Turku sugar studies XXI

Xylitol-, sorbitol-, fructose- and sucrose-induced physico-chemical changes in saliva

EVA SÖDERLING, MAARIT REKOLA, KAUKO K. MÄKINEN & ARJE SCHEININ

Institute of Dentistry, University of Turku, Finland

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The aim was to study eventual physico-chemical changes occurring in whole saliva due to sweetened and unsweetened stimulators. The assay was carried out in 10 female subjects with regard to changes of pH, buffering capacity and electrolytes in saliva as influenced by chewing of fructose, sucrose, sorbitol and xylitol gum, gum base and paraffin. The flow rate of saliva was measured in relation to use of xylitol and sucrose chewing gum and unsweetened gum base. These sweeteners increased significantly the salivary flow rate in comparison to the unsweetened gum base. Generally, xylitol and sorbitol on one hand, and sucrose and fructose on the other, behaved in an almost similar way. Increased buffering capacity and elevation of pH in saliva was found in the presence of the polyols tested.

Key-words: Saliva; chewing gum; xylitol; sorbitol; fructose; sucrose

Eva Söderling, Institute of Dentistry, University of Turku, SF-20520 Turku 52, Finland

The principal biochemical findings on whole saliva in the present series of studies indicated that there were no significant differences between the sugar groups on strict sucrose, fructose and xylitoldiet with regard to calcium, sodium and inorganic phosphate concentrations (*Mäkinen & Scheinin*, 1975). It should be noted, however, that these determinations were carried out in paraffin-stimulated saliva. Hence no conclusions were drawn with regard to the eventual immediate effects of the sugars tested on these parameters.

On the other hand, it has been recently observed that intake of various sucrose-containing sweets has resulted in a marked increase of the ioncontent and flow rate of saliva (*Shannon, Suddick & Dowd*, 1974). These observations indicated a correlation between the salivary flow rate and the calcium concentration, which were particularly influenced through consumption of sucrose-containing acidic candies.

It was thus decided to study the eventual physico-chemical changes occurring in whole saliva in relation to the presence of a disaccharide (sucrose), a monosaccharide (fructose), a 5-C-polyol (xylitol) and a 6-C-polyol (sorbitol). The effects of these sweeteners were investigated through the use of chewing gums sweetened with the respective sugars. Unsweetened gum base and paraffin were used as controls.

MATERIALS AND METHODS

1. *Subjects and collection of samples.* Ten female dental students aged 20—22 years

acted as voluntary subjects in the investigation. Before every test the subjects had been fasting for two hours. The following sweetened chewing gums were used: sucrose, xylitol, sorbitol and fructose gum. The composition of the chewing gums used was principally the same as earlier (*Scheinin et al.*, 1975). The only differences between the products were the sweeteners mentioned above which were used in a concentration of 50 %. Unflavored chewing gum base and paraffin were used as unsweetened stimulators.

The experiments were carried out on consecutive days in the morning (9—11 a.m.) according to the following schedule:

Day 1. Resting saliva
Fructose chewing gum

Day 2. Paraffin
Xylitol chewing gum

Day 3. Chewing gum base
Sucrose chewing gum

Day 4. Sorbitol chewing gum

On each day involving two series of experiments the second set was initiated after resting saliva conditions were obtained.

The stimulation by chewing lasted for two minutes and thereafter the stimulator was discarded. The first saliva samples (2 ml) were collected immediately and the following samples 10 and 30 min. after the stimulation. The flow rate was measured in a graduated test tube by collecting resting saliva for 10 min. and stimulated saliva for 3 min.

2. *Chemical methods.* All reagents were E. Merck's a-grade. The water (specific resistance 10^6 Ωcm) was distilled and treated with an ion exchange resin.

