, in extreme cases, loss of vertical dimension, pulp exposure or abscesses, since in deciduous teeth the enamel and dentin thickness is smaller and the pulp chamber is larger than the crown, with prominent pulp horns, increasing the chance of pulp exposure.<sup>3</sup>

The etiology of dental erosion is multifactorial.<sup>4</sup> The consumption of acidic beverages and foods plays an important role in the development of erosion.<sup>7</sup> Changes in the lifestyle of the population, increased consumption of industrialized products, such as artificial powdered fruit juices and soft drinks that contain acids in their composition, the replacement of water intake by natural juices in tropical countries, the greater concern with healthy dietary habits stimulating the consumption of fruits and natural juices, influence the high incidence of dental erosion.<sup>8-10</sup>

In Brazilian preschool and school children, sweetened beverages are the most commonly consumed, especially chocolate-based beverages, processed fruit juices and soft drinks.<sup>11</sup> However, in addition to the sugar present in these beverages, their erosive potential should be considered. The objective of this study was to verify the in vitro erosive potential of beverages frequently consumed by Brazilian children.

## METHODS

### Sample selection

Newly extracted deciduous, healthy bovine teeth were selected without lesions, cracks, fractures or opacities. After selection, the teeth were cleaned and stored in 0.5% chloramine T solution at 4°C for 30 days, with weekly changes of storage solution.

The sample size was determined from a previous study,<sup>12</sup> and from the results obtained, the calculation was performed, stipulating a beta error of 0.80 and an alpha error of 0.05. Thus, it was stipulated that at least six teeth per group would be sufficient to achieve the statistically necessary power for statistically significant differences between the groups to be obtained. The sample consisted of 42 deciduous bovine teeth.

**Preparation of teeth and experimental groups**

After the roots were removed with a diamond disk (Isomet, Buehler, Germany), the buccal and palatal faces of the deciduous bovine tooth crowns were worn out in Politriz (Ecomet 3 - Buehler Ltd, Lake Bluf, IL, USA), with #600 and #1500 grain silicon carbide water sandpaper, under cooling, maintaining parallelism between the surfaces, until flat surfaces were obtained.

The sample was randomly divided into six groups (table 1). All groups presented n=7, except for the Chocolate Powder group, which presented a problem when reading a specimen, totaling n=6. The pH of each beverage was measured using tapes (Macherey-Nagel, Duren, Germany) and a calibrated digital pH-meter (Tecnal, São Paulo, SP, Brazil). The control solutions (buffer) used for the measurements were potassium biphtalate (pH 4.00) and disodium phosphate (pH 7.00).

**Table 1.** Characteristics of the evaluated beverages. Caxias do Sul-RS, 2018.

Beverage	(n)	Commercial brand	pH	
			Tape	PhMeter
Regular cola soft drink - Traditional	n=7	1	3.00	2.73
Sugar zero cola soft drink	n=7	1	3.00	3.18
Orange flavored fruit nectar (industrialized box juice)	n=7	2	4.00	3.63
Powdered solution for orange flavored beverages (1L powder juice)	n=7	3	3.00	3.12
Chocolate milk drink	n=6*	4	6.00	5.64
Tap water	n=7	5	6.00	7.00

\*Problems were identified when reading 1 specimen.

**Erosive Challenge**

Before starting the procedure, the samples were cleaned from possible impurities, in distilled water, ultrasonically. The teeth were submitted to an erosion-remineralization cycling model.<sup>13</sup> Each specimen was individually immersed for one minute in 10mL of the tested beverage at room temperature twice a day, for 28 consecutive days, totaling 56 immersion cycles. After each immersion in the test products, each specimen was washed with distilled water and immersed in 10mL of artificial saliva for six hours (between cycles) and 18 hours (between the last and first cycles of the following day). At each cycle, the evaluated products were discarded and the saliva was changed before the first cycle of each day. After the periods of erosion-remineralization, the specimens were washed in distilled water and carefully dried on paper towels.

**Surface roughness evaluation**

The initial surface roughness reading was performed on specimens prior to any treatment. After erosion-remineralization cycling, the final surface roughness reading was performed. At both times, the specimens were taken to the profilometer with a contact type pen (Mitutoyo SJ-410, Kanagawa, Japan), with the following test configurations: λc - 0.8 mm and speed of 0.5 mm/s. The parameter used was arithmetic roughness (Ra), determined by the mean (µm) of three readings, starting from three different positions. The mean of the three readings resulted in the mean roughness of each deciduous enamel surface.

**Statistical Analysis**

The normality of the data was assessed by the Kolmogorov-Smirnov and Shapiro-Wilk tests, and the homogeneity of the variances by the Levene test. For the groups with normal distribution, the paired t-test was performed, while for the groups in which the distribution was not normal, the Wilcoxon test was used. The level of significance adopted for all analyses was 5%. The statistical analysis of roughness values (Ra, µm) was performed with SPSS (Statistical Package for the Social Sciences), version 23.0.

