Assessment of Acid Neutralizing Capacity in Cola-based Drinks and Energy Beverages by Artificial Saliva

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ABSTRACT

Objective: To assess the pH of regular and light cola-based drinks and energy drinks, and examine the acid neutralizing capacity by the addition of artificial saliva.

Materials and methods: Ten packages of each product purchased locally at different stores were evaluated. The pH was measured by a pH-electrode calibrated in standard buffer solutions. To assess the pH neutralizing capacity, 1 ml/min of artificial saliva was added until a pH of 5.5 set as a cutoff point was reached in the tested solution. The data were subjected to analysis of variance (ANOVA), at the 0.05 significance level.

Results: The surveyed beverages had an initial acidic pH, ranging between 2.3 and 3.4. The average amount of saliva required to raise the pH of the cola-based drinks above 5.5 ranged between 6.0 and 6.8 ml. Energy drinks used volumes between 11.3 and 12.5 ml; however, it was not possible to achieve a pH of 5.5.

Conclusion: According to the methodology used, it was concluded that: (1) All beverages analyzed showed an initial acidic pH. (2) There was no statistical difference between the initial pH level and acid neutralization by the addition of artificial saliva in both regular and light drinks. It was not possible to reach the appropriate pH, set as the cutoff point, for the energy drinks.

Keywords: Dental erosion, Beverages, Drinks, Artificial saliva.

Dental erosion can be defined as the chemical removal of minerals from the tooth structure without the participation of bacterial processes, often occurring through the involvement of acidic substances and affect deciduous or permanent teeth.2,3 Usually this lesion initiate in areas where enamel not exist or is less mineralized such as near the gingival margins retracted or areas of defect, and often have to be restored.

It is also possible to observe a relationship between the intake of acidic fatty foods and the development of dental erosion injuries, for which treatment should start with the elimination of the causative agent.4

Erosion is a natural process and can be considered physiological.5 However, in certain circumstances, and in some individuals, tooth wear over time can be considered excessive and therefore, pathological.6 According to Shaw; Smith7 (1998), pathological tooth wear is becoming increasingly common.

This is largely due to the changing eating habits of the population, which includes the proven increase in industrialized drinking beverages with acidic pH in the last two decades.2 As an example, Larsen; Nyvad8 (1999), through an in vitro study, investigated the erosive potential of soft drinks, mineral water and orange juice and compared erosion depth relating to pH and buffering capacity of drinks. They reported that erosion was minimal in drinks with pH above 4.2, but became more evident with pH below 4.0.

To prevent erosion, saliva has been considered the most important biological factor. It works in two ways; first, it has a significant influence on the oral environment acidity for its neutralizing capacity of the pH.9 Second, it has a remineralizing capacity, balancing the demineralization process occurred by ingestion of acidic substances.10,11

The objective of this study was to evaluate the pH of cola-based drinks and energy drinks and examine the
Possibility of the pH neutralization in these beverages through the addition of artificial saliva.

MATERIALS AND METHODS

Two brands of cola-based drinks (light and regular) and two brands of energy drinks (one light and regular and other just regular) were randomly acquired in ten different markets. Just one sample per product was acquired in each market.

The sample groups were divided in seven groups with 10 samples each as follow: regular cola-based drinks 1 (RCD1), regular cola-based drinks 2 (RCD2), light cola-based drinks 1 (LCD1), light cola-based drinks 2 (LCD2), regular energy drinks 1 (RED1), light energy drinks 1 (LED1) and regular energy drinks 2 (RED2).

The pH level of each sample was measured using a pH-meter (Tecnal–Piracicaba, São Paulo state) with electrode (Digimed, São Paulo) previously calibrated using standard buffer solutions at pH 4.0 and pH 7.0, with an uncertainty range of +0.2. After opening each package, the beverage was housed in glass tube, and the initial measurement of the pH level was performed for each sample.

Immediately after the completion of the initial pH measurement, 6 ml of each sample was added in a test tube, remaining in homogenization on a magnetic stirrer (Faalk, Paulo Lopes, Santa Catarina state). Then, 1 ml of artificial saliva per minute was added using a pipette. The pH readings were performed immediately after each addition of saliva. The procedure was repeated until reaching the cutoff point established for the pH or when it reached stability levels, i.e., when it remained constant for five consecutive additions of artificial saliva.

