



## Evaluation of masticatory and salivary parameters in preschool children with different morphological occlusion

Avaliação dos parâmetros mastigatórios e salivares em crianças pré-escolares com diferente oclusão morfológica

Maria Carolina Salomé MARQUEZIN<sup>1</sup>, Clizia Genoveze GAUCH<sup>1</sup>, Camila Alvarez DE SIQUEIRA<sup>1</sup>, Fernanda Yukie KOBAYASHI<sup>1</sup>, Fernando Luiz Affonso FONSECA<sup>2</sup>, Paula Midori CASTELO<sup>2</sup>

1 - Department of Pediatric Dentistry - Piracicaba Dental School - University of Campinas (UNICAMP) - Piracicaba - SP - Brazil.

2 - Department of Pharmaceutical Sciences - Federal University of São Paulo (UNIFESP) - Diadema - SP - Brazil.

### ABSTRACT

**Objective:** To better understand the relationship between malocclusion and masticatory and salivary functions in the primary dentition, the aim was to evaluate the differences in masticatory performance (MP), facial morphology (FM), maximal bite force (BF) and salivary parameters in 65 preschool children with normal occlusion (n = 22), functional posterior crossbite (n = 20) and anterior open bite (n = 23), and to explore the relationship between these variables. **Material and Methods:** MP, FM and BF were assessed by sieving method, anthropometry and gnatodynamometer, respectively. Stimulated (SS) and unstimulated (US) saliva flow and composition were measured by automated colorimetric technique. Data were analyzed using ANOVA/Kruskal-Wallis, t-test/Wilcoxon and Pearson/Spearman correlation test. The relationship between SS flow rate and the independent variables were assessed using multiple linear regression. **Results:** The lower face dimension was smaller in crossbite-group, and a decreased BF in the crossbite-side of the dental arch was observed. BF correlated positively with intergonial width in open bite group. In malocclusion groups, better MP correlated with bizigomatic and intergonial widths. US flow rate was lower in crossbite-group and total protein concentration differed between SS and US saliva only in the crossbite-group, being lower in US. Amylase activity was higher in SS than US in all groups. SS flow rate related positively with age and negatively with the presence of malocclusion. **Conclusion:** In young subjects, significant correlations were found between masticatory parameters and facial dimensions; in addition, some important masticatory and salivary parameters differed between children with different

### RESUMO

**Objetivos:** Para compreender melhor a relação entre má oclusão e as funções mastigatória e salivar na dentição decídua, objetivou-se avaliar as diferenças na performance mastigatória (PM), morfologia facial (MF), força de mordida (FM) e parâmetros salivares em 65 pré-escolares com oclusão normal (n = 22), mordida cruzada funcional posterior (n = 20) e mordida aberta anterior (n = 23) e explorar a relação entre essas variáveis. **Materiais e Métodos:** PM, MF e FM foram avaliados pelo método de peneiragem, antropometria e gnatodinamômetro, respectivamente. O fluxo salivar estimulado (SE) e não estimulado (SNE) e composição salivar foram mensurados por técnica colorimétrica automatizada. Os dados foram analisados utilizando-se ANOVA/Kruskal-Wallis, teste-t/Wilcoxon e teste de correlação de Pearson/Spearman. A relação entre fluxo salivar e as demais variáveis independentes foi avaliada por regressão linear múltipla. **Resultados:** A dimensão facial inferior foi menor no grupo com mordida cruzada e observou-se menor FM no lado cruzado do arco dentário. A FM correlacionou-se positivamente com a largura intergoniaca no grupo com mordida aberta. Nos grupos com má oclusão, uma melhor PM relacionou-se com as larguras bizigomática e intergoniana. O fluxo de SNE foi menor no grupo com mordida cruzada e a concentração de proteína total diferiu entre SE e SNE apenas no grupo com mordida cruzada, sendo menor na SNE. A atividade da amilase foi maior na SE em relação à SNE em todos os grupos. O fluxo de SE mostrou relação positiva com a idade e negativa com a presença de malocclusão. **Conclusão:** Em crianças de pouca idade foram encontradas correlações significativas entre os parâmetros mastigatórios e as dimensões faciais; além disso, parâmetros mastigatórios e salivares importantes diferiram entre crianças com diferentes tipos de

morphological occlusions, hence the importance of the early assessment of these parameters.

## KEYWORDS

Bite force; Dental occlusion; Masticatory system; Primary dentition.

oclusão, mostrando a importância da avaliação precoce destes parâmetros.

