



## Comparing Properties (Concentration, PH and mutans streptococcus Saliva in Both Status Resting Saliva and Stimulated Saliva in Preschoolers of Kerman city

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### ABSTRACT

This paper aimed to compare the characteristics (concentration, PH and mutans streptococcus) saliva in both status resting saliva and stimulated saliva in preschoolers of Kerman city. In this study, 100 children aged 5 years among patients admitted to the pediatric ward of Kerman dental school and dental offices, some experts in Kerman dental school participated. Resting and stimulated saliva (after chewing oral paraffin) children collected and in concentrations, PH and the amount of mutans streptococcus was measured. Mc Nemar test to compare the frequency of positive and negative cultures before and after stimulation as well as paired t-test to compare the saliva pH and concentration of not stimulated saliva and stimulated saliva in two modes was used. The significance level was set less than 0.05. The mean resting salivary osmolality of the population:  $30.42 \pm 87.41$  and the average salivary osmolality of the total population were 79.81. Osmolality differences in saliva before and after stimulation with each other was significant ( $p = 0.009$ , paired t-test). The mean of resting saliva in the total population PH  $0.45 \pm 7.78$  and the average PH stimulated saliva in the total population was 8.22 and the difference before and after each significant ( $p = 0.02$ , paired t-test). In mutans streptococcus in test samples in all 71 patients (71%) positive test and 29 patients (29%) had a negative test that number of positive cultures are equal before and after stimulation of saliva and thus the difference between the two groups ( $p > 0.05$ ) was observed. In terms of comparing the properties of resting and stimulated saliva can conclude that salivary stimulated PH was significantly higher than resting saliva. While stimulated saliva osmolality was significantly less than resting saliva and the frequency of positive test mutans streptococcus in saliva before and after stimulation had no significant difference ( $p > 0.05$ ). This means that test results on samples of mutans streptococcus in stimulated saliva and resting saliva is same.

**Keywords:** resting saliva, stimulated saliva, PH, concentration of mutans streptococcus

### INTRODUCTION

Dental caries is a multifactorial disease, the role of each factor in its development can partially be independent of other factors that caused or exacerbated. Among the most important factors in the pathogenesis of dental caries can be fed such as the type and amount of carbohydrate intake, oral hygiene and controlling dental plaque and saliva compounds noted. Among the most important factors affecting susceptibility to the development of dental caries, is the quality and quantity of saliva. The rate of decay is significantly higher than that in terms of quality and quantity of natural saliva [1]. It is generally accepted that the process of tooth decay naturally present in saliva is controlled largely with safety mechanism. Many of the properties of saliva to understand their possible role in the process of

decay were evaluated [2]. PH of saliva, the ability to neutralize acid, calcium, phosphorus and fluoride is very important. Long as the addition of these features, affect the flow rate and concentration of saliva in caries. Therefore, it seems all the known characteristics of saliva in a way connected with the process of tooth decay [3]. Saliva concentration of dissolved substances in saliva can be expressed as a percentage and as mentioned factors can affect the caries process but few studies have examined its significance and the factors, influencing it can be noted nutrition person [4]. PH saliva based on acidic or basic saliva properties can be defined the normal value is between 5.3 to 7.8. This makes it identified as a factor in the process of dental caries [5]. Factors affecting it allow medications, age, stress and physical activity named [6]. The most important and most pathogenic microorganism *Streptococcus mutans* in saliva is described rot and is associated with the onset and progression of dental caries [7]. On the other hand, not stimulated saliva in two non-tied and stimulus (food, chewing, etc) and stimulated secretion in response to gustatory stimuli. They may be because the combination can be different and compare them in terms of risk of caries is important [8].

Due to the lack of studies comparing stimulated saliva and not stimulated in this age as well as the low number of subjects and emphasize only one or two properties in their saliva, in this study tries to saliva properties (Concentration, PH and buffering power) in both stimulated and not stimulated saliva examined and its relationship with age, gender and decay in the baby's mouth with DMFT index determined by our assessment [9].

The results of this study determine the qualitative characteristics of saliva in future and to control dental caries in this age range through considering the characteristics of saliva will help.