The following physico-chemical properties of saliva were measured: The pH-values were determined immediately from whole saliva with a pH-meter (glass- and calomel-electrodes, Radiometer pHM 26). P_{CO_2} was measured immediately from whole saliva (E5036/D616 P_{CO_2} single Electrode Assambly combined with PHM 71 Mk2 Acid Base Analyzer, Radiometer). Calcium was titrated with EDTA from centrifuged saliva ($12000 \times g$, 10 min, +4° C). Inorganic phosphate was determined from centrifuged saliva by the method of *Fiske* and *Subbarow* (1957). Sodium was determined from centrifuged saliva with atomic absorption (Perkin-Elmer 303). Chloride was titrated with Ag^+-ions (Aminco, automatic coulometric chloridetitrator).

3. *Statistical methods.* The level of significance of the difference between the arithmetic or logarithmic means were calculated by using Student's t-test. The significance levels were: $p < 0.1$ almost significant, $p < 0.05$ significant.

RESULTS

The following physico-chemical changes in saliva were observed:

1. *pH.* Fig. 1 shows the changes in pH of saliva due to different stimulations. All stimulators caused an immediate and significant rise in pH-values.

It is to be observed that still after 30 min. sucrose and fructose stimulations resulted in significantly lower pH-values than observed in relation to paraffin or gum base chewing.

2. *Buffer systems of saliva*

a) *Inorganic phosphate.* All stimulators caused an immediate and significant decrease in phosphate values as compared to resting saliva (Fig. 2).

The HPO_4^{2-}-diagrams were calculated from phosphate and pH-values (Fig. 3). The sweetened stimulators caused a

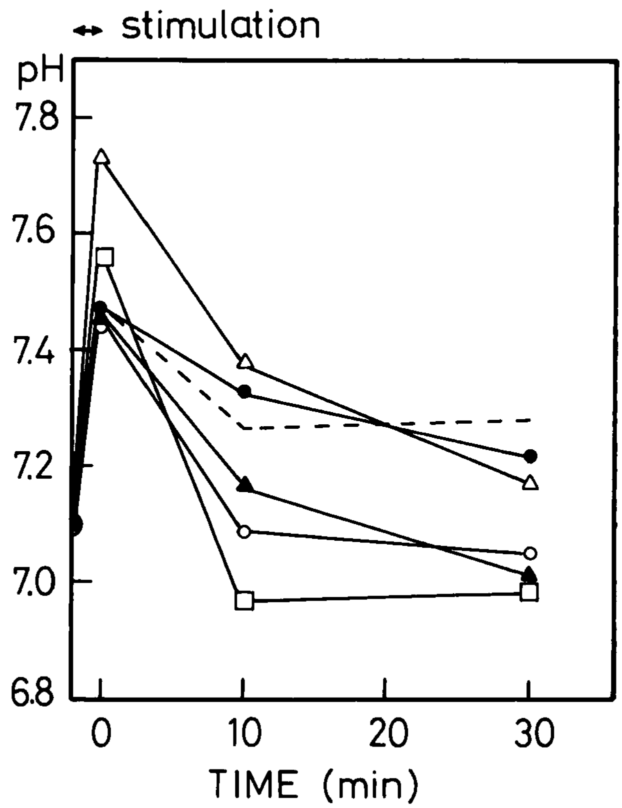

Fig. 1. The pH of saliva due to stimulation with sweetened and unsweetened stimulators. The diagrams represent logarithmic means of pH-values from 10 test persons. The stimulation time was 2 min. Stimulators: ● chewing gum base, - - - paraffin, △ xylitol gum, ▲ sorbitol gum, ○ sucrose gum, □ fructose gum. ⊙ Resting saliva.

