Course of Changes in Salivary pH-Values after Intake of Different Beverages in Young Children

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Purpose: The aim of the study was to determine possible differences in decrease of pH-values of whole saliva, following the intake of different beverages.

Materials and Methods: Twelve boys and 13 girls (4.9 ± 0.9 years old) participated in this study. A dental examination was performed (dmft). Orange juice (pH = 3.67), instant fennel tea (pH = 7.38), whole milk (pH = 6.84) and mineral water (pH = 5.88) were tested. All beverages were given at the same time of day. Salivary pH and buffering capacities of the beverages were determined with a portable pH-meter. Immediately after intake of a beverage, and 5, 10, 15 and 25 minutes later, whole saliva was collected, and the pH-value was measured again. The statistical evaluation was performed using the Wilcoxon test for paired samples.

Results: Fifteen children had healthy dentitions. Ten subjects had a mean dmft of 1.1 ± 2.3. The mean base salivary pH was 7.09 ± 0.07, without differences between the children with and without dental decay. Mineral water led over the whole period of measurements to a significant rise in salivary pH ($P < 0.05$). Orange juice caused a significant reduction in the salivary pH during the first 10 minutes. After intake of instant tea or milk, significant reductions were found in the period of 5 to 10 minutes. After the intake of instant tea, the reduction was still significant after 15 minutes. During the period of 5 to 10 minutes, the change in pH (ΔpH) in whole saliva differed significantly only between consumption of mineral water and other beverages ($P < 0.01$).

Conclusion: With regard to dental health, a regular consumption of orange juice or sweetened instant teas should be discouraged.

Key words: children, instant tea, milk, orange juice, salivary pH

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In recent decades, an increase in the consumption of soft drinks, including fruit-flavoured drinks, fruit juices and carbonated beverages, has been observed in developed countries (Nielsen et al, 2002). This tendency is reflected in the parents' behaviour in that they offer fruit juices and soft drinks in feeding bottles and cups more and more often. Almost a quarter of the daily energy intake in young children stems from the carbohydrate content of soft drinks (Watt, 2000).

Fruit juices and soft drinks contain sugars and organic acids as natural ingredients, which can not only lead in children to health problems with long-term consequences but also damage dental hard tissues (Rugg-Gunn et al, 1998; Ribeiro et al, 2001; O'Connor et al, 2006).

In the presence of good oral hygiene, a moderate consumption of acidic and sugar-containing beverages has no adverse effects on the teeth of healthy individuals (Tenovuo and Rekola, 1977). However, the frequency and the form of intake of a beverage, e.g. with a glass, straw or feeder cup, the type of sugar and

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Table 1 Timetable of the study

Day 1	Day 2	Day 3	Day 4	Day 5
08:30–10:00 dental examination measurement of SFR and BC of stimulated saliva	08:30–09:00: tooth brushing			
	09:00–10:30: measurement of unstimulated salivary pH before, immediately after and 5, 10, 15 and 25 minutes after beverage intake			
	Test beverage: orange juice	Test beverage: instant tea	Test beverage: whole cow's milk	Test beverage: mineral water

SFR, salivary flow rate; BC, buffering capacity.

acids in the beverage, and the exposure time of the enamel surface to a low pH can influence the extent of damage on the teeth (Millward et al, 1997; Edwards et al, 1998; Hughes et al, 2000; Lussi et al, 2000; West et al, 2000).

The erosive effect of an acidic beverage on the tooth surface depends not only on the contents of the beverage, but also on its temperature, the concentration of the constituent acids, and on the amount of calcium and phosphate, which can influence the buffering capacity (Edwards et al, 1999; Larsen and Nyvad, 1999; Hughes et al, 2000; Azrak et al, 2003). In addition, factors like flow rate, buffering capacity, pH, and calcium and phosphate concentrations of the saliva, as well as frequency of fluid intake can influence the extent of erosion (Hughes et al, 2000; O'Sullivan and Curzon, 2000).