## MATERIALS AND METHODS

This cross-sectional study and the target population in this study was 100 children aged 5 years among patients admitted to the pediatric ward of Kerman and Kerman dental school and clinic in Kerman children had some dental professionals (The selection only children 5-year age effect on the properties of saliva) [10]. The objectives and procedures described in this study for parents and informed consent form was obtained from them [11] (Sample form in the appendix) and then people who have been systemic disease or syndrome, have a cold, flu or any other viral disease or were active bacterial, those (parents) have refused to sign a written consent, were excluded [12].

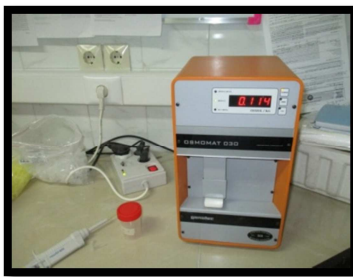
The more samples selected for cooperation and prevent their failure to cooperate: (1) Oral examination, (2) Fluoride therapy (3) confirm the test results to parents, and (4) a number toothbrush and toothpaste for children (age appropriate) samples for free, all participants were awarded.

On arrival at the clinic and then provide the necessary explanations and informed consent of the parents of children under the intraoral examination and demographic data as well as medical history and dental them through interviews with parents then his name and address and contact number to send laboratory results were recorded on a separate form.

Dmft indicator (diseased, missed and filled teeth) dental mirror and probe beneath the unit for each individual study and number as it was recorded [13]. Then a number of children placed at the disposal toothbrush and toothpaste and asked them to clean their teeth under the supervision of clinic staff and they were asked to avoid eating or drinking for 50 minutes of each species (excluding the effect of nutrition on levels of saliva). Children studied at this time at the clinic and remained under the supervision of personnel to ensure they do not eat or drink. In the next step of the selected children were asked to take down the head, not swallow your mouth water for 2 minutes. In addition, entirely within the empty container dial that placed at their disposal [14]. A time to rest another 10 minutes and repeats the previous step. The difference is that this time, before collecting saliva, saliva by chewing paraffin for the company GC, Japan was aroused. The patient was discharged. Other steps experimentally were done.

### The method of measuring parameters

**Concentration of saliva:** in each saliva samples were prepared (either at rest or in a state of excitation) using osmometers (Gonotec Company, Germany) were digitally, after the removal of 500 micrometers of instances of each series with the sampler and for it and then transfer it to the micro-pit and put in the device, using cryotherapy osmolality. Each of the samples in hot Smolwere then displayed on the display device and then recorded [15].



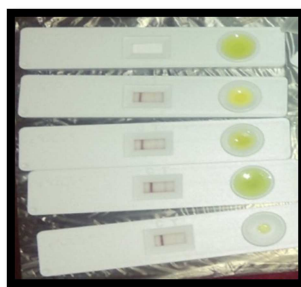
**Figure 1: Sampler and micropipette with osmometers devices**

Check PH: PH any saliva samples taken (either at rest or in a state of excitation) using PH meter (Consort Company, Elgium) digitally and using sensitive sensor device was examined and ph results were recorded [9].



**Figure 2: PH meters and measuring devices PH samples**

The amount of Streptococcus mutans in saliva: the Streptococcus mutans samples collected using the kit for measuring the amount of streptococcus mutans (GC Company, Japan) was investigated and the results positive (high) and negative (low) recorded.



**Figure 3: Test kit for streptococcus mutans**

### **Statistical analysis**

Descriptive statistics were used to describe the overall population, the distribution of data Kolmogrov-Smirnov test was assessed, and if the data had normal distribution, parametric tests were used. To compare the osmolarity of saliva and salivary pH in both genders and different dmft (caries free and caries active) independent of the t-test was used. To compare the number of positive and negative cultures before and after stimulation Mc Nemar test was used when considering the similarity of culture before and after stimulation, this difference was not statistically significant. To compare the pH of saliva and saliva concentrations in saliva stimulated and not stimulated in two paired t-test was used [5].

### **Ethical Considerations**

This study was observational and specific interventions in order to advance the goals of the research on children were done. Necessary tests on the saliva and send the results to a screening of families considered and can be useful. However all stages of the study were explained to the parents and informed consent was obtained. Only children whose parents have released their written consent were studied..

