

Action of the Salivary Microorganisms having Sorbitol as Substratum

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ABSTRACT

Microorganisms living in the saliva are able to ferment some sweetener types and to promote the fall of pH that has its critical point around 5.5; in this situation, they equal the input and the calcium output in the processes of remineralization and demineralization. For iteration of the three essential factors to the installation of the dental caries (microorganism, host and diet) suggested the accomplishment of a work that involved one of these factors. This work aims to study this issue, through the indication of substitute sweetener of the saccharose, whenever possible, it solved to evaluate pH of the saliva, exposed to sorbitol.

Keywords: Sorbitol, Sweetener and dental caries, Microorganism.

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INTRODUCTION

The installation of the dental caries depends on the interaction of three essential factors: host (tooth), microbiota (with potential cariogenic) and the diet (rich in fermentable carbohydrates).¹ Alterations in any of these factors influence directly in the appearance of dental caries. With the objective of work in the source of the problem, main attention should be given to the diet, mostly in respect

to the sugar consumption, mostly responsible person for the occurrence of the caries² and whose relation is irrefutable.³

The dental plaque is a consistent mass, adherent, full of microorganisms that colonizes the surface of the tooth, which are able to form acids from saccharose, which corresponds to 99% in the constitution of sugar. Through glicolitic activity, it decreases mouth pH, causing demineralization of the dental tissue.⁴⁻⁷

It is believed that the main maneuver with the objective of favoring the dental health be the retreat of the sugar from the oral environment, or in a less radical situation, the reduction of consumption frequency of food that contains saccharose. Such measure (standard action) is base on the dietary education, which is proved an effective mean of reduction of the dental caries.⁸

LITERATURE REVIEW

Westergren et al,⁹ analyzed the genetics transfer possibility of the fermentation ability of the sorbitol. The authors concluded that *Streptococcus sanguis* non-fermentable of the sorbitol acquired such capacity, through its exhibition to the DNA of *S. sanguis* and *mitior*, possessors of such a property.

Glass,¹⁰ accomplished a clinical experiment of 2 years with bubble gums sweetened with sorbitol in children (540) in an area that was not fluoridated. A group did use of the chewing gums twice a day for 2 years and another group did not chew the chewing gums. In regard to the average level of the caries, there were no significant differences found and-according to the author, the results demonstrated that sorbitol was not cariogenic.

Slee and Tanzer,¹¹ studied the metabolism of sorbitol by *Streptococcus mutans* 'in vitro'. They noted that the sweetener used by these bacteria via sorbitol phosphotransferase/sorbitol-6-phosphate dehydrogenises, being this utilization quickly repressed by the presence of the glucose.

Roe,¹² related that the exhibition of the humans for normal levels of sorbitol or xylitol did not convey any risks to neoplasm development.

Flisykowska,¹³ studied the effect of the sweetener substances (saccharose to 3%, glucose to 6% and sorbitol to 10%) in the development of the caries, the bovine

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enamel adapted in volunteers' removable prosthesis. The volunteers were guided that they should immerse their prosthesis in the sweetener solutions (each group with your respective substitute of the sugar) daily for 10 minutes up to 6 weeks. The initial destruction of enamel by the treatment with the saccharose belonged to 28.8%, 22.2% with the glucose and 8% with sorbitol.

Izumitani et al,¹⁴ analyzed the caries inductor capacity of a sweetener (77% of maltitol, 2% of sorbitol and 21% several sweetener) in mice infected with *S. mutans*. The results indicated that the caries index of mice group fed with that sweetener was significantly smaller when compared to the saccharose.

Jensen,¹⁵ analyzed the variations of pH of the interproximal board after ingestion of sugary food and the effect in pH using bubble gums sweetened with sorbitol. The chewed gum; 15 minutes after the beginning of the ingestion for 10 minutes provoked a fast increase of pH on the board that was kept for 30 minutes in a safe considered level for the tooth. Therefore, the author concluded that all the foods (chocolate bars, drought grapes, chocolate crackers stuffed with cream, cakes with cream frosting and stuffing, and cherry pies) produced pH fast decrease of the plaque.

Firestone and Naivia,¹⁶ analyzed *in vivo* the caries incidence and the variations of pH of the molar teeth furrows board of mice after topical applications of sorbitol and saccharose. The authors concluded that there was adaptation on the board with mice on a diet of 20% of sorbitol and that the furrow caries incidence in the group of sorbitol was significantly smaller than in the group of the saccharose.