Sweetened instant teas and whole cow's milk are non-erosive beverages, which are also popular among young children and are often placed in nursing bottles. However, such teas have a high cariogenic potential due to their high sucrose content. The low cariogenicity of cow's milk was recently shown (Bowen and Lawrence, 2005).

The aim of this study was to determine in young children the possible differences in decrease of the pH-values (ΔpH) of whole saliva, following the intake of orange juice, instant fennel tea, whole milk or mineral water and relate it to type of beverage and salivary parameters.

MATERIALS AND METHODS

Twenty-five healthy children (12 males, 13 females) with a mean age of 4.9 ± 0.9 years, attending kindergarten, participated in this study. Written informed consent was obtained from the parents of the children. At the first visit, the number of decayed, filled and extracted teeth (dmft) was recorded. In the next step, the flow rate (SFR) of stimulated saliva was determined. The subjects chewed on a piece of paraffin wax (CRT Paraffin Vivadent®, Schaan, Liechtenstein) for one minute, and the saliva that had collected during this period in the oral cavity was swallowed. Afterwards, saliva was collected for 5 minutes in a 30 ml beaker. The total amount of saliva was measured and the flow rate was expressed as ml/minute. The buffering capacity of stimulated saliva was determined employing the CRT Buffer Test (Ivoclar Vivadent®, Schaan, Liechtenstein). Saliva was applied to the test strips, and the colour of the test area was compared with the colour pattern supplied by the manufacturer. The colour blue indicates a high (pH ≥ 6.00), green a medium (pH = 4.5–5.5), and yellow a low (pH = ≤ 4) buffering capacity.

The test beverages used in this study were as follows: orange juice (from concentrate; sugar content in 100 ml: 8.8 g), instant tea for children (fennel; sugar content in 100 ml: 3.0 g glucose, 1.6 g sucrose), whole cow's milk (lactose content in 100 ml: 4.8 g), mineral water. These beverages were tested on all children in such a way that on four subsequent days a different beverage was given to the child (Table 1). In order to account for the circadian rhythm, the beverages were given to each child at the same time of day.

On each day, at least 60 minutes after breakfast and 15 minutes after tooth cleaning (toothbrush: Oral B Stages 2; toothpaste: Elmex for children), which was supervised but not assisted, the pH of unstimulated whole saliva (base value) was measured. Whole saliva was collected in small plastic tubes with the help of plastic funnels. The pH of saliva and of the tested beverages was determined with a portable pH-meter equipped with a microelectrode (Novodirect, Kehl, Germany). The portable pH-meter had an accuracy of ± pH 0.01. It was calibrated each day prior to the first measurement with buffering solutions from IUPAC (Radiometer, Copenhagen, Denmark). A test beverage (50 ml) was given to the children in special cups (Nuk®, Zeven, Germany), to ensure a uniform intake of the fluids. Immediately after the intake of a beverage (0), and

5, 10, 15 and 25 minutes later, whole saliva was collected as described above and the pH-value was measured again. For measuring the salivary pH, only a small amount (about 200 µl) of saliva was necessary.

In addition, the titratable acids, and the calcium and phosphorus contents of the beverages were measured. The total acids were determined by titration with 0.2 N NaOH and subsequent measurements of the resulting pH values. The phosphorus and calcium contents of the beverages were determined spectrophotometrically (Shimadzu UV-1202/UV-VIS, Shimadzu Europe, Duisburg, Germany) by means of a colorimetric assay, using test kits (Sigma®, Deisenhofen, Germany).

The statistical evaluation of the data was performed using the SPSS program at the Institute for Medical Biometrics, Epidemiology and Computer Sciences, Johannes-Gutenberg University, Mainz, employing the Wilcoxon test for paired samples.