The volume of saliva was estimated based on what was described by Dawes and Kubieniec (2004). They obtained a flow rate of 1 ml/min of saliva secreted under stimulation.

The cutoff point of pH 5.5 was determined based on the study by Cairns et al, which they claimed that at that level enamel begins to demineralize.

Artificial saliva was produced by UNISUL (University of Southern Santa Catarina, Florianópolis, SC, Brazil) in the pharmacy laboratory as proposed by Perdigão et al (1998): 25 mM KH2PO4, 24 mM Na2HPO4, 150 mM KHCO3, 100 mM NaCl, 1.5 mM MgCl2, 15 mM CaCl2 and 6 ml of 25 mM citric acid (pH 6.8), qsp 100 ml.

Average values of initial pH were compared to the amount of artificial saliva used to test the neutralizing capacity of each group, by using ANOVA statistical analysis at the 0.05 significance level.

RESULTS

It was observed that the pH of the analyzed solutions was acidic, varying significantly between groups (Table 1). In general, regular and light soft drinks showed a low initial pH level.

However, the cola-based drinks, whether regular or light, required a smaller volume of saliva to reach the cutoff point (pH 5.5) than energy drinks (Table 2).

The cola-based drinks, whether regular or light, required a smaller volume of saliva to reach the cutoff point (pH 5.5) than energy drinks. Furthermore, the Table 2 shown that the energy drinks needs twice higher saliva volume to reach pH near 5.0, although all energy drink samples failed to reach the pH 5.5 set as the cutoff point.

### Table 1: Average pH values and standard deviation of the different beverages evaluated

<table>
<thead>
<tr>
<th>Groups</th>
<th>RCD 1</th>
<th>LCD 1</th>
<th>RCD 2</th>
<th>LCD 2</th>
<th>RED 1</th>
<th>LED 1</th>
<th>RED 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Average pH values</td>
<td>2.3a</td>
<td>2.9a</td>
<td>2.5a</td>
<td>2.7a</td>
<td>3.3b</td>
<td>3.4b</td>
<td>2.6a</td>
</tr>
<tr>
<td>Standard deviation</td>
<td>0.11</td>
<td>0.09</td>
<td>0.11</td>
<td>0.05</td>
<td>0.05</td>
<td>0.05</td>
<td>0.13</td>
</tr>
<tr>
<td>Minimum pH</td>
<td>2.2</td>
<td>2.7</td>
<td>2.4</td>
<td>2.7</td>
<td>3.3</td>
<td>3.3</td>
<td>2.5</td>
</tr>
<tr>
<td>Maximum pH</td>
<td>2.5</td>
<td>3.1</td>
<td>2.7</td>
<td>2.8</td>
<td>3.4</td>
<td>3.5</td>
<td>2.8</td>
</tr>
<tr>
<td>N</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Anova (p-value)</td>
<td>4.19E-40</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Average pH values followed by same letters, a or b, do not differ statistically.

### Table 2: Mean volumes of artificial saliva needed to raise the pH level to 5.5

<table>
<thead>
<tr>
<th>Item</th>
<th>RCD 1</th>
<th>LCD 1</th>
<th>RCD 2</th>
<th>LCD 2</th>
<th>RED 1</th>
<th>LED 1</th>
<th>RED 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean volume of saliva (ml)</td>
<td>6.8</td>
<td>5.7</td>
<td>5.3</td>
<td>6.0</td>
<td>&lt;12.5</td>
<td>&lt;12.7</td>
<td>&lt;11.3</td>
</tr>
<tr>
<td>Standard deviation</td>
<td>0.9</td>
<td>0.9</td>
<td>0.6</td>
<td>0.5</td>
<td>2.4</td>
<td>2.2</td>
<td>2.0</td>
</tr>
<tr>
<td>Minimum saliva (ml)</td>
<td>5.3</td>
<td>5.0</td>
<td>4.3</td>
<td>5.1</td>
<td>9.3</td>
<td>9.5</td>
<td>8.3</td>
</tr>
<tr>
<td>Maximum saliva (ml)</td>
<td>8.0</td>
<td>7.6</td>
<td>6.6</td>
<td>7.0</td>
<td>16.6</td>
<td>16.6</td>
<td>14.3</td>
</tr>
<tr>
<td>N</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Anova (p-value)</td>
<td>6.34E-21</td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
</tbody>
</table>
Graph 1 illustrates the pH adjustment behavior in a sample from each group of cola-based drinks and energy drinks. Cola-drinks need low volumes of artificial salivary to reach pH above 5.0, however, is observed a resistance of energy drinks to have been neutralized by artificial saliva used in this study attributed to the high titratable acidity of these beverages.