Kalfas and Birkhed,¹⁷ analyzed the aerobic terms effect or anaerobic in the acidogenic activity of sorbitol, by the dental board and for *Streptococcus* oral. The authors noted that the acids production by the dental board neither of sorbitol nor of the glucose were affected by the experimental terms; they were very small, the differences of the concentrations of the four final metabolic products (succinate, formate, acetate, and ethanol). Suspension pH of *S. mutans* in glucose was the same. However the experimental situation, pH in the suspension of *S. mutans* incubated with sorbitol, however, was considerably smaller under aerobic than under anaerobic condition.

Rugg-Gunn,¹⁸ analyzed the alterations in pH of the board caused by Iycasin (to 70%) or sorbitol (to 70%) to replace the saccharose (to 70%) in medicinal syrups and in cooked candies. The author noted that the changes in pH of the board caused by sorbitol and by Iycasin were very small and with negligible differences, 50 much in the candies as in the syrups, while the saccharose provoked greater alterations. The author concluded that

the substitution of the saccharose for Iycasin or sorbitol in medicinal syrups was considered beneficial to the children's dental health who use this type of treatment.

Makinen and Isokangas,¹⁹ related that it has been existing growth in the substitution indication of the saccharose for sweetener less cariogenic in products consumed during the secondary meals.

Leach et al,²⁰ studied for remineralization of artificial lesions of caries on human enamel, *in situ*, for bubble gums sweetened with sorbitol. Enamel portions with carious lesions produced were artificially cemented in bands with the molar teeth of adult individuals. Soon after the meals, for 3 weeks, they chewed five gums a day for 20 minutes. The authors demonstrated that according to the content of mineral nominated in the microscopic exam of polarized light, there was significantly remineralization.

Kashket et al,⁵ analyzed the use of bubble gum sweetened with sorbitol in demineralization of the enamel induced by the saccharose. The authors concluded that the use of bubble gum with sorbitol soon after the sugar ingestion can reduce for demineralization of the enamel, and it can also increase for subsequent remineralization.

Söderling et al,²¹ studied the effects of sorbitol (10.9 gm/day) of xylitol (10.9 gm/day) and of a xylitol and sorbitol mixture (8.5 and 2.49 respectively/day) under bubble gum forms in the dental boards of three adults' groups (average age of 22.5 years). A group of quarter of habitual user of gum with saccharose was used as a control. The authors concluded that the individuals who used gums with xylitol and xylitol-sorbitol showed a significantly larger ability to resist the falls of pH than the group of sorbitol; there was a decrease of the board quantity in the groups of xylitol-sorbitol (24.3%) and xylitol (29.4%), and an increase in the group of sorbitol (48.3%); in the group of sorbitol the *S. mutans* levels in the dental board and in the saliva increased and in the groups where xylitol was used, it decreased.

Siebert and Forsthuber,²² related how several proxies of sugar that were not cariogenic acted as modifying of glucan synthesis and therefore they contributed for their properties that were not cariogenics and sometimes even anticariogenics.

Grenby et al,²³ studied *in vitro* the favorable and the unfavorable properties to the dental health of lactitol in comparison to other three glucose saccharose sweeteners; sorbitol, manitol and xylitol. The cultures were incubated by 24 hours contended the sweetener as energy source. The attack to the tooth, generated by the acid, was measured by calcium and match analyses. The results showed that for demineralization was more severe with the glucose and the saccharose; the minor acid productions were generated by sorbitol and by manitol.

With demineralizations reduced; the fermentations of lactitol and of xylitol were soft, carrying pH high levels. With demineralizations extremely low; the polysaccharides quantity synthesized by the incubated microorganisms introduced in decreasing order on the following way: saccharose glucose > sorbitol > manitol > lactitol > xylitol.

Kalfas et al,²⁴ analyzed the adaptation of the dental board to sorbitol in people with normal levels and short salivary secretion. The authors concluded that the frequent exhibition of sorbitol to the oral cavity carries to an adaptation of the dental board to this polyol. The acidity created by the board under the exhibition of sorbitol, in people with drop level of salivary secretion, it was of larger greatness and of longer duration when compared with people of normal salivation; pH of the board reached critical levels for demineralization of the teeth of people with smaller natural protection against the caries, such as the ones that own low salivary secretion, considering the fact that sorbitol is, in many cases, included in regularly used products.