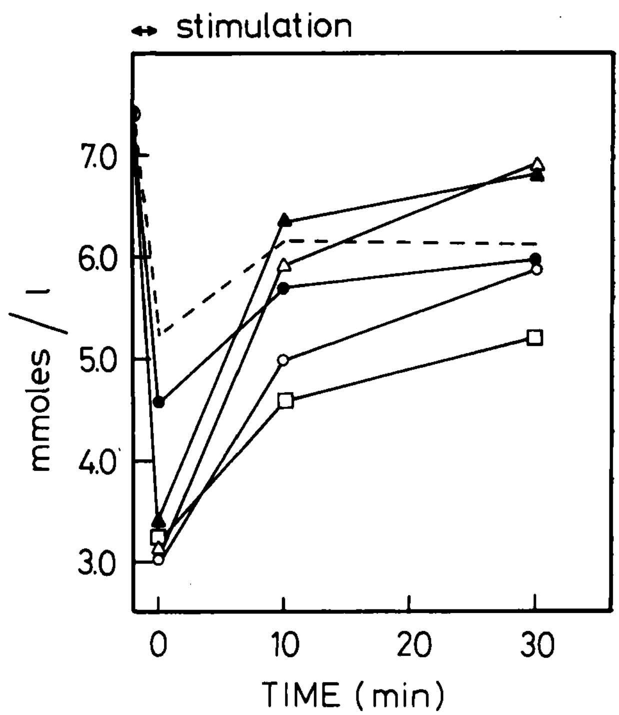

Fig. 2. Inorganic phosphate of saliva due to stimulation with sweetened and unsweetened stimulators. The phosphate values are arithmetic means of values from 10 test persons. Other details as in Fig. 1.

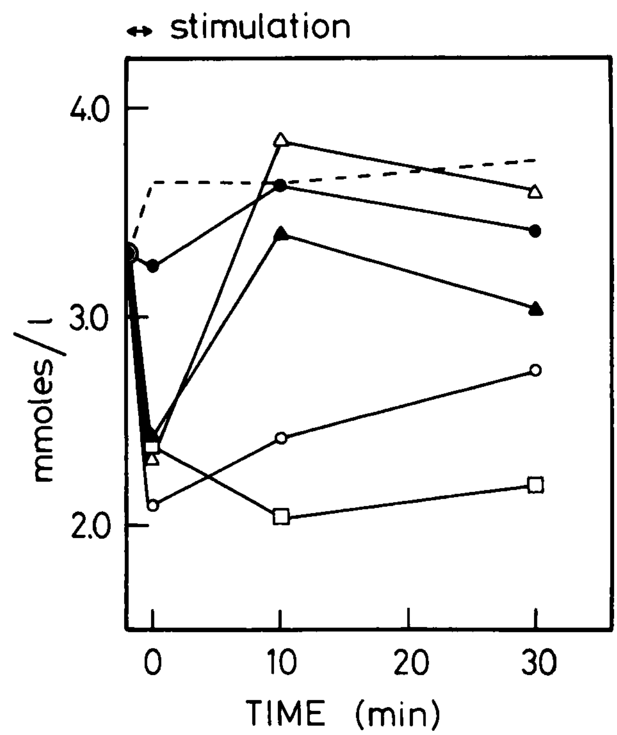

Fig. 3. Salivary HPO_4^{2-}. The diagrams were calculated from phosphate and pH. Other details as in Fig. 1.