## PALAVRAS-CHAVE

Força da mordida; Oclusão dentária; Sistema mastigatório; Dentição decídua.

## INTRODUCTION

Chewing is the first oral step in the process of digestion, when food is prepared for swallowing and further processing. The reduction of food particles is determined by a multifactorial process, which depends on factors as bite force (BF), the coordination of the masticatory muscles and the number of occluding pairs of teeth [1]. In the oral cavity, it is subjected to several mechanical and chemical processes, being diluted and broken down by saliva, forming a bolus to finally be swallowed. Components of saliva participate and facilitate the motor functions of chewing, swallowing and speech, as well as the functions of the sensory perception of flavor, taste and texture of food in the oral cavity [2], contributing to food acceptability [3].

The primary occlusion may improve or worsen as the child moves from primary to mixed and permanent dentition. Occlusion is an important predictor in craniofacial development, as to reach a normal growth, it is necessary to enable a proper morphological and functional development of the jaws. Malocclusions, when present, may alter some stomatognathic functions, such as speech and chewing [4].

The early assessment of masticatory apparatus seems to be of great importance, and the hypothesis to be tested was whether the malocclusion can impact the masticatory and salivary functions in young children. Thus, the purpose of this study was to evaluate the differences in masticatory and salivary

parameters in preschool children with different morphological occlusions and to explore the relationship between these variables.

## MATERIAL AND METHODS

### *Sample selection and clinical examination*

A convenience sample was selected from the Pediatric Dentistry Clinic, Department of Pediatric Dentistry, University of Campinas (UNICAMP) and consisted of 65 subjects with primary dentition divided into three groups: normal occlusion ( $n = 22$ ; 14 boys and 8 girls, mean age  $56.36 \pm 8.59$  months), unilateral functional posterior crossbite ( $n = 20$ ; 8 boys and 12 girls, mean  $56.00 \pm 6.46$  months), and anterior open bite ( $n = 23$ ; 2 boys and 11 girls, mean  $56.74 \pm 10.72$  months), diagnosed according to Foster and Hamilton [5] and Keski-Nisula et al. [6] The children and their parents/guardians consented to participate in the study, which was approved by the Ethics Committee of Piracicaba Dental School (Protocol No. 004/10).

Sample size was calculated from results found by Sonnesen et al. [7], which examined the differences in maximal BF between children aged 7-13 years with unilateral crossbite and neutral occlusion. Considering a power of the test = 0.80 and alpha level = 0.05, 15 subjects in each group would be sufficient to perform such evaluation. Considering the secondary endpoint, sample size was calculated from results found in a previous study [8] which evaluated the correlation between masticatory performance

(MP) and facial morphology in adolescents with different morphological occlusions. Considering the correlation coefficients obtained between masticatory performance (MP) and bigonial ( $r = -0.343$ ) and bizygomatic width ( $r = -0.391$ ), 65 subjects in total would be sufficient.

Body weight and height were also determined and the body mass index (BMI) was calculated as:  $BMI = \text{Kg}/\text{m}^2$ .

The inclusion criteria were the presence of complete primary dentition. The exclusion criteria were: systemic disturbances in general, ingestion of medicines that could interfere directly or indirectly with muscular activity and that potentially affect salivary function, uncooperative behavior, pain of dental origin, premature tooth loss, anomalies of shape, number, structure and/or changes that might compromise the mesiodistal tooth dimensions, tooth decay, trauma, and soft tissue abnormalities.

#### *Masticatory performance evaluation*

MP was assessed by the determination of the individual capacity of fragmentation of a test-material: Optocal [9]. The subjects received 10 cubes (Figure 1), which were chewed for 20 mastication cycles. After drying, the particles were removed from the paper filter, weighed and passed through a series of 10 granulometric sieves with meshes ranging from 5.60 mm to 0.71 mm, connected in decreasing order and closed with a metal base (Figure 2). The particles retained on each sieve were removed and weighed on an analytical scale with a precision of 0.001 g. The distribution of the particles by weight was described as “b” by cumulative function (Rosim–Ramler equation). The degree of fragmentation of the material was then given by the median particle size ( $X_{50}$ ), which is the aperture of a theoretical sieve through which 50% of the particles could pass [8]. The evaluations were performed in replicates and for purposes of final results, there were chosen

the replication which showed the smallest loss after sieving.