**RESULTS**

The means representing the roughness of the specimens are presented in Tables 2 and 3. In general, the surface roughness values of deciduous bovine dental enamel increased in relation to the initial values. For the groups water (p=0.134), chocolate powder (p=0.283) and juice (p=0.091), the roughness did not show significant intragroup difference before and after the erosive challenge. Regular cola soft drink (p=0.043) and sugar zero cola soft drink (p=0.049) showed significant erosive effect on deciduous bovine teeth.

**Table 2.** Mean and standard deviation of roughness (Ra) before and after erosive challenge, according to paired T test. Caxias do Sul-RS, 2018.

Beverage	Ra Before (±SD)	Ra After (±SD)	P Value
Sugar zero cola soft drink	0.3578 (0.0862) a	0.4184 (0.0495) b	0.049*
Chocolate milk	0.3534 (0.0531) a	0.3814 (0.0460) a	0.283
Water	0.4452 (0.1286) a	0.3910 (0.1548) a	0.134

Means followed by the same lowercase letter on the lines do not differ significantly at 5% probability; \*statistical significance.

**Table 3.** Mean and standard deviation of roughness (Ra) before and after erosive challenge, according to Wilcoxon's test. Caxias do Sul-RS, 2018.

Beverage	Ra Before (±SD)	Ra After (±SD)	P Value
Regular cola soft drink	0.3992 (0.0437) a	0.5014 (0.1338) b	0.043*
Box juice	0.3724 (0.0567) a	0.5030 (0.1915) a	0.091
Powdered solution for orange flavored	0.4262 (0.0757) a	0.5941 (0.3711) a	0.091

Means followed by the same lowercase letter on the lines do not differ significantly at 5% probability; \*statistical significance.

## DISCUSSION

In view of the increased consumption of industrialized products by children, such as soft drinks, artificial powder juices, juices and dairy drinks in boxes, it is essential to assess the erosive potential of these beverages. The present study showed that certain beverages consumed during childhood may represent an erosive challenge for the deciduous enamel, contrasting with a previous study conducted with human deciduous teeth, in which no beverage evaluated available in the market significantly influenced the roughness values.<sup>14</sup>

The acids present in certain beverages are able to demineralize the inorganic matrix that makes up the dental structure when exposed to an inorganic aqueous solution with a pH below 5.<sup>15</sup> However, even in beverages with an acidic pH, such as apple juice (pH 4.0),

these acid components, after an erosion-remineralization cycling in young bovine teeth, have not significantly altered their surface roughness.<sup>15</sup> This is in line with the results found in the present study, in which the box juices (pH 3.63) and powder juices (pH 3.12), despite showing an increase in the roughness values, were not able to significantly alter the enamel surface.

The results obtained for soft drinks show that both regular cola soft drink and sugar zero cola soft drink have a significant erosive effect on deciduous enamel. These beverages showed the lowest pH values or their erosive potential is associated with it, since the solubility of the enamel hydroxyapatite increases as the pH decreases.<sup>16</sup> Although no direct comparison was made between the two, the mean surface roughness value of the enamel after erosion challenge with regular cola soft drink is higher. Previous studies, which also performed profilometry, found that sugar zero cola soft drink resulted in less enamel wear than the regular version.<sup>17,18</sup> This difference may be explained by the fact that sugar-free cola-based soft drinks have a higher pH value and contain aspartame, which may be responsible for lower erosive potential.<sup>17</sup> But both hypotheses need further studies, since these are only hypotheses that do not minimize the results of the present study.

The findings that cola-based beverages significantly alter the enamel roughness and that this did not happen with orange juices is consistent with a previous study, which compared the erosive effect of these beverages in the first minute of exposure to hydroxyapatite crystals.<sup>19</sup> The same study found that the erosive potential of all types of cola beverages was ten times greater than that of orange juice. It can also be suggested that differences in the viscosity of cola soft drink and juices influence these results. More viscous substances facilitate the adhesion of the product to the dental substrate, which is a chemical factor that can influence the biocorrosion process.<sup>13</sup>

The consumption of dairy beverages and yoghurts has a protective effect against dental erosion, due to the amount of calcium and phosphate in its composition, and can control the demineralization of the surface.<sup>20</sup> In the present study, the chocolate milk beverage was tested to deny its erosive potential, since it is one of the most consumed by Brazilian children.<sup>11</sup>

This study has the limitation of being in vitro, not taking into account biological factors present in the oral cavity. However, keeping the specimens in artificial saliva in the intervals of exposure to the beverages had the objective of approaching what occurs in vivo, in which there is the buffer action of saliva, neutralizing the deleterious effects.<sup>21</sup> The use of deciduous bovine dental enamel is justified by the similarity with human enamel and the easy acquisition of this product.<sup>22</sup>

## CONCLUSION

Both cola soft drink significantly influence the roughness of deciduous bovine tooth enamel, demonstrating greater erosive potential among the beverages most consumed by Brazilian children.

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**Contributors**

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