**RESULTS****Average dmft index**

Average Dmft index for the total population was  $2.92 \pm 6.46$ . Dmft index for boys and for girls  $2.95 \pm 6.44$   $2.91 \pm 6.47$ .

**Table 1: The average dmft index for girls and boys (n)**

Gender		Minimum	Maximum	Average	Standard deviation
(100) Boy	Dmft	.00	13.00	6.4706	2.91447
(100) Girl	Dmft	.00	13.00	6.4490	2.95847

**Average PH and osmolality of saliva in the resting saliva in the total population according to gender:**

Average PH resting saliva of the population:  $0.45 \pm 7.78$

The average osmolality of resting saliva of the population:  $30.42 \pm 87.41$

Boys: Average PH  $0.43 \pm 8.08$  respectively. The mean osmolality was  $30.98 \pm 86.13$ .

Girls: Average PH  $0.47 \pm 8.08$  and the average osmolality was  $30.09 \pm 88.73$ .

**Table 2: Average PH and osmolality in resting saliva in the total population according to gender (number)**

	Standard deviation	Average	Maximum	Minimum
PH rest Total sample (100)	6.94	9.40	7.7867	.45193
Osmolality rest Total sample (100)	41.00	197.00	87.4100	30.42165
PH rest Osmolality rest Boy (51)	6.94 41.00	9.40 159.00	8.845 86.1373	.43732 30.98130
PH rest Osmolality rest Girl (49)	7.06 43.00	9.09 197.00	8.0890 88.7347	.47119 30.09068

The number of samples tested positive of mutans streptococcus in saliva rest in all sample according to gender:

A positive test for the bacteria more than  $5 * 10^5 \frac{\text{CFU}}{\text{ml saliva}}$  and test negative for the bacteria is less than or equal to this amount. In all samples, 71 (71%) tested positive and 29 patients (29%) had a negative test. 38 samples were positive in boys and in girls 33 samples were positive. This means that 74.5% of boys and 67.3 percent girls had a positive test, the difference was statistically significant. The amount of mutans streptococcus significantly is higher in boys.

**Table 3: Number of samples tested positive for mutans streptococcus in resting saliva in all cases according to gender (number)**

		Number	Percent
Total (100)	Negative	29	29.0
	Positive	71	71.0
Boy (51)	Negative	13	25.5
	Positive	38	74.5
Girl (49)	Negative	16	32.7
	Positive	33	67.3

Average PH and salivary osmolality of all samples and boys and girls separately:

Average PH in the saliva stimulated in the total population was 8.22 and the average stimulated salivary osmolality of the total population was 79.81

Average PH in the stimulated saliva interest in boys was 8.28 and the average stimulated salivary osmolality of boys was 78.72

Average PH in stimulated saliva was 8.15 in girls and the average stimulated salivary osmolality of girls was 80.93

**Table 4: Average PH and osmolality stimulated saliva and distinguish gender in the general population (number)**

	Minimum	Maximum	Average	Standard deviation
Total (100) PH Osmolality	6.65	9.26	8.22	0.48
	31.00	156.00	79.8100	25.10129
Boy (51) PH Osmolality	6.65	9.26	8.2873	.47539
	31.00	155.00	78.7255	25.70454
Girl (49) PH Osmolality	6.76	9.16	8.15	.49
	46.00	156.00	80.9388	24.67236

The number of positive tests Streptococcus mutans in stimulated saliva in general and differentiated by gender:  
 The stimulated saliva in all the samples tested, 71 were positive and 29 were negative test  
 The stimulated saliva in a total of 38 positive and 13 negative test was boys  
 The stimulated saliva the girls tested, 33 were positive and 16 were negative test

**Table 5: Number tested positive for mutans streptococcus in stimulated saliva in general, according to gender (number)**

	Cultivation	Number	Percent
Total (100)	Negative	29	29.0
	Positive	71	71.0
Boy (51)	Negative	13	25.5
	Positive	38	74.5
Girl (49)	Negative	16	32.7
	Positive	33	67.3

Compare dmft, PH and osmolality between boys and girls before and after stimulated saliva (t-test):  
 In none of the cases, there was no significant difference between boys and girls ( $p > 0.05$ , t-test).