#### *Facial Morphology*

The evaluation of facial morphology was performed by anthropometry, using a caliper (caliper Bone aluminium 240 mm Cescorf, Brazil) (Figure 3). For each subject, seven craniometric points located by palpation/inspection were marked with pencil directly on the skin: n, nasion; zy, zygion (left/right); go, gonion (left/right); sn, subnasale; gn, gnathion [10]. The following dimensions were then determined by the same examiner (CAS): face height (n-gn), nose height (n-sn), lower face (sn-gn) [11], mid-facial width (zy-zy), and bigonial width (go-go) [12].



**Figure 1** - Test-material used to determine the individual capacity of fragmentation (Optocal).



**Figure 2** - Ten granulometric sieves in which the particles of Optocal are retained.



**Figure 3** - Caliper used in the evaluation of facial morphology.

### ***Maximal Bite Force***

Maximum unilateral BF was assessed with a digital gnathodynamometer (Digital Dynamometer DDK, Kratos Equip. Ind. Ltda., Cotia, Brazil), using a fork strength of 10 mm connected to a digital device. The children were instructed to bite the fork as forcefully as possible in each side of the dental arches (left/right). The recordings of each side were performed two times at the level of the first permanent molars. The maximum value for each side was considered, with an accuracy of 0.01 N.

### ***Evaluation of salivary flow and biochemical analysis***

Stimulated (SS) and unstimulated (US) saliva were collected in the morning (CGG),

with all subjects having refrained from eating, drinking or brushing their teeth for a minimum of 2h before collection. Subjects were trained to avoid swallowing saliva and asked to lean forward and spit all the US saliva produced for five minutes into a cooled tube, through a glass funnel. After this, SS saliva was collected by subjects chewing on 0.3 g of an inert and tasteless material (Parafilm, Merifield, USA), for approximately 70 cycles/min [13]. SS and US flow rates were defined as the weight of saliva secreted per min (g/min). Next, samples were centrifuged at 11000 g and 4 °C, fractionated and frozen for further analyses (-80 °C).

Biochemical analyses were performed in duplicate, using automated technique (Flexor E6002-190 Clinical Chemistry Analyzer, Vital Scientific, Dieren, Switzerland), at the Clinical Analyses Laboratory of ABC Medical School (FMABC, Santo André, SP, Brazil). The total protein (TP) concentration (mg/dL) was determined by the Bradford method [14] in pure saliva (Proti U/LCR, Wiener Laboratories, Rosario, Argentina). Salivary amylase activity (AMY) was measured using enzymatic method, with a commercially assay kit in diluted saliva 1:25 (Amylase SL 50 mL, ELI Tech, France) and expressed as U/L.

### ***Measurement errors***

For assessment of method error of the studied variables, the intraclass correlation coefficient (ICC) were calculated from data obtained from 15 subjects not included in the studied sample, on two separate occasions at an interval of 14 days (BioEstat 5.0; Mamirauá, Belém, PA, Brazil) (Table 1). ICCs for MP were obtained correlating the particles of test-material retained on each sieve from two consecutive assessments, and the values obtained for “X50” and “b” were 0.63 and 0.55, respectively, showing a moderate to substantial agreement [15]. ICCs for the other variables ranged from substantial to almost perfect agreement.

**Table 1** - Method error of the studied variables assessed by means of intraclass correlation coefficients (ICC), using data from 15 volunteers

Variable	First evaluation	Second evaluation	ICC
	Mean (SD)	Mean (SD)	
X <sub>50</sub>	4.23 (1.17)	4.00 (0.91)	0.63
b	1.77 (0.73)	1.92 (0.49)	0.55
n-sn	5.70 (0.39)	5.65 (0.40)	0.88
sn-gn	5.27 (0.55)	5.27 (0.49)	0.94
go-go	12.11 (0.89)	12.07 (0.92)	0.96
n-gn	10.95 (0.81)	10.92 (0.80)	0.95
zy-zy	13.19 (0.75)	13.16 (0.67)	0.96
Right BF	321.68 (116.12)	318.47 (103.91)	0.77
Left BF	289.84 (114.43)	294.63 (114.55)	0.72

SD, standard deviation; ICC, intraclass correlation coefficient; X<sub>50</sub>, median particle size of test-material; b, distribution of the particles in different sieves; n-sn, nasion-subnasale; sn-gn, subnasale-gnathion; go-go, gonion-gonion; n-gn, nasion-gnathion; zy-zy, zygonion-zygonion; BF, bite force.