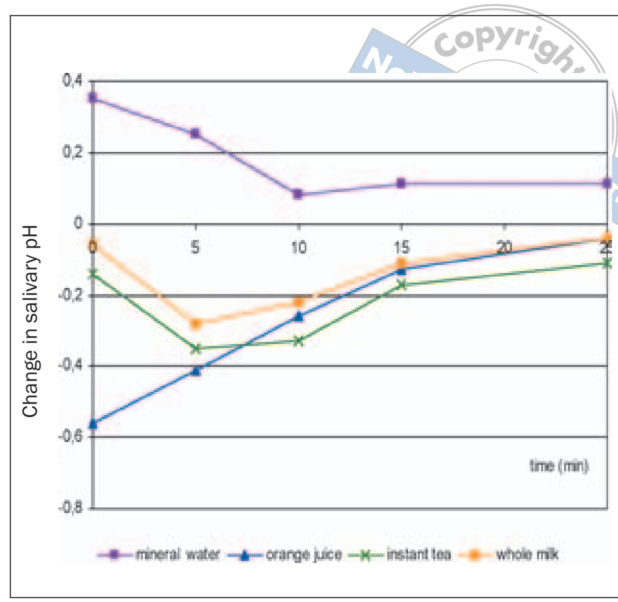

Fig 1 The course of changes of salivary pH after intake of the test beverages over a period of 25 minutes (n = 25).

RESULTS

Fifteen children had caries-free dentitions (dmft = 0), while the dmft values of the remaining 10 subjects ranged from 1 to 7 with a mean of 1.1 ± 2.3. The mean base value of the pH of whole saliva was 7.09 ± 0.07. There were no significant differences in the levels of salivary pH in children with or without dental decay. The flow rates of stimulated saliva could only be determined in 15 children: 40% had a high (>1 ml/min), 53% a medium (0.7-1 ml/min) and only one child had a low salivary flow rate (< 0.7 ml/min). The median value for SFR was 1 ml/min. In 28% of the subjects (n = 15), saliva had a medium and in 72% a high buffering capacity; in none of the children was it low. No correlations could be found between the dmft values of the children or the buffering capacity of their saliva and the initial pH changes of the whole saliva after intake of the tested beverages.

The mineral water led immediately after intake to a statistically significant ($P < 0.05$) rise in salivary pH, which remained above the base salivary pH over the whole period of measurements (Fig 1 and Table 2). Orange juice caused a statistically significant reduction in the salivary pH-values during the first 10 minutes. Immediately after the intake of instant tea or milk, the reductions in the salivary pH-values were not significant; however, in the period of 5 to 10 minutes after consumption of these beverages, significant reductions were found. After intake of instant tea, the reduction was still significant even after 15 minutes.

Except for mineral water, the consumption of the beverages decreased the pH of the whole saliva significantly only in the period of 5 to 10 minutes. Therefore, this period was chosen for the statistical evaluation of the differences of ΔpH of whole saliva after intake of the test beverages. During this period, the course of the changes of ΔpH in whole saliva differed significantly ($P < 0.01$) between mineral water and the other beverages (Table 3). No significant differences were found between juice, instant tea and milk.

Milk contained considerably more calcium (1.55 mg/ml) and phosphorus (0.48 mg/ml) than the other beverages (Table 4). Milk and instant tea contained no acids, while orange juice had the highest buffering capacity concerning the acidic pH range as well as the highest amount of titratable acids (Fig 2).

DISCUSSION

In the present study, 60% of the children had caries-free dentitions. Epidemiological studies from the same area found caries-free dentitions and comparable dmft levels in 50 to 57% of 3- to 6-year-old children (Franzen, 2000). The mean pH of unstimulated whole saliva in the present study was 7.08, which is comparable with the findings of previous studies (Andersson et al, 1974; Azrak et al, 2003).

