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Light cola drink is less erosive than the regular one: An *in situ/ex vivo* study

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Light cola drink is less erosive than the regular one: an *in situ*/*ex vivo* study

Abstract

Objective: This *in situ/ex vivo* study assessed the erosive potential of a light cola drink when compared to a regular one.

Methods: During 2 experimental 14-days crossover phases, eight volunteers wore palatal devices with 2 human enamel blocks. The groups under study were: group light, erosive challenge with light cola drink and group regular, erosive challenge with regular cola drink. During 14 days, erosive challenges were performed extraorally 3X/day. In each challenge, the device was immersed in 150 ml of light cola (group light) or regular cola (group regular) for 5 minutes. Erosion was analysed by surface profilometry (µm) and surface microhardness change (%SMH). The data were statistically analyzed using paired t test ($p<0.05$).

Results: Group light (0.6 ± 0.2 µm) showed significantly lesser wear than group regular (3.1 ± 1.0 µm). There was no significant difference between the groups for the %SMH (group light -63.9 ± 13.9 and group regular -78.5 ± 12.7).

Conclusions: The data suggest that the light cola drink is less erosive than the regular one.

1. **Introduction**

Erosion is defined as the loss of dental hard tissue resulting from non-bacterial chemical attack usually involving acidic substances.[1,2] The consumption of acidic foodstuffs and beverages plays a major role in the development of erosion.[3] Along with the change of lifestyle through the decades, the total amount and frequency of consumption of acidic foods and drinks have also changed.[4] Soft drink consumption in the USA increased by 300% in 20 years.[5] The increased consumption of acidic products in the presence of other risk factors (behavioural and biological) might increase the risk for dental erosion.[2,4,6]

The potential erosive effect of a soft drink depends on a number of conditions such as pH and buffering capacity, type of acid (pK_a values), *adhesion* of the product to the dental surface, chelating properties and calcium, phosphate and fluoride concentration.[2,7] Since the composition of regular and light drinks is different, especially due to the presence of sweeteners, such as aspartame, in the last ones, it was speculated that they could have a different erosive potential. This study was therefore designed to compare the erosive potential of a light version of a cola drink to its regular version. For this purpose an *in situ/ex vivo* model was chosen, using profilometry as response variable. Percentage of superficial microhardness change was also employed as a complementary response variable.

Keywords: Erosion, Soft drinks, *In situ*, Enamel

2. Material and methods

2.1. Experimental design

This study was approved by the IRB of the Faculty of Dentistry of Bauru, University of São Paulo. Eight adult volunteers with average age of 26.8 yr (range 21-32 yr), and normal stimulated saliva flow rate (> 1 mL/min) took part in this study after signing an informed, written consent. The study involved a randomized complete block design, performed in two *in situ* crossover phases of 14 days, with an interval of one week in between. In each phase, the volunteers wore an acrylic palatal appliance containing two human enamel blocks. The appliance was renewed after the first phase. The factor under evaluation was the version of cola drink (light or regular). The groups under study were: group light, erosive challenge with light cola drink and group regular, erosive challenge with regular cola drink. The response variable was depth of enamel surface wear (µm). Percentage of superficial microhardness change (%SMH) was also employed as a complemental response variable.

2.2. Chemical Properties of the studied beverages

The pH and phosphorus, calcium and fluoride concentrations were analyzed using standard procedures (Light cola- pH:3.0; 13.7 mg Ca/L; 15.5 mg P/L and 0.31 mg F/L–/ regular cola- pH: 2.6; 32.1 mg Ca/L; 18.1 mg P/L and 0.26 mg F/L). The amount of base (NaOH 0.2 M) added to raise the pH to 7.0 was measured using a pH electrode, and both colas were titrated with 0.125 mL.

2.3. Enamel blocks and palatal device preparation

Enamel blocks (4 x 4 mm) were obtained from recently extracted, caries-free, unerupted human third permanent molars, which were stored and sterilized in 2% formaldehyde solution pH 7.0 for 30 days at room temperature.[8] The enamel surface of the blocks was ground flat with water-cooled carborundum discs (320, 600 and 1200 grades of Al_2O_3 papers; Buehler, Lake Bluff, USA) and polished with diamond spray (1µm; Buehler, Lake Bluff, USA), resulting in removal of about 100 µm depth of enamel, which was controlled with a micrometer. A surface Knoop microhardness test was performed (5 indentations in different regions of the block, 25 g, 5 s, HMV-2000; Shimadzu Corporation, Tokyo, Japan) to select 32 human enamel blocks (320-358 KHN).

