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ORIGINAL ARTICLE

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Buffer capacity, pH, and flow rate in saliva of children aged 2–60 months with Down syndrome

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Abstract This study measured the flow rate, pH, and buffer capacity of saliva from children with Down syndrome aged 2–60 months. We compared 25 children with Down syndrome with 21 children without Down syndrome. The whole saliva was collected under slight suction and the pH and the buffer capacity were determined using a digital pHmeter. The buffer capacity was measured by titration with 0.01 N HCl. The Down syndrome group demonstrated higher buffer capacity than the control group in the pH ranges of 6.9–6.0, 5.9–5.0, and 4.9–4.0. The flow rate was low in the Down syndrome group. Boys from the Down syndrome group demonstrated higher buffer capacity in the pH ranges 6.9–6.0, 5.9–5.0, and 4.9–4.0 than controls, while girls from the Down syndrome group showed no difference compared with controls. Girls with Down syndrome demonstrated significant difference only in the range of pHi–pH 7.0. Conclusion. These data suggest that the Down syndrome persons present a better buffer capacity, supporting the results observed in several studies which found the low dental caries in persons with Down syndrome.

Keywords Buffer capacity · Saliva · Flow rate · pH · Down syndrome

Introduction

Down syndrome is a genetic disorder caused by trisomy of chromosome 21 [1]. The incidence of the Down syndrome is 1 in 600–700 live births [2, 3]. Several systemic manifestations such as cardiac anomaly, recurrent infections, and hypothyroidism are linked to the Down syndrome. In relation to the oral manifestations of this syndrome some authors have reported a relationship between the Down syndrome and low prevalence of dental caries and high prevalence of periodontal disease [4]. Some differences in salivary components of persons with Down syndrome compared to individuals of a control group have been reported [5]. There are conflicting results in the salivary pH of individuals with Down syndrome; no difference [6, 7, 8], higher pH [9], or lower pH [10] was observed. Previous report from our laboratory [11] showed that the pH of boys (aged 6–10 years) with Down syndrome was lower than the pH of boys from the control group. On the other hand, no difference was found in the pH of girls with Down syndrome and girls of the control group. The flow rate and buffering capacity of saliva are important protective factors in oral health [12]. In saliva there are three major systems contributing to the buffer capacity, bicarbonate, phosphate, and protein buffer systems [13]. An interrelationship between pH, buffer capacity, and flow rate has been reported [14, 15, 16]. A reduced salivary flow rate in persons with the Down syndrome has been described [10, 11, 17]. On the other hand, no difference in buffer capacity of saliva from individuals with Down syndrome aged 7–22 years than the control group has been observed [8].

To our knowledge this is the first study evaluating the pH and the buffer capacity of saliva from children with Down syndrome aged below 5 years. Thus the aim of this investigation was to determine the pH, buffer capacity, and flow rate of whole saliva in children with Down syndrome aged between 2 and 60 months compared with a matched control group.

Materials and methods

This study included 46 children: 25 with Down syndrome and 21 without. The children with Down syndrome were selected from those attending the "Associação de Pais e Amigos dos Excep-

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cionais, APAE- São Paulo" and the Darcy Vargas Children Hospital of the city of São Paulo. The control group was selected from children attending the Dental Clinic of Madre Rita Institute. None of the children had any systemic diseases and did not take any medication for at least 15 days before saliva collection. The protocol of this study was approved by the Ethics Committee of the Faculty of Dentistry of the University of São Paulo. After being informed of the objectives of the investigation, each parent or guardian provided a written consent for the child to participate in the study.

The saliva sample were collected between 9:00 and 10:00 a.m. to minimize the circadian rhythms effects, 2 h after the last meal by a slight suction through a soft plastic catheter. No stimulation was used, although the presence of the soft catheter might have provided a slight stimulation. The saliva collected during the first 10 s was discarded; saliva was then collected for 2 min so that the initial flow rate could be calculated. After this period the sampling continued until 3.5 ml saliva was collected. During the collection period the children remained comfortably seated on the lap of their parents or guardians in a well-ventilated and well-lit room. If a child cried during sampling, its sample was excluded. Immediately after saliva collection both the pH and the buffer capacity were determined using a portable pHmeter (Digimed DU-2). The buffer capacity was determined by titration using 1 ml saliva, with 0.01 N HCl and after each addition of the acid the change in pH was monitored up to pH 4.0.

For statistical analysis the data are presented as a mean ±SD. Student's *t* test was used to determine differences between the mean salivary values of children with Down syndrome and those of the control group.

Results

The data of this study are presented in the Tables 1 and 2. The buffer capacity of whole saliva was analyzed taking ranges of pH. The volume of acid added to the saliva was calculated for each interval considered. Table 1 shows significant difference in flow rate and in buffer capacity of saliva in the pH ranges 6.9–6.0, 5.9–5.0, and 4.9–4.0. Table 2 compares the results obtained for boys and girls with Down syndrome with controls. The salivary flow rate was lower in both boys and girls with Down syndrome than in controls. The buffer capacity of saliva in the range pHi–pH 7.0 was higher in girls with Down syndrome than controls. On the other hand, boys with Down syndrome showed a higher buffer capacity of saliva in the pH ranges of 6.9–6.0, 5.9–5.0, and 4.9–4.0 than controls, while no difference statistically significant was observed between the buffer capacity of saliva from girls with Down syndrome and girls from the control group.