**Table 6: Compare dmft, PH and osmolality between boys and girls before and after stimulation of saliva**

		t-test for data distribution		Degrees of freedom	p-value	mean deviation	Degrees of freedom	95% confidence interval of measure	
		T value						Fewer	More
dmft	Equal variance assumption	.037		98	.971	.02161	.58734	-1.14394	1.18716
	Unequal variances assumption	.037		97.700	.971	.02161	.58752	-1.14434	1.18756
PHRest	Equal variance assumption	-.049		98	.961	-.00447	.09086	-.18478	.17584
	Unequal variances assumption	-.049		96.726	.961	-.00447	.09100	-.18509	.17615
Osmolarity rest	Equal variance assumption	-.425		98	.672	-2.59744	6.11089	-14.72429	9.52942
	Unequal variances assumption	-.425		97.988	.672	-2.59744	6.10729	-14.71717	9.52229
PH stimulatory	Equal variance assumption	1.355		98	.178	.13093	.09660	-.06077	.32263
	Unequal variances assumption	1.354		97.496	.179	.13093	.09666	-.06091	.32276
Osmolarity stimulatory	Equal variance assumption	-.439		98	.662	-2.21329	5.04186	-12.21869	7.79212

*Compare the pH of saliva before and after stimulation:*

The difference before and after each significant ( $p = 0.02$ , paired t-test) as a result of stimulated saliva PH was significantly higher than the rest of saliva.

**Table 7 comparing the pH of saliva before and after stimulation**

	T value	Degrees of freedom	significance level p-value
resting PH against stimulated PH	-2.370	99	.020

Comparing osmolality of saliva before and after stimulation of saliva:

Osmolality differences in saliva before and after stimulation with each other was significant ( $p = 0.009$ , paired t-test) result was significantly less salivary osmolality of resting saliva.

**Table 8: Comparison of salivary osmolality of the samples before and after stimulation**

	T value	Degrees of freedom	significance level p-value
Resting osmolality in front of stimulated osmolality	2.682	99	.009

Frequency tested positive for mutans streptococcus in saliva before and after stimulation:

The number of positive cultures is equal before and after chewing paraffin. Hence, the analysis showed no significant difference between the two groups Mc Nemar ( $p > 0.05$ ).

Comparing PH and osmolarity of saliva dmft less than or equal to two groups of five and more than five (caries free and caries active):

PH and osmolarity of saliva before and after chewing paraffin in the two groups were not significantly different.

**Table 9: Comparison of salivary PH and osmolarity less than or equal dmft into two groups of five and more than five (caries free and caries active) before stimulate saliva**

		t-test for data distribution						
		T value	Degrees of freedom	p-value	mean deviation	Degrees of freedom	95% confidence interval of measure	
							Fewer	More
PHRest	Equal variance assumption	-1.522	98	.131	-.13950	.09164	-.32136	.04236
	Unequal variances assumption	-1.569	91.712	.120	-.13950	.08892	-.31611	.03711
Osmolarity rest	Equal variance assumption	.385	98	.701	2.40000	6.23668	-9.97650	14.77650
	Unequal variances assumption	.378	78.560	.706	2.40000	6.34843	-10.23733	15.03733
PH stimulatory	Equal variance assumption	-.299	98	.765	-.02975	.09945	-.22710	.16760
	Unequal variances assumption	-.304	87.903	.762	-.02975	.09797	-.22444	.16494
Osmolarity stimulatory	Equal variance assumption	.629	98	.531	3.23333	5.13949	-6.96581	13.43248
	Unequal variances assumption	.621	79.962	.536	3.23333	5.20664	-7.12828	13.59495

*Comparison of saliva testing positive for mutans streptococcus dmft less than or equal to two groups of five and more than five (caries free and caries active):*

Of the 100 samples studied, 60 of them in the group Caries active and 40 of them were in the group Caries free. Of the 60 patients in Caries active group, 52 of them (86.66%) tested positive for Streptococcus mutans. Of the 40 patients in Caries free group, 19 cases (47.5%) tested positive for Streptococcus mutans.