Custom-made acrylic palatal devices were made with two recessions (6 x 6 x 3 mm). One enamel block was randomly assigned to each of the two recessions and fixed with wax. In order to maintain reference surfaces for the profilometric measurement and microhardness analysis, two layers of nail varnish were applied on half of the blocks' surfaces.[8]

2.4. Intraoral phase

One day before the experimental phases, the device was worn and the blocks were not subjected to erosion to allow the formation of a salivary pellicle.[9] During the following 14 days, erosive challenges were performed extraorally 3 times per day (8hr, 14hr, and 20hr). In each challenge, the device was immersed in a cup containing 150 mL of a freshly opened bottle of the regular or light cola drink (Coca Cola; Companhia Fluminense de Refrigerantes, Porto Real, Brazil), for 5 minutes. Before reinserting the device into the mouth the volunteers were instructed to take one sip of the beverage.

Moreover, the volunteers were instructed to avoid acidic foods and to wear the intraoral devices throughout the intraoral phase of the study. The devices should be removed only for the above-described procedures, during meals and for oral hygiene purposes. Throughout the experimental phase, the volunteers brushed their teeth with fluoridated dentifrice (1,100 ppm F as NaF, pH 6.8; Crest, USA).

2.5. Microhardness and wear analysis

At the end of each experimental phase, the nail varnish over the reference surfaces was carefully removed and the blocks were removed from the device. After that, surface microhardness of the enamel blocks was measured again using a microhardness tester (Shimadzu HMV-2000, Shimadzu Corporation, Japan) with a Knoop diamond under a 25-gram load for 5 seconds. Ten indentations were made on each specimen, five on the previously protected enamel surface (SMH) and five on the experimental areas (SMH_1). The percentage of superficial microhardness change (%SMH) was calculated (%SMH=[SMH_1-SMH]/SMH]*100).

The enamel wear was determined profilometrically in relation to the reference surface (Hommel Tester T 1000, Hommelwerke, Schwenningen, Germany). The tracing parameters were established at Lt: 1.5 mm and Lc: 0.25 mm. Five readings were performed on each block. These profilometric traces were taken by moving the stylus from the reference surface to the exposed surface. The average wear depth of an experimental unit was computed by using the 5 readings: one block x five readings.

2.6. Statistical analysis

The assumptions of equality of variances and normal distribution of errors were checked for the tested response variables. Since the assumptions were satisfied, paired Student's *t* tests (α=0.05) were performed. The software GraphPad Prism 4 version 4.0 for Windows, Graph Pad Software (San Diego, CA, USA) was used.

3. Results

The light cola drink caused a 5-fold decrease in the wear when compared to regular cola drink (group light/$0.6^a \pm 0.2$ µm and group regular/$3.1^b \pm 1.0$ µm) ($p<0.05$). The percentage of superficial microhardness change was not significantly different between the groups, even though the regular cola drink ($-78.5^a \pm 12.7$) presented a higher SMH% than the light cola drink ($-63.9^a \pm 13.9$).

4. Discussion

Several *in vitro* studies have evaluated the erosive potential of different beverages, by using different parameters, such as pH and buffering capacity, type of acid (pK_a values), adhesion of the product to the dental surface, calcium, phosphate and fluoride concentration, dental demineralization and wear.[2,10-13] Some of these studies did not show differences in the erosive potential between the regular and light drink[2,11,12] while others showed that the diet cola drink caused less demineralization than the regular ones.[10,13] In the present study, for the first time, the erosive wear of a light cola drink was compared to that of a regular cola drink using an *in situ* protocol. The light cola drink caused demineralization of tooth surface, which consequently led to reduction of the surface microhardness values, but without a relevant surface wear when compared to the regular cola drink. Thus, the light cola drink did not demineralize the enamel surface in sufficient manner to allow a high loss of enamel surface. For this reason the wear was considered the best response variable to show the effect of the erosive challenge into the enamel and the %SMC should be considered as a complementary data.

This result can be partially explained by the different pH values of the drinks (light cola pH 3.0 and regular cola 2.6). As shown by Hannig et al.,[14] the erosive effects of different acids are pH dependent and vary distinctly between pH 2 and pH 3. This means that even a small decrease of the pH can result in a distinct increase of enamel loss.[14] On the other hand, the drinks presented the same buffering capacity. However this chemical characteristic might be of minor importance in the present study, as the enamel samples were exposed to a large volume of the respective drinks for 5 min only in each erosion cycle. Therefore, the pH of the drink samples might be constant during the exposure time.