In the present study, only 15 children were able to collect saliva according to the instructions. Andersson

Table 2 The median ΔpH of whole saliva at the various times of measurement after intake of test drinks (n = 25)

Time of measurement	Median ΔpH Mineral water	Orange juice	Instant tea	Whole milk
Immediately after intake	0.35*	-0.56*	-0.14	-0.06
After 5 min	0.25*	-0.41*	-0.35*	-0.28*
After 10 min	0.08	-0.26*	-0.33*	-0.22*
After 15 min	0.11*	-0.13	-0.17*	-0.11
After 25 min	0.11*	-0.04	-0.11	-0.04

*Statistically significant P < 0.05

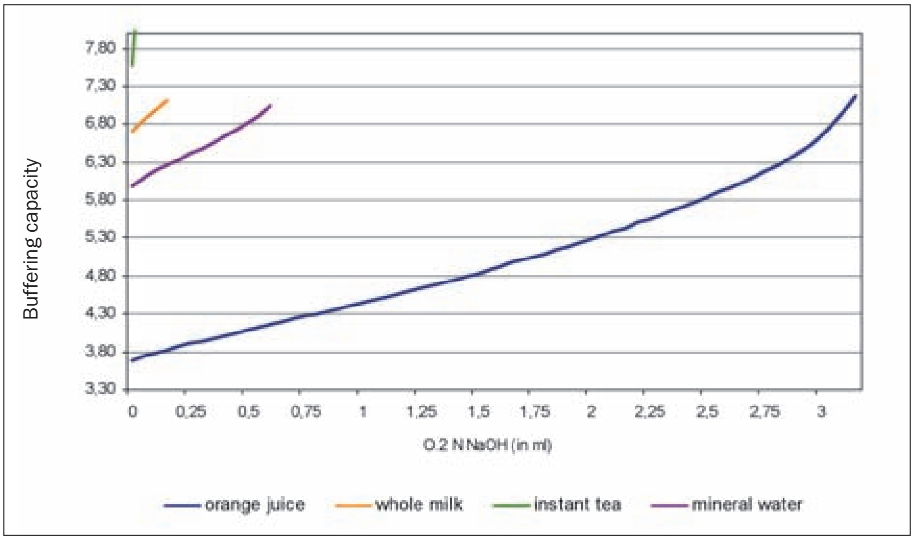

Fig 2 Buffering capacities of the test beverages.

et al (1974) considered the possibility that children under 7 years of age might swallow the saliva instead of collecting it. The median value for stimulated salivary flow rate was 1 ml/min. Previous studies found in comparable age groups similar values for stimulated salivary flow rate (Azrak et al, 2003; Sanchez et al, 2003). None of these children from the present study had a low buffering capacity of stimulated saliva. This is supported by the investigation from Sanchez et al (2003), who found in only 8% of 4- to 10-year-old children a low buffering capacity of stimulated saliva. They found in 47% of the children of the same age group a medium buffering capacity, whereas only 28% of the children from the present study had a medium buffering capacity. However, Kavanagh and Svehla (1998) demonstrated a high variability in buffering capacity in children, when its measurements were repeated in the same individuals monthly over a period of 11 months. No correlation of buffering capacity to salivary pH changes after intake of sugar- and acid-containing beverages could be detected in the present study, although the salivary buffering capacity was described as an important protective factor in oral health by neutralising acids in saliva (Edgar, 1992). The missing correlation in this study could be attributed to the low number of the subjects.

In the present study, different salivary pH changes were observed immediately after intake of the tested beverages and in the period of 25 minutes, corresponding to their composition. Oral clearance of sugar-containing beverages can take up to 30 minutes, with very high salivary levels within the first 10 minutes (Meurman et al, 1987). The salivary properties, bacterial composition of saliva and residual plaque, but also type of acids and sugars and their concentrations in the beverages, and their baseline pH-values influence the changes in salivary pH. The inherent buffering capacity of a beverage competes with the salivary

Table 3 The median ΔpH differences of whole saliva 5–10 minutes after the intake of different test drinks and the *p* values (n = 25)

Beverages		Median of differences	*P* value
Orange juice	Mineral water	-0.41	0.0001*
Orange juice	Instant tea	0.04	0.5825
Orange juice	Whole milk	-0.03	0.3709
Instant tea	Mineral water	-0.42	0.0001*
Instant tea	Whole milk	-0.15	0.1620
Whole milk	Mineral water	-0.35	0.0001*