### Statistics

Statistical analysis was performed using BioEstat 5.0 (Mamirauá, Belém, PA, Brazil) and SigmaPlot 11 (Systat Software Inc., Chicago, EUA) with a 5% significance level. Descriptive statistics analysis (mean, standard deviations, medians, interquartile ranges) and Kolmogorov-Smirnov normality test were applied. The proportion of girls and boys among groups was verified by means of the Chi-square test.

The comparison between groups for the variables under study was performed using ANOVA (and Tukey post-test) or Kruskal-Wallis test (and Dunn post-test), where appropriate.

The intragroup comparisons were evaluated using paired t-test or Wilcoxon. Correlations between BF, MP and facial dimensions were assessed using Pearson or Spearman correlations tests.

A multiple linear regression model with stepwise backward elimination was used to verify the relationship between SS flow rate, as the dependent variable, and the following independent variables: gender, age, presence of malocclusion, BMI, BF, and AMY and TP concentrations.

## RESULTS

The descriptive statistics of the demographic data and values of facial dimensions are shown in Table 2. According to the results found, age, BMI and the proportion of boys and girls among groups did not differ significantly. The dimension n-sn was significantly differed between groups, being smaller in the crossbite-group.

The results of unilateral BF and the results of the comparison between the sides of the dental arches (left/right or normal/crossbite-side) are shown in table 3. BF magnitude did not show a significant difference among groups; however, the comparison between the normal and crossbite-side in the crossbite-group showed a significant difference, the magnitude in crossbite-side being smaller ( $p < 0.0001$ ). Table 3 also shows the description of the MP parameters, and X<sub>50</sub> and “b” did not differ significantly among groups ( $p > 0.05$ ).

**Table 2** - Demographic data and descriptive data for facial dimensions (mm) of the sample divided in three groups

Group	n (♂/♀)†	Age‡ (months) Mean (SD)	BMI‡ (Kg/m <sup>2</sup> ) Mean (SD)	n-sn Median (IQR)	sn-gn Median (IQR)	go-go Median (IQR)	n-gn/zy-zy Median (IQR)
Normal	22 (14/8)	56.36 (8.59)	16.07 (2.05)	4.63 (0.68)	4.18 <sup>A</sup> (0.44)	9.85 (0.72)	0.78 (0.11)
Crossbite	20 (8/12)	56.00 (6.46)	16.42 (1.51)	4.70 (0.66)	3.75 <sup>B</sup> (0.51)	9.85 (0.75)	0.77 (0.04)
Open bite	23 (12/11)	56.74 (10.72)	16.83 (2.49)	4.70 (0.45)	4.20 <sup>AC</sup> (0.43)	9.80 (0.60)	0.80 (0.07)

BMI, body mass index; SD, standard deviation; IQR, interquartile range; n-sn, nasion-subnasale; sn-gn, subnasale-gnathion; go-go, gonion-gonion; n-gn, nasion-gnathion; zy-zy, zygonion-zygonion.

†  $p > 0.05$  (Chi-square test)

‡  $p > 0.05$  (Kruskal-Wallis test)

A≠B≠C ( $p < 0.05$ ; ANOVA/Tukey post-test or Kruskal-Wallis test/Dunn post-test).

**Table 3** - Descriptive values of unilateral bite force (BF) and masticatory performance

Group	BF right side/crossbite (N)	BF left side/normal (N)	BF: difference between sides dental arches (N)	X <sub>50</sub>	b
	Mean (SD)	Mean (SD)	Mean (SD)	Median (IQR)	Median (IQR)
Normal	199.93 (42.90)	185.95 (39.44)	29.36 (26.30)	4.54 (1.37)	2.52 (1.02)
Crossbite	153.66* (37.39)	179.32* (35.57)	30.24 (19.36)	5.12 (1.06)	2.56 (0.47)
Open bite	180.46 (54.48)	198.49 (60.78)	30.53 (22.70)	4.75 (1.48)	2.23 (0.60)

SD, standard deviation; IQR, interquartile range; BF, bite force; X<sub>50</sub>, median particle size of the test-material; b, distribution of particles in different sieves.

\* p < 0.001 (difference = -27.685; CI = -38.874 to -16.496; power = 0.99; paired t-test).