In addition, the adhesion of the product to the dental surface and calcium, phosphate and fluoride concentration might influence the development of erosion.[2,12,,15] Ireland et al.,[15] found that regular coke drinks showed a better adhesion to enamel surfaces than light coke drinks. However, it has to be taken into account that the above mentioned study was performed on flat, polished enamel surfaces without the impact of a salivary pellicle.

Finally, it can be hypothesized that this lower erosive potential of the light cola drink is related to the presence of the amino acid phenylalanine, which is provided from the hydrolysis of aspartame in the presence of saliva. The possible mechanism of action of phenylalanine is not known so far. This amino acid could act as a buffer system, increasing the neutralization and buffering the acids from the cola drink. Hung and Hung[16] showed that phenylalanine presented a protective effect against gastric hemorrhagic erosion in acid-irrigated stomachs of rats. Taking into account the results of Hung and Hung[16], another possibility is the formation of an amino acid based layer on dental surfaces. This layer might reduce dental erosion by acting as a diffusion barrier or a perm-selective membrane preventing the direct contact between the acids and tooth surface, such as the salivary pellicle.[17]

The findings of this study demonstrated the importance of studying other parameters, such as amino acids and proteins present in drinks that could interact with components of saliva, resulting in an impact on dental erosion *in vivo*.

5. Conclusions

The data suggest that the light cola drink is less erosive than the regular one. Further studies should be conducted to elucidate if the presence of phenylalanine is the real responsible for the lesser erosive potential of the light cola drink.

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References

1. ten Cate JM, Imfeld T. Dental erosion, summary. *European Journal of Oral Science* 1996;**104**:241-244.

2. Lussi A, Jaeggi T, Zero D. The role of diet in the aetiology of dental erosion. *Caries Research* 2004;**38**:34-44.

3. Johansson AK, Johansson A, Birkhed D, Omar R, Baghdadi S, Khan N, et al. Dental erosion associated with soft-drink consumption in young Saudi men. *Acta Odontológica Scandinavica* 1997;**55**:390-397.

4. Lussi A. Erosive tooth wear – a multifactorial condition of growing concern and increasing knowledge. *Monographs in Oral Sci*ence 2006;**20**:1-8.

5. Calvadini C, Siega-Riz AM, Popkin BM. US adolescent food intake trends from 1965 to 1996. *Archives of Disease in Childhood* 2000;**83**:18-24.

6. Moss SJ. Dental erosion. *International Dental Journal* 1998;**48**:529-539.

7. Larsen MJ, Nyvad R. Enamel erosion by some soft drinks and orange juices relative to their pH, buffering effect and contents of calcium phosphate. *Caries Research* 1999;**33**:81-87.

8. Rios D, Honório HM, Magalhães AC, Delben CAB, Machado MAAM, Silva SMB, et al. Effect of salivary stimulation on erosion of human and bovine enamel subjected or not to subsequent abrasion: an in situ/ ex vivo study. *Caries Research* 2006;**40**:218-230.

9. Hara AT, Turssi CP, Teixiera EC, Serra MC, Cury JA. Abrasive wear on eroded root dentine after different periods of exposure to saliva in situ. *European Journal of Oral Science* 2003;**111**:423-427.

10. Grobler SR, Senekal PJ, Laubscher JA. In vitro demineralization of enamel by orange juice, apple juice, pepsi cola and diet pepsi cola. *Clinical Preventive Dentistry* 1990;**12**:5-9.

11. Jensdottir T, Holbrook P, Nauntofte B, Buchwald C, Bardow A. Immediate erosive potential of cola drinks and orange juices. *Journal of Dental Research* 2006;**85**:226-230.

12. Lussi A, Jaeggi T. Chemical factors. *Monographs in Oral Sci*ence 2006;**20**:77-87.

13. Jain P, Nihill P, Sobkowski J, Agustin MZ. Commercial soft drinks: pH and in vitro dissolution of enamel. *General Dentistry* 2007;**55**:150-154.

14. Hannig C, Hamkens A, Becker K, Attin R, Attin T. Erosive effects of different acids on bovine enamel: release of calcium and phosphate in vitro. *Archives of Oral Biology* 2005;**50**:541-552.

15. Ireland AJ, McGuinness N, Sherriff M. An investigation into the ability of soft drinks to adhere to enamel. *Caries Research* 1995;**29**:470-476.

16. Hung CR, Hung PC. Protective effects of several amino acid-nutrients on gastric hemorrhagic erosions in acid-irrigated stomachs of septic rats. *The Chinese Journal of Physiology* 1999;**42**:161-169.

17. Hara AT, Lussi A, Zero DT. Biological factors. *Monographs in Oral Sci*ence 2006;**20**:88-99.