Bowen et al,²⁵ analyzed the sweetener influence in solutions, in the formation of dental caries in mice without salivation. The results showed that for sucralose (trichlorogalactosucrose), sorbitol and aspartame did not induce the formation of dental caries in the mice infected with *Streptococcus sootinus* and *Actinomyces viscosus*; in contrast, the saccharose and for fructose induced greater dental destructions; *Actinomyces visosus*' population was very small with the animals that received fructose and saccharose and it was greater with the animals that received sucralose, sorbitol and aspartame.

Birkhed et al,²⁶ analyzed the caries level and with the factors relating them with individuals who had prolonged use of sorbitol. The discoveries of these studies confirmed the previous observations that the frequent consumption of sorbitol can cause small adaptation of the dental board; however, these alterations did not result in high activity of caries.

More et al,²⁷ compared for remineralization of lesions on human enamel, *in vitro*, through the bubble gums used with sorbitol or with fluoride. The volunteers were divided into two groups. They brushed their teeth 3 times a day with fluoride-free toothpaste, for a control period of 21 days within the two test periods. Each group chewed 1 chewing gum 5 times a day. The results demonstrated that both regimes produced increase of the mineral density, but the group of the fluoride produced a remineralization significantly larger than the group of sorbitol, at the same time in which it was more resistant to the acidogenic activity.

Park et al,²⁸ analyzed the bubble gum effects with saccharose or sorbitol in pH of the dental board of

volunteers. After a board accumulation for 3 to 6 days, they chewed a chewing gum with sorbitol or saccharose for 10, 15 or 20 minutes. The pH was measured by 2 hours and the results pointed that the gum with saccharose produced an acidogenic, so the answer was significantly greater than the gum with sorbitol.

Rogers et al,²⁹ analyzed the sweetener effects in *S. mutans* and *Streptococcus mittett* cultures. The results showed that the glucose to fructose, the lactose to xylose and sorbitol provided increase in *S. mutans* population and did not affect significantly *Streptococcus milleri*, and xylitol demonstrated an antimicrobial effect in *S. mutans* and did not affect *S. milleri*.

A study found that both sorbitol and xylitol could be fermented by reducing the pH and demineralize tooth enamel.³⁰ However, the supply may reduce the cariogenic plaque pH to more than just sorbitol levels.

This research aims to evaluate the fermentation of the salivary microorganisms exposed to the sweetener sorbitol.

MATERIALS AND METHODS

Pool of Saliva

A pool of saliva was used. Thirty-six people (85.5% of dentistry students) donated their saliva for this research. It was gathered without stimulation and it was put in a glass with sterilized cover, and then it was packed properly in a temperature box on the rocks for posterior carries until the microbiology laboratory of the University Camilo Castelo Branco.

Preparation of the Mean Culture Exempted of Carbon Source

The substances used to prepare the mean culture are on Table 1.

Preparation of the Solution to 10% of the Sweetener Sorbitol

It was used 1.09 of the sweetener in 9 ml of distilled water. After it dissolved completely, the solution was sterilized in a millipore filter with porosity of 0.22 micrometer.

Preparation of the BASE (Culture Medium) and Sweetener

In each 4.5 ml of culture medium, previously sterilized, 0.5 ml of the solution was added. Fifty as to obtain a final concentration of 1% of the sweetener in the culture medium.

Preparation of Salivary Inoculum in the Culture Medium

It added 0.5 ml of saliva in a series of four test tubes with cover contend sweetener + culture medium, and another



Table 1: Substances used to prepare the mean culture

Substances	
Magnesium sulfate	20 gm
Ammonium hydrogen phosphate	1.00 gm
Potassium phosphate	1.00 gm
Sodium chloride	5.00 gm
Bromotimol-indicator blue	0.08 gm
Distilled water	1000.0 ml
The final pH of the environment was 6.8	

Table 2: Values of the pH on the solution of sorbitol and control group

Hours		Sorbitol	Control
24	Mean	6.7	7.0
	No saliva	7.0	7.0
48	Mean	5.5	7.0
	No saliva	7.0	7.0
96	Mean	4.7	7.0
	No saliva	7.0	7.0

tube received only the culture medium + the sweetener, in other words, without saliva to be the microcontrol.

The tubes were incubated to 37°C, where after 24, 48 and 96 hours was accomplished visual reading and pH mensuration through indicators toothpicks, the tubes during the reading remained with the same time outside the stove, 50 that it kept the standardization.

Another series of four tubes was used as macrocontrol, in other words, three of the four tubes received only the saliva culture medium, and the remaining tube was the macro-control, it received only the culture medium.