## DISCUSSION

The PH level of saliva before and after the conclusion was that stimulate salivary PH in the saliva of subjects above the rest and this difference PH saliva before and after stimulation with significant ( $p = 0.02$ , paired t-test). This means that PH saliva stimulated salivary flow was significantly higher than the rest in this age group. Very few studies on the comparison of PH in resting and stimulation saliva were done [13]. In a study by Gittings et al concluded that salivary PH was significantly higher than resting saliva [9]. So in terms of higher PH stimulated saliva and resting saliva the results of this study are similar studies.

PH more is stimulated saliva due to differences in electrolytes compared to the resting saliva. So that we stimulate salivary secretion of saliva and more than parotid gland and parotid gland is predominantly serous secretion, which is rich in electrolytes and enzymes (unlike other glands that secrete mucus-rich glycoprotein). The electrolyte content in the stimulated saliva compared to the resting saliva is PH [7-14]. In terms of the level of concentration in resting saliva and stimulated saliva must first point material mole is the osmolality values that are dissolved in liquids or in other words solute expressed in ml. The resting saliva and stimulated saliva by measuring the average osmolarity conclusion was that the resting salivary osmolality is higher in subjects and this difference osmolarity of saliva before and after stimulation with significant ( $p = 0.009$ , paired t-test). This means that it is significantly less resting salivary osmolality saliva in this age group. Very few studies comparing the osmolality or concentration of resting and stimulated saliva are done. In a study Gittings et al did, it concluded is significantly less salivary concentrations than resting saliva [9]. So in terms of osmolality less than stimulated saliva and resting saliva the results of this study are similar studies. Less salivary concentrations in resting saliva compared to two reasons:

1. Salivary flow leads to a relative increase in more liquid soluble in stimulated saliva.
2. The content of mucin (glycoproteins and oligosaccharides) due to lower salivary secretion of mucin content is much lower parotid glands more tubers sublingual, submandibular and palatine (main officials salivating rest) [9].

In terms of comparing the frequency of positive test mutans streptococcus in saliva before and after stimulation because equal number of positive cultures before and after saliva stimulation with each other, Mc Nemar analysis showed no significant difference between the two groups ( $p > 0.05$ ). This means that test results on samples of mutans streptococcus in saliva and saliva stimulation rest is the same. Since, paraffin candies to stimulate saliva samples for (GC Company) were used and it did not make any transform in the mutans streptococcus in saliva. There is no similar study on comparison of mutans streptococcus in stimulated and resting saliva found the results of this study would be satisfied.

In terms of comparison between the genders, the mean dmft in boys sample was slightly higher than girls, PH saliva sample was almost equally in boys and girls resting, resting salivary osmolality slightly higher in girls than boys sample, PH in stimulated saliva sample was slightly higher in boys, Osmolarity stimulates saliva sample was slightly higher in girls. However, in none of the cases there was no significant difference between boys and girls ( $p > 0.05$ , t-test). This means that girls and boys in this age group in terms of dmft index and examined the saliva properties (PH and osmolality) or at resting saliva or while on stimulated saliva was no significant difference. In a study conducted by Pandey et al was conducted on 120 children 7 to 15 years old the result was a significant correlation between saliva and dmft PH values are not the same age and gender [15]. Therefore, in terms of the lack of difference in dmft, PH saliva between the genders at resting and stimulated saliva results of this study are similar studies.

Compare also on salivary osmolality between girls and boys was similar study has been done. As a result, the results of this study can be satisfied in this case. While in the prevalence of mutans streptococcus in saliva tests positive for resting and stimulated by each gender;

Referring to the order of the positive test bacteria levels more than  $5 * 10^5 \frac{\text{CFU}}{\text{ml saliva}}$  and test negative for the bacteria is less than or equal to this amount, the result was that 74.5% of boys and 67.3 percent girls had positive test this difference is statistically significant and the bacteria mutans streptococcus significantly higher in boys sample.

Comparing the two genders in the field of mutans streptococcus in the same study found and in this context the results of this study will be satisfied. Perhaps due to the increased amount of mutans streptococcus in the oral health of their sons observe less because they pay less attention to this case.

Comparing PH and osmolality of saliva dmft less than or equal to two groups of five and more than five (caries free and caries active) first. It should be noted that according to similar studies [9] Caries free samples and Caries active groups were divided according to their dmft, which means that people with dmft five and less in the group Caries free and people with higher dmft were five in the group Caries active. Then to understand the role of PH and osmolality of saliva on dental caries in this study as a secondary objective, two groups were compared and the following results were obtained.