**Table 4** - Correlation coefficients obtained between facial dimensions, bite force and masticatory performance (only significant coefficients are shown)

	r (p-value)	zy-zy	go-go	n-sn
Open bite group	BF†	-	0.43 (0.038)	-
Crossbite group	X <sub>50</sub> †	-0.57 (0.012)	-0.56 (0.014)	-
Open bite group	X <sub>50</sub> ‡	-	-0.50 (0.021)	-0.50 (0.025)

r, correlation coefficient; BF, bite force; X<sub>50</sub>, median particle size of test-material; n-sn, nasion-subnasale; go-go, gonion-gonion; zy-zy, zygion-zygion.

†Pearson correlation test

‡Spearman correlation test

Table 4 shows the significant correlation coefficients obtained between BF, MP and facial dimensions in the three groups. In the open bite group, BF was positively correlated with intergonial distance, that is, a greater mandibular width. Median particle-size (X<sub>50</sub>) correlated negatively with go-go and zy-zy dimensions in the crossbite-groups, and go-go and n-sn in the

open bite group. These findings suggest that smaller median particle-size (X<sub>50</sub>) correlates with larger dimensions of the faces in the open bite and crossbite groups. The coefficients found for normal occlusion group did not reach the level of significance.

The results found in the evaluation of salivary parameters are shown in Table 5. US flow rates significantly differed between groups with malocclusion, being lower in the crossbite group. There was no significant difference in the concentrations of AMY and TP among the three groups. The comparison between SS and US showed a significant difference in flow rate and AMY concentration in all groups (p < 0.05). TP concentration differed between SS and US only in the crossbite-group, and it was lower in US.

The multiple linear regression model showed a significant positive relationship between SS flow and age and a negative relation

**Table 5** - Descriptive data for salivary parameters

Group	Unstimulated flow rate (g/min)	Stimulated flow rate (g/min)	Unstimulated Amylase (U/L)	Stimulated Amylase (U/L)	Unstimulated Total Protein (mg/dL)	Stimulated Total Protein (mg/dL)
	Median (25-75%)	Median (25-75%)	Median (25-75%)	Median (25-75%)	Median (25-75%)	Median (25-75%)
Normal	0.2* (0.2-0.4)	0.4* (0.3-0.8)	2299.0* (1351.9-3375.6)	3985.8* (2604.5-5032.4)	36.1 (29.8-51.4)	38.6 (30.8-47.1)
Posterior Crossbite	0.2* <sup>A</sup> (0.1-0.2)	0.3* (0.1-0.4)	2108.5* (1633.5-3031.0)	2919.0* (2429.0-3801.5)	47.1* (34.6-69.9)	41.1* (26.9-50.8)
Anterior open bite	0.3* <sup>B</sup> (0.2-0.4)	0.6* (0.3-0.6)	1442.0* (872.0-2616.5)	2509.0* (1312.0-4535.0)	43.5 (31.1-53.0)	44.2 (32.3-53.8)

A≠B in the same column: comparison between groups (p < 0.05, Kruskal-Wallis test and Dunn post-test)

\* p < 0.05 (paired t-test).

with the presence of malocclusion (Table 6). The model used accounts for about 25% of the variation of SS flow in the studied sample.

## DISCUSSION

The comparison of facial morphology among groups showed a smaller dimension of the lower face (sn-gn) in the crossbite-group, probably because functional posterior crossbite is generally associated with a distal step in the crossbite-side (which may persist as a Class II subdivision) and rotational closure of the mandible [16]. The mean BF did not show differences between groups; however, when the magnitude was compared between the sides of the dental arches (left/right or normal/crossbite-side), the crossbite-group showed smaller BF magnitude in the crossbite-side. These findings corroborate past studies that showed several changes in the growth and function of orofacial structures related to the persistency of this malocclusion, such as decrease in BF magnitude [17], asymmetrical masticatory muscle activity, muscle and hard tissues asymmetries [4,17] and abnormal chewing pattern [18]. Since these conditions may lead to changes in masticatory function, early diagnosis and treatment plan are advised to normalize the occlusion and create conditions for normal growth and development.

The MP parameters evaluated, i.e. X50 and b, also did not differ between groups, although some past studies showed a negative effect of malocclusion on subjects' ability to process and break down food or a test-material [19,20]. The literature shows that the relationship between

food comminution and malocclusion remains controversial, especially in young subjects or subjects with less severe types of malocclusion [8,21]. In these cases, it can be inferred that adaptive mechanisms may compensate morphological and/or functional asymmetries, which occurs early in development [16,22]. Unilateral chewing is a way to compensate for a functional deficiency and, probably, that is the reason why BF and MP did not differ among groups. Moreover, at this stage of dentition, the child is still under learning process of mastication, and MP depends on many other factors involved in this process, such as the number of teeth in contact, age, BMI [20,21] and facial morphology, as will be discussed below.