DISCUSSION

The etiology of noncarious lesions is related to both intrinsic and extrinsic factors of the individual. Diet is a major extrinsic factor for dental erosion. It should be noted that the consumption of industrialized beverages, which contain components that decrease the pH of saliva, leading to mineral loss of tooth structure and increasing the risk of dental erosion.  

Soft drinks and energy drinks are both widely available in supermarkets, drug stores and vending machines all across the world. People consume these beverages on a daily basis, unaware of their nutritional information. While soft drinks and energy drinks share many similarities, there are important differences between the two types of beverages.

Soft drinks consist of mostly carbonated water and sugar. The other ingredients of the beverage are designed to enhance the taste, color the product or preserve the freshness of the beverage. Energy drinks, on the other hand, typically include a variety of supplemental ingredients designed to serve specific purposes. A most common ingredient in energy drinks is caffeine (often in the form of guarana or yerba mate). Energy drinks contain about three times the amount of caffeine as cola.

Soft and energy drinks contain high levels of sugar too, because this many brands offer artificially sweetened ‘light’ versions. In this case, as in the brands tested in this study, they use aspartame to substitute sugar.

Confirming the dental erosion risks, Table 1 shows the initial average pH of cola-based drinks and energy drinks investigated. It can be noted that all the beverages had an initial pH level below the tolerance limits independent if regular or light.

According to the literature, a pH below 5.5 is considered critical for starting enamel dissolution. With this in mind, we tried to estimate the average amount of saliva required to raise the pH to 5.5 to reach a safe level for the control of mineral loss and the development of erosive lesions. According to the results shown in Table 2, the cola-based drinks required a smaller volume of saliva to raise the pH level compared to energy drinks. This happened regardless of whether the beverages were regular or light.

Furthermore, as shown in Graph 1, the behavior was different for the samples of energy drinks in relation to soft drinks. The former required a larger volume of saliva than the latter to raise the pH to the desired level.

Despite some brand secrets to composition of the drinks evaluated, it is believed that this may have occurred because energy drinks are often associated with citric acid and sodium citrate, among other substances, which is known to minimize the buffering effect of pH variation, preventing to reach the cutoff point after the addition of artificial saliva by the method used.

One of the functions of saliva is to regulate the pH of the oral environment by salivary buffers. According to Ganss (2006), the buffering capacity corrects the pH alterations that occurred with the formation of acidic and basic ions, protecting the teeth against enamel demineralization. Dawes, Kubieniec (2004) measured the salivary flow and, as a result, they obtained 1 ml/min saliva secreted under stimulation and 0.37 ml/min without stimulation. From a conservative perspective, taking into account the value of 1ml/min, the results presented in Table 2 and the data
proposed by Lagerlöf and Dawes\(^\text{19}\) (1984), according to whom, when taking liquids, the remaining average volume in the oral cavity will be 1 ml, one can estimate that it will take 5 to 7 minutes, on average, to raise the pH level above 5.5 after taking cola-based drinks. As for energy drinks, it will take at least 11 to 12 minutes, since in none of the samples the pH established as a cutoff point was reached.

Therefore, according to the results, one can observe that the cola-based drinks required a smaller volume of saliva to raise the pH level, and probably are neutralized faster than the energy drinks.

Finally, it should be highlighted that, due to the nature and high prevalence of noncarious lesions of the erosion type, as well as the change in eating habits of the population, with increasing consumption of these types of drinks, associated with the frequent launching of new food products in the market, further studies on the subject should be constantly carried out and published. The research design should include randomized clinical trials, whenever possible.

**CONCLUSION**

According to the methodology used, it can be concluded that:
1. All beverages analyzed showed an initial acidic pH, which is likely to potentiate the risk of erosive lesions.
2. The cola-based drinks that were subject to the addition of artificial saliva reach the pH value set as a parameter, whereas the energy drinks did not reach the pH set as the cutoff point, raising the risk for erosive lesions.
3. There was no statistical difference between the initial pH level and acid neutralization by the addition of artificial saliva in both regular and light drinks.

**REFERENCES**


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