#### **To stimulate saliva**

The rest of PH Caries active in the group is slightly higher than in Caries free and the rest in the group Caries free osmolality of saliva is slightly higher than in Caries active. However, the amount of PH and osmolality of saliva rest in the two groups had no significant difference. This means that the PH and osmolality of saliva rest is not much different in the two groups Caries active and Caries free and so PH and osmolality of saliva break with the incidence of dental caries in this age group is not connected.

#### **After stimulate saliva**

PH is approximately equal to the stimulated salivary flow rates in the two groups. However, the salivary osmolality of Caries free group is slightly higher than Caries active. Nevertheless, the amount of salivary PH and osmolality of the two groups had no significant difference. This means that the salivary PH and osmolality is not much different in the two groups Caries active and Caries free and therefore salivary PH and osmolality is not associated with the incidence of dental caries in this age group. In a study conducted by Preethi et al on saliva break the conclusion was that there was poor communication between PH saliva and caries activity. Tuhunoglu et al also studied and noted that this was a result of no connection between PH saliva and caries activity regardless of gender and age of the person is not [16].

In a study conducted by Pandey et al over the rest of saliva that over 120 children 7 to 15 year old it was concluded that the correlation between PH levels in saliva and caries activity regardless of age and gender does not exist [15]. In a study conducted on 100 children under six years by Jayaraj et al and by stimulated saliva and resting saliva PH It was concluded that a significant difference between the two groups rot and decay without it [17]. In a study conducted by Dogra et al on 80 patients 7 to 14 years and this result was achieved on the PH saliva and saliva rest Caries active in the group is reduced but this reduction is not statistically significant [18]. So in terms of the difference PH and osmolarity resting saliva and irritation in the Caries active Group and Caries free the results of this study are similar studies. In terms of the level of saliva testing positive for mutans streptococcus in caries free and caries active groups due to the fact the Caries active Group 86.66% of tests were positive for mutans streptococcus and in Caries free 47.5% tested positive for mutans streptococcus. It can be concluded that Caries active in people testing positive for mutans streptococcus and therefore the amount of mutans streptococcus is higher than Caries free. In a study that was conducted by Ghasempour et al concluded the amount of mutans streptococcus significantly higher in the group Caries active is Caries free [19]. So in terms of the rate of Streptococcus mutans in Caries active group compared to the group Caries free and the results of this study are similar studies.

## CONCLUSION

*According to obtained the following results:*

In terms of comparing the properties of resting and stimulated saliva can conclude that salivary PH was significantly higher than resting saliva. While salivary osmolality was significantly less than resting saliva and the frequency of positive test mutans streptococcus in saliva before and after stimulation had no significant difference ( $p > 0.05$ ). This means that test results on samples of mutans streptococcus in resting saliva and stimulated saliva is same. In terms of comparison between the gender PH and osmolarity in stimulated and resting saliva between boys and girls before and after stimulated saliva had no significant difference ( $p > 0.05$ , t-test). However, the bacteria mutans streptococcus significantly higher in boys than girls sample. In terms of comparison between the two groups Caries active and Caries free, PH and osmolarity of saliva in caries free and caries active both before and after saliva stimulation had no significant difference and therefore salivary PH and osmolarity is not associated with the incidence of dental caries in this age group. While Caries active in people testing positive for mutans streptococcus and therefore the amount of mutans streptococcus is higher than Caries free.

### Practical suggestions and aspect needs more study

Although the specification (PH and osmolarity) resting saliva and saliva stimulated in the age group of subjects differ (as others based on other articles have investigated) but this difference is associated with the incidence of caries in these patients is poor. Therefore, it is suggested that in other research to other etiologic factors other than the characteristics of dental caries (PH and concentration) saliva was noted.