The moderate coefficients obtained between BF, MP and facial dimensions showed that a smaller median particle-size (X50) correlated with larger dimensions of the face, both in the open bite and crossbite-groups, as well BF magnitude related positively with the lower jaw width. This relationship was also observed in the normal-occlusion group, although the coefficients were not significant. These findings agree with previous studies which showed that MP improves with chronological age, body size, BF, and increased occlusal contact area [21], that is, good conditions to generate force to break down food in smaller particles. Even in young children, these relationships showed their importance in the development of mastication.

AMY activity and TP concentrations did not differ among groups and an increase

**Table 6** - Multiple linear regression model with stepwise backward elimination used to verify the relationship between stimulated salivary flow rate and studied variables (only significant results are shown)

Dependent variable	Independents variables	Coeff.	p value	Model		
				R <sup>2</sup>	p-value	Power 5%
Stimulated salivary flow rate	Constant	-0.090	-	0.247	0.001	0.962
	Age	0.011	0.005			
	Malocclusion	-0.172	0.018			

Normality test: p=0.070  
Constant of variation: p=0.322.

in flow rate and AMY activity from US to SS was observed in all groups. These findings are similar to that found by Arhakis et al. [23], who showed an increase in salivary flow rate followed by the increase in AMY activity under no stressful conditions in adults. However, according to past studies, AMY activity is independent from saliva flow [24]; thus, AMY activity and flow rate increased from US to SS saliva, although they probably did not increase together, as observed in the results found by the linear regression model.

By analyzing the results found, the crossbite-group showed lower US flow rate in relation to the other groups; also, according to the linear regression model used, the SS flow rate related negatively with the presence of malocclusion. Salivary secretion is predominantly under the control of the autonomic nervous system, although other types of stimuli induce salivary secretion, such as smell and taste of food and chewing [25]. During mastication, stimulation of mechanoreceptors in soft tissues may result in saliva secretion by eliciting the masticatory-salivary reflex. Since subjects with malocclusion may present impairments in jaw mobility during chewing cycles [22], alterations in dental occlusion may have an influence on the amount of salivary secretion. A past study showed that during unilateral chewing in children, saliva is not well distributed around the mouth and tends to stay on the chewing side of the mouth [26], where saliva is predominantly produced.

In the crossbite-group, lower TP concentration in SS was observed, which means that increasing flow rate by increasing the degree of stimulation, a protein dilution effect may occur [2]. de Campos et al. [13] also showed a decrease in TP concentration under chewing stimulation, when the parotid glands participate to a greater extent.

Age may also influence salivary flow [27], and the literature reported a significant correlation between salivary flow rate and age

in children [28] and adolescents. In this way, salivary flow rate increases with age [27] and salivary glands seem to be fully developed at the age of 15 [28]; in addition, a past study showed that a high or low flow rate pattern is a constant individual trait in children [29]. The results found may also be influenced by the child's cooperation with sampling techniques, and children may acquire learning on how to spit over the years, although every child included in this study has been instructed previously.

Food is submitted to several actions in the mouth: lubrication with saliva, reduction to small particles and starch hydrolysis, showing the importance of oral digestion to global digestion [30]. To date, little is known about masticatory and salivary parameters in young subjects and to what extent these characteristics may influence the subject's nutritional status; thus, further studies are needed to confirm these hypotheses.

In conclusion, the present study showed that the early assessment of masticatory and salivary functions is of importance when considering different morphological occlusions in the primary dentition. In children with crossbite, the magnitude of BF in the crossbite-side was smaller than the normal side. In children with open bite or crossbite, larger dimensions of the faces correlated with smaller median particle-size (X50), that is, better masticatory performance. In addition, unstimulated salivary flow differed between groups, being lower in the crossbite group. Taken together, these findings draw attention to the early assessment of changes in functions related to the masticatory system.

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**Prof. Paula Midori Castelo  
(Corresponding address)**

Universidade Federal de São Paulo (UNIFESP) – Campus Diadema  
R. São Nicolau, 210 – Diadema – SP / Brasil / 09972-270  
E-mail: pcastelo@yahoo.com

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