RESULTS

Values of the pH on the solution of sorbitol and control group measured to the 24, 48 and 96 hours are on Table 2.

DISCUSSION

The purpose of this study was to evaluate the influence on salivary pH front to the sorbitol. The study confirmed what other researchers have shown related to the effect of the sorbitol in reducing the salivary pH.^{24,31}

Gil and Cury,³¹ evaluated the cariogenic potential of six chewing gums, among them, one possessed a sorbitol composition. As from the metabolism of sugar containing sucrose by salivary bacteria, there was a decrease in the pH 1.5 to 2.0 units. In the gum containing sorbitol, it decreased 0.4 units.

Kalfas et al, in 1990,²⁴ studied sorbitol and glucose acids production under anaerobic terms of bacteria of the belonging dental board to *Streptococcus*, *Lactobacillus* and *Actinomyces*. The results demonstrated that sorbitol fermented more slowly. The pH reduction activity

decreased acid production of the two carbohydrates, wherein the total amount of acids formed to smaller sorbitol.

Burt,³² compared the caries-inhibitory action of sorbitol and xylitol in sweetened chewing gums and assessed the role of these products in caries prevention. When compared with sugar-sweetened gum, sorbitol sweetened gum had low cariogenicity when it was chewed no more than three times per day.

Holgerson et al,³³ studied the reduction of dental plaque through the use of chewing gums containing xylitol or sorbitol. He observed a significant increase in salivary flow in chewing gum containing sorbitol.

Runnel et al,³⁴ collected samples of saliva and plaque from individuals 7 to 8 years of age. Subjects were randomized into three groups consumed erythritol, xylitol or sorbitol candy. As a result, it was observed that the use of bullets containing erythritol, has been associated with reduced growth of the plates and reduced oral count *mutans streptococci* compared with the consumption of sweets containing xylitol or sorbitol.

Raju et al,³⁵ concluded that the sorbitol liquid and gum significantly increase the salivary flow. However, the use of liquid sorbitol would be beneficial in the treatment hypo salivation due to only topical action.

CONCLUSION

During the experiment, sorbitol presented a decreasing pH that resulted in a value (pH = 4.7) considered harmful to the dental health.

REFERENCES

- Keyes PH. Recent advances in dental caries research. Bacteriology. Bacteriological findings and biological implications. Int Dent J 1962;12(4):443-464.
- Carlos JP. A debate over the role of sugars in the etiology of dental caries. J Pedod 1983;7(4):330-332.
- Newbrun E. Sucrose in the dynamics of the carious process. Int Dent J 1982;32(1):13-23.
- Edgar WM. The role of sugar in the aetiology of dental caries: 3. The physicochemical evidence. J Dentistry 1983;11(3):199-205.
- Kashket S1, Yaskell T, Lopez LR. Prevention of sucrose-induced demineralization of tooth enamel by chewing sorbitol gum. J Dent Res 1989;68(3):460-462.
- Mühlemann HR. The effect on dietary carbohydrate counselling of plaque pH telemetry. Quint Int 1983;14(2):201-206.
- Pinheiro CE. Biochemistry course of dental caries I. Biochemistry of saliva. Odont Paul Rev 1982;4(4):40-47.
- Fonseca YPC, Guedes-Pinto AC. Diet food control in pediatric dentistry patients with high incidence of caries. APCD Rev 1984;38(4):289-301.
- Westergren G, Krasse B, Birkhed D, Edwardsson S. Genetic transfer of markers for sorbitol (D-glucitol) metabolism in oral streptococci. Archs Arch Oral Biol 1981;26(5):403-407.