*Statistically significant $P < 0.05$

Table 4 Calcium and phosphorus contents and buffering capacity (in ml 0.2 N NaOH) of the test beverages

Beverage	pH	Buffering capacity ↑pH 5.5	↑pH 7.0	Phosphorus mg/ml	Calcium mg/ml
Orange juice	3.67	2.20	3.15	0.1653	0.0770
Mineral water	5.88	-	0.60	0.0002	0.1001
Whole milk	6.84	-	0.15	0.4826	1.5500
Instant tea	7.38	-	0.05	0.0013	0.0089

buffering systems. The salivary pH changes, induced by natural buffering systems, can become compensated by the buffering capacity intrinsic to the beverages, so that lower pH-values in saliva and in plaque are maintained longer (Edwards et al, 1999; Larsen and Nyvad, 1999; Lussi et al, 2000). In the present study, the children brushed their teeth prior to the test periods to remove the bacterial plaque; the tooth cleaning was only supervised, but not assisted. In this age group, tooth brushing and therefore complete removal of plaque cannot be sufficiently performed without assistance by an adult (Koroluk et al, 1994). In addition to residual plaque, acid-forming bacteria are also present on the tongue and mucosa as well as in saliva (Tanner et al, 2002).

Mineral water had, as in other studies, a very low buffering capacity (Edwards et al, 1999; Azrak et al, 2003). Despite an initial pH of 5.88, it caused an increase in the salivary pH after intake, and the positive ΔpH remained significant almost for the whole observation period. In the present study, the mineral water had a strikingly salty taste and could therefore have led to a more pronounced stimulation of salivary flow. A gustatory stimulation, as described by Dawes and Watanabe (1987) and Edgar (1992) can explain the rise of the pH-value in whole saliva after the intake of mineral water, which had a considerably lower pH than the base value of salivary pH in the subjects.

The orange juice utilised in the present study showed a higher buffering capacity compared with the other tested beverages, verifying the findings of previous investigations (Larsen and Nyvad, 1999). The course of the curve for the salivary pH changes within the 25 minutes for orange juice was comparable to the course of its titration curve. Immediately after intake of orange juice, the salivary pH dropped to the lowest level in the observation period and then increased continuously. Like ‘salty’, ‘sour’ and ‘sweet’ are also stimulants for salivary flow (Dawes and Watanabe, 1987; Edgar, 1992), but ‘sweet’ is a poorer stimulant than ‘sour’. Orange juice without added sugar tastes rather sour and can stimulate the salivary flow in children.

Both instant tea and milk led to similar changes in salivary pH after intake. They both had similar sugar concentrations but contained different types of sugar: the instant tea had glucose and sucrose as additives, and milk has lactose as a natural constituent. Their initial pH was close to 7, and their sugar concentration was almost half that of orange juice. Thus, significant decreases in pH were observed not immediately after intake of the beverages but 5 minutes later, possibly after acid formation by residual oral bacteria. In contrast to milk and orange juice, 15 minutes after intake of instant tea the difference in the salivary pH compared with the initial value was still significantly lower, and the salivary pH increased at the end of the observation period only by a small amount. Another explanation could be that neither milk nor instant tea had any distinctive taste and, therefore, SFR was not stimulated by these beverages.

Milk contains not only lactose but also caries-protective components like calcium, phosphorus, glycoproteins, proteoglycans and lactophorins, and especially cow's milk is accepted as a noncariogenic beverage (Grenby et al, 2001; Bowen and Lawrence, 2005). However, cariogenicity depends not only on the type of constituent sugar but also on the form, frequency and length of the intake of a beverage (Marshall et al, 2007). In conclusion, with regard to dental health, only non-carbonated or carbonated mineral water can be recommended for children without reservation. Despite the caries-protective components, because of the lactose content, milk can only be recommended for limited consumption. Regular consumption of orange juice or sweetened instant teas should be discouraged altogether.

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