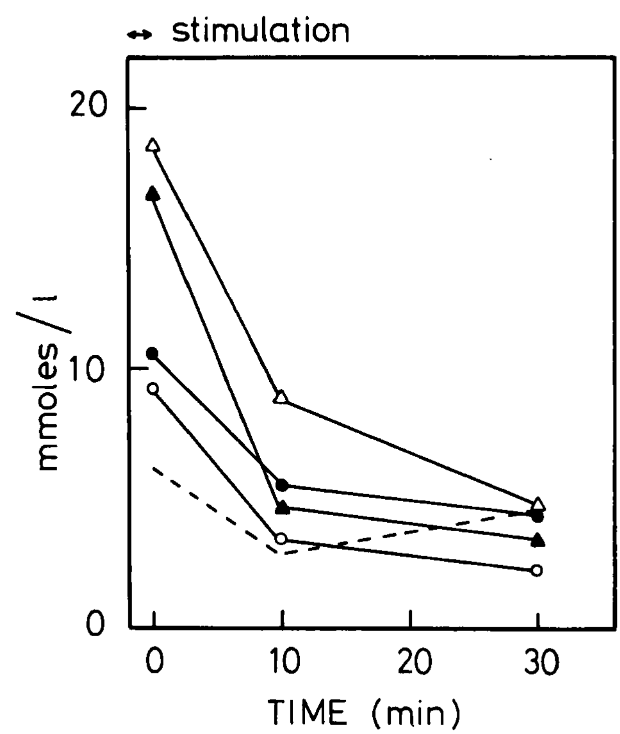

Fig. 4. Salivary HCO_3^- due to stimulation with sweetened and unsweetened stimulators. The arithmetic means of values from 10 test persons. Stimulation time was 2 min. Stimulators: ● chewing gum base, - - - paraffin, △ xylitol, O sucrose, ▲ sorbitol.

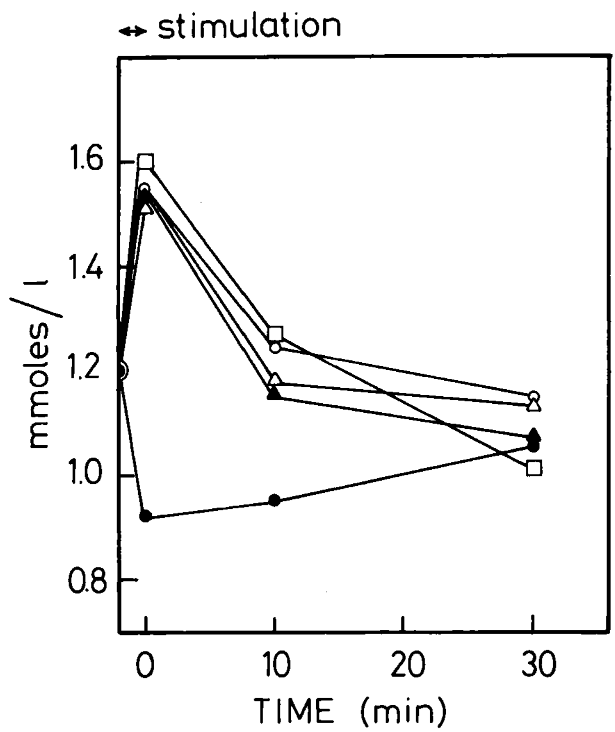

Fig. 5. Salivary Ca^{2+}-levels due to stimulation with sweetened and unsweetened stimulators. Arithmetic means of values from 10 test persons are given. Other details as in Fig. 1.

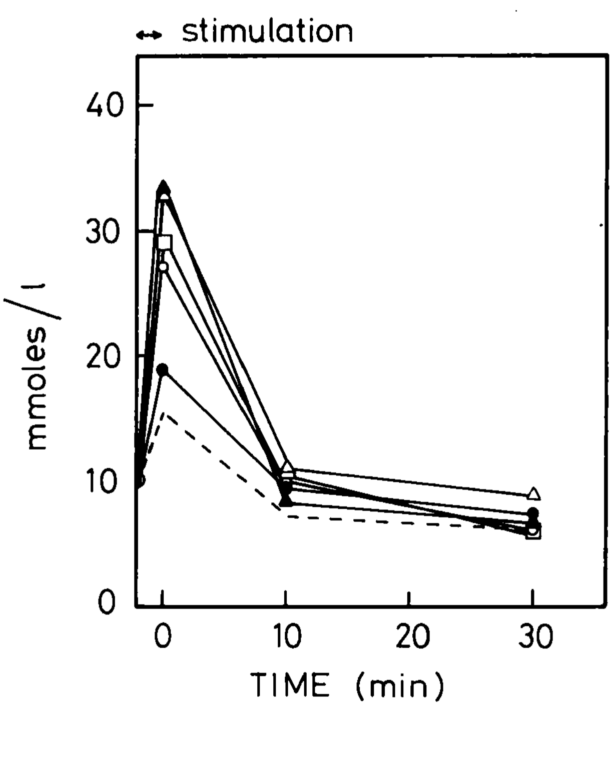

Fig. 6. Salivary Na^{+} -concentrations due to stimulation with sweetened and unsweetened stimulators. Arithmetic means of values from 10 test persons are given. Other details as in Fig. 1,