10. Glass RL. A two-year clinical trial of sorbitol chewing gum. *Caries Res* 1983;17(4):365-368.
11. Slee AM, Tanzer JM. The repressible metabolism of sorbitol (D-glucitol) by intact cells of the oral plaque-forming bacterium *Streptococcus mutans*. *Arch Oral Biol* 1983;28(9):839-845.
12. Roe FJ. Perspectives in carbohydrate toxicology with special reference to carcinogenicity. *Swed Dent J* 1984;8(3):99-111.
13. Flisykowska AK. Study on the cariogenic effects of various substances used for sweetening in an intraoral model. *Czasop Stomat* 1985;38(5):357-363.
14. Izumitani A, Sumi N, Kusamura Y, Ooshima T, Sobue S. Caries-inducing activity of a maltitol-rich sweetener in the experimental dental caries of rats. *Shoni Shikagaku Zasshi*. 1985;23(1):56-61.
15. Jensen ME. Effects of chewing sorbitol gum and paraffin on human interproximal plaque pH. *Caries Res* 1986;20(6):503-509.
16. Firestone AR, Navia JM. In vivo measurements of sulcal plaque pH in rats after topical applications of xylitol, sorbitol, glucose, sucrose, and sucrose plus 53 mM sodium fluoride. *J Dent Res* 1986;65(1):44-48.
17. Kalfas S, Birkhed D. Effect of aerobic and anaerobic atmosphere on acid production from sorbitol in suspensions of dental plaque and oral streptococci. *Caries Res* 1986;20(3):237-243.
18. Rugg-Gunn AJ. Effect of Lycasin upon plaque pH when taken as a syrup or as a boiled sweet (short communication). *Caries Res* 1988;22(6):375-376.
19. Makinen KK, Isokangas P. Relationship between carbohydrate sweeteners and oral diseases. *Prog Food Nutr Sci* 1988;12(1):73-109.
20. Leach SA, Lee GT, Edgar WM. Remineralization of artificial caries-like lesions in human enamel in situ by chewing sorbitol gum. *J Dent Res* 1989;68(6):1064-1068.
21. Söderling E, Mäkinen KK, Chen CY, Pape HR Jr, Loesche W, Mäkinen PL. Effect of sorbitol, xylitol, and xylitol/sorbitol chewing gums on dental plaque. *Caries Res* 1989;23(5):378-384.
22. Siebert G, Forsthuber F. Activations of glucosyltransferases from *Streptococcus mutans* AHT by noncariogenic polyols and isomaltulose. *Caries Res* 1989;23(2):127.
23. Grenby TH, Phillips A, Mistry M. Studies of the dental properties of lactitol compared with five other bulk sweeteners in vitro. *Caries Res* 1989;23(5):315-319.
24. Kalfas S, Maki Y, Birkhed D, Edwardsson S. Effect of pH on acid production from sorbitol in washed cell suspensions of oral bacteria. *Caries Res* 1990;24(2):107-112.
25. Bowen WH, Pearson SK, Falany JL. Influence of sweetening agents in solution on dental caries in desalivated rats. *Arch Oral Biol* 1990;35(10):839-844.
26. Birkhed D, Svensäter G, Edwardsson S. Cariological studies of individuals with long-term sorbitol consumption. *Caries Res* 1990;24(3):220-223.
27. More F, Lamb W, Corpron R. Remineralization of enamel lesions with fluoride-containing versus sorbitol chewing gum. *J Dent Res* 1991;70(Spec. no):453.
28. Park KK, Schemehorn BR, Bolton JW, Stookey GK. Effect of sorbitol gum chewing on plaque pH response after ingesting snacks containing predominantly sucrose or starch. *Am J Dent* 1990 Oct;3(5):185-191.
29. Rogers AH, Pilowsky KA, Zilm PS, Gully NJ. Effects of pulsing with xylitol on mixed continuous cultures of oral streptococci. *Aust Dent J* 1991;36(3):231-235.
30. Mann J. Sugar revisited; again. *Bull World Health Organ* 2003; 81(8):552.
31. Gil PSS, Cury JA. Cariogenic potential of chewing gums. *RGO (Porto Alegre)* 1986;34(2):121-124.
32. Burt BA. The use of sorbitol- and xylitol-sweetened chewing gum in caries control. *J Am Dent Assoc* 2006;137(2):190-196.
33. Holgerson PL, Sjöström I, Stecksén-Blicks C, Twetman S. Dental plaque formation and salivary mutans streptococci in schoolchildren after use of xylitol-containing chewing gum. *Int J Paediatr Dent* 2007;17(2):79-85.
34. Runnel R, Mäkinen KK, Honkala S, Olak J, Mäkinen PL, Nömmela R, Vahlberg T, Honkala E, Saag M. Effect of three-year consumption of erythritol, xylitol and sorbitol candies on various plaque and salivary caries-related variables. *J Dent* 2013;41(12):1236-1244.
35. Raju GS, Morghal MM, Hossain MS, Hassan MM, Billah MM, Ahamed SK, Rana SM. Assessment of pharmacological activities of two medicinal plant of Bangladesh: *Launaea sarmentosa* and *aegialitis rotundifolia* roxb in the management of pain, pyrexia and inflammation. *Biol Res* 2014;28;47(1):47-55.