Table 1 Age, weight, flow rate, pH, and volume of acid 0.01 N HCl used in pH ranges of whole saliva from children aged 2–60 months with Down syndrome and controls (*parenthesis* number of samples)

	Controls (*n*=21)	Down syndrome (*n*=25)
Age (months)	28.40±13.21	29.56±11.98
Weight (kg)	13.01±1.81	13.20±1.78
Flow rate (ml/min)	0.56±0.18	0.34±0.14*
pH	7.42±0.40	7.35±0.33
Buffer capacity (ml acid/ml saliva)		
pHi–7.0	0.41±0.16	0.43±0.20
pH 6.9–6.0	0.71±0.22	0.95±0.35*
pH 5.9–5.0	0.52±0.30	0.77±0.28*
pH 4.9–4.0	0.53±0.37	0.78±0.29*

* $P<0.05$ (Student's *t* test)

Discusson

A negative correlation between the buffer capacity of saliva and dental caries has been reported [14, 18, 19]. Several methods to determine the buffer capacity of saliva are available, including colorimetric and electrometric methods. In this study we used the titration method with 0.01 N HCl solution, monitoring the changes in pH at each acid addition, so that it was possible to analyze the buffer capacity at different pH range. The Van Slyke formula, $\beta=\Delta Ca/\Delta$ pH, where β is the buffer capacity, ΔCa denotes the amount (in gram equivalent per liter) of acid added to the saliva at each pH range, and ΔpH the change in pH induced by the addition of acid, was used in the present investigation. For practical purposes we express the buffer capacity in volume (ml) of the acid added to 1 ml saliva in the pH range considered instead of equivalents of H^+. Information on the effect of diseases on the salivary buffer capacity is scarce. In individuals with cystic fibrosis the salivary buffer capacity is reported to be higher than in saliva from controls [20], possibly due to higher phosphorus level [21]. In saliva of individuals with insulin-dependent diabetes the buffer capacity is reported to be higher than control [22], but this was not confirmed by Tenovuo et al. [23].

Table 2 Age, weight, flow rate, pH, and volume of acid 0.01 N HCl used in pH ranges of whole saliva comparing boys and girls of the control and Down syndrome groups (*parenthesis* number of samples)

	Boys		Girls	
	Control (*n*=10)	Down (*n*=11)	Control (*n*=11)	Down (*n*=14)
Age (months)	27.14±10.61	27.90±11.52	31.13±10.01	33.28±12.38
Weight (kg)	12.87±2.56	12.13±3.24	13.16±1.49	14.37±2.71
Flow rate (ml/min)	0.63±0.22	0.37±0.14*	0.47±0.27	0.32±0.19*
pH	7.48±0.43	7.37±0.38	7.34±0.36	7.25±0.24
Buffer capacity (ml acid/ml saliva)				
pHi–pH 7.0	0.47±0.17	0.32±0.15	0.35±0.13	0.54±0.18*
pH 6.9–6.0	0.66±0.19	1.04±0.34*	0.76±0.23	0.89±0.36
pH 5.9–5.0	0.38±0.15	0.80±0.27*	0.66±0.35	0.74±0.29
pH 4.9–4.0	0.38±0.22	0.86±0.28*	0.67±0.43	0.71±0.29

* $P<0.05$ (Student's *t* test)

In our study, taking the results from boys and girls together, the buffer capacity of saliva in the pH range 6.9–6.0 of persons with Down syndrome was higher than in controls. In this pH range the amount of acid consumed was higher than the amount consumed in the other ranges. This may be explained by the fact that pK values of $H_2PO_4^-$ and H_2CO_3 are around pH 6.8 and 6.1 in saliva, respectively, depending on the ionic strength. Both H_2CO_3 and $H_2PO_4^-$ are components of the buffer system of saliva.

Yarat et al. [8] working with individuals aged 7–22 years found no difference in the buffer capacity comparing the Down syndrome and control. They suggested a relationship between the low dental caries prevalence and the high concentration of bicarbonate found in persons with Down syndrome groups. However, these authors determined the pH and buffer capacity of saliva by using a colorimetric method, that is, with a pH indicator paper. In our investigation, using a pHmeter and considering the sexes separately, we found that boys with Down syndrome presented higher buffer capacity in all pH ranges studied with the exception of pHi–pH7.0. On the other hand, the buffer capacity of saliva from girls with Down syndrome showed a statistically significant difference only in the range pHi–pH7.0. In the other ranges studied, although showing higher values than controls, the differences were not statistically significant.

In this work we found a reduction in the flow rate of about 41% and 32% for boys and girls with Down syndrome, respectively compared with the controls. No difference in sexes was observed, comparing boys and girls of the control groups with boys and girls with Down syndrome groups, respectively. Many studies have shown sex differences in salivary flow rate, although not always to the level of statistical significance [24, 25, 26, 27]. Dezan et al. 2002 [28] observed in children aged, 18, 30, and 42 months no statistically significant sex differences in the initial flow rates in any age group. Winer et al. [29] reported higher sodium, calcium, and bicarbonate concentration in saliva of Down syndrome group than control. Cutress [5] reported high concentrations of calcium, potassium, and phosphorus, but not sodium in the whole saliva of persons aged 6–23 years with Down syndrome compared with a control group. However, in parotid saliva sodium was the only electrolyte to show higher concentration in the Down syndrome group than controls. Stabholz et al. [7] working with individuals aged 8–13 years observed lower salivary pH in the Down syndrome group than controls; however, they found no differences between Down syndrome and a mentally retarded group. Shapira et al. [6] found no differences in the pH of saliva from adults (20–48 years) comparing the Down syndrome with the controls.

Conclusion

These findings clearly show, using an electrometric method, that the saliva from children aged 2–60 months with Down syndrome have higher buffer capacity than children from a control group.

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