significantly higher initial decrease in HPO_4^{2-}-values than paraffin or chewing gum base. Subsequently the HPO_4^{2-} values remained at a significantly lower level on sucrose and fructose stimulation as compared to other stimulators. It is to be noted, however, that the pK-values for H_3PO_4 used in the calculations were those of H_3PO_4 in aqueous solutions.

b) *Bicarbonate.* Fig. 4 shows bicarbonatevalues in saliva due to stimulation with sweetened chewing gums, paraffin and chewing gum base. All stimulators caused an initial increase in HCO_3^--levels. The immediate increase in HCO_3^--values due to sorbitol or xylitol chewing gum were almost significantly higher than those to caused by gum base. It is to be noted, that the HCO_3^--values were calculated with an empirical equation for calculation of blood HCO_3^-.

c) *Calcium.* Fig. 5 shows a notable difference in calcium level changes of saliva due to sweetened stimulators as compared to chewing gum base. The gum base stimulation caused an immediate and significant decrease in calcium concentration opposite to the sweetened stimulators which all increased significantly the level of calcium of saliva as compared to resting saliva.

3. *Electrolytes.* Fig. 6 shows the effect of stimulators on the sodium ion content of saliva. The differences between the values obtained immediately after stimulation for sorbitol and xylitol in relation to gum base were significant and those between fructose and sucrose as compared to gum base were almost significant.

A small-scale study on saliva chloride levels, the results not being reported here, suggested that the chloride levels followed the sodium levels in saliva.

4. *Flow rate.* Both xylitol and sucrose increased the flow rate of saliva significantly as compared to chewing gum base (Table I).

Table I. *Effect of xylitol and sucrose on the flow rate of saliva. Arithmetic means of values from 10 test persons are given*

Stimulator	Flow rate (ml × min^{-1}) Mean	S.D.
None	0.64	2.39
Chewing gum base	2.55	1.90
Xylitol chewing gum	3.37	1.65
Sucrose chewing gum	3.39	1.59

DISCUSSION

These pilot-scale observations made in order to examine the acute physico-chemical effects in whole saliva in relation to the presence of various sugars suggested a capacity of the carbohydrates tested to evoke measurable changes with regard to the observed variables. It is thus seen that in addition to the well known facts about differences in the physico-chemical characteristics between unstimulated and stimulated saliva, certain sugar-induced effects were observable. These effects appeared as differences between unsweetened and sweetened stimulators with regard to pH, buffer systems (HCO_3^-, HPO_4^{2-}, Ca^{2+}) and electrolytes (Na^+, Cl^-) of saliva. Generally the unsweetened stimulators tested, paraffin and gum base, seemed to behave in a similar way. Correspondingly, the addition of fructose, sucrose, sorbitol or xylitol to the gum base evoked a trend pattern, characteristic to these substances in general. Stimulation of salivary secretion, due to the influence of the sugars tested resulted in increased concentrations of Na^+-, Cl^--, Ca^{2+}- and HCO_3^--ions in saliva. This was accompanied by a general increase in salivary pH-values, notably in the case of xylitol. The polyols tested, xylitol and sorbitol on one hand, and sucrose and fructose on the other, appeared to behave in an almost similar way. The similar trend evoked by the polyols might be explained by their selective osmotic effects on the oral mucosa.

It is concluded that the stimulation of the salivary secretion, elevation of pH and certain electrolyte concentrations, and increased buffering of saliva as observed in the presence of some of the sugars tested, might exert protective mechanisms in the process of caries development. These protective effects might be of special value in the case of high pH values in the immediate vicinity of tooth surfaces.

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