## REFERENCES

- [1] KhwajaHassani, R., Pvraslamy, H., check glucose levels in saliva of children with early childhood caries compared to normal healthy children, PhD thesis General Dentistry, Kerman, Kerman University of Medical Sciences, 2016, 40 p.Fejerskov, E; Kidd :Dental Caries: The Disease and Its Clinical Management, 2nd ed. Wiley-Blackwell 2008;chap 9:781-4051-3889-5.
- [2] Mcdonald. A.D., Avery, D.A. Dean. J.A. Dentistry for the child and Adolescent. St.louis: Mosby,2004; 240-250.
- [3] C. Fenoll-Palomares, J. V. Muñoz-Montagud, V. Sanchiz, B. Herreros, V. Hernández et all,Unstimulated salivary flow rate, pH and buffer capacity of saliva in healthy volunteers, REVESPENFERMDIG (Madrid) , Vol. 96. N.º 11, pp. 773-783, 2004.
- [4] ZabokovaBilbilova E, SotirovskaIvkovska A, Georgiev Z, Stefanovska E. Evaluation of buffer capacity of saliva in caries-free and caries-active children. Prilozi. 2013;34(2).
- [5] P.L. Fogilio-Bonda, M. Migliario, V. Rocchetti, F. Pattarino,A. Foglio-Bonda. Daily and annuallyvariation of unstimulated whole saliva flow rate and pH and their relation with body profile in healthy young adults , EuropeanReviewforMedicalandPharmacological Sciences, 2013; 17: 2545-2538.
- [6] PariziTorabi, Maluku, oral health, nutrition and other factors, Tehran, Press Organization (SID), 1383; 28-60.
- [7] M Moritsuka, Y Kitasako, MF Burrow, M Ikeda, J Tagami, The pH change after HCl titration into resting and stimulated saliva for a buffering capacity test, Australian Dental Journal, 2006; 51(2):170-174.
- [8] Sally Gittings, Neil Turnbull, Brian Henry, Clive J. Roberts, Pavel Gershkovich, Characterisationofhuman saliva as a platform for oral dissolution medium development, European journal of pharmaceutics and biopharmaceutics,2015;16-24.



- [9] VildanErdem, Mehmet Yıldız, TeomanErdem, The Evaluation of Saliva Flow Rate, pH, BufferCapacity, Microbiological Content and Indice of Decayed, Missing and Filled Teeth in Behçet'sPatients , *Balkan Med J* 2013; 30: 211-14.
- [10] Anupama Gaur, N Anup, Rajesh Sharma , Variation in Salivary Parameters and its Correlation withPlaque and Gingival Status among 12 to 15 Years Schoolchildren of Rural and Urban Jaipur City in Winter and Summer Seasons, 10.5005/jp-journals-10005-113204.
- [11] H Islas-Granillo, SA Borges-Yañez, CE Medina-Solís, CA Galan-Vidal, JJ Navarrete-Hernández , MESCOFFIÉ-Ramirez ,Salivary Parameters (Salivary Flow, pH and Buffering Capacity) in Stimulated Saliva of Mexican Elders 60 Years Old and Older , *West Indian Med J.* 2014; 63(7): 758.
- [12] A.R. Moreira, I.A. Passos, F.C. Sampaio, M.S.M. Soares and R.J. Oliveira, Flow rate, pH andcalcium concentration of saliva of children and adolescents with type 1 diabetes mellitus , *Salivary flow rate, pH and calcium in T1DM* , *Brazilian Journal of Medical and Biological Research*, 2009; 42: 707-711.
- [13] Shahrabi M et al. A comparison of salivary calcium, phosphate, and alkaline phosphatase inchildren with severe, moderate caries, and caries free in Tehran's kindergartens. *J Indian SocPedod Prevent Dent*, 2008; 74-77.
- [14] Vibhakar PA, Patankar SR, Yadav M, Vibhakar P. Correlation of Salivary Glucose Levels with DentalCaries: A Biochemical Study. *IntJOral Maxillofacial Patho*, 2014;1(1):17-20.
- [15] Çağlar E, Cildir SK, Ergeneli S, Sandalli N, Twetman S(2006).Salivary mutans streptococci andlactobacilli levels after ingestion of the probiotic bacterium *Lactobacillus reuteri* ATCC 55730 by straws or tablets. *ActaOdontolScand* 64,314–318.
- [16] P Pandey, N Venugopol, C.P. Ehaudhary, Estimation of salivary flow rate, PH, buffer capacity, calcium, total protein and total antioxidant capacity in relation to dental caries severity, age and gender, contemporary clinical dentistry mednow publication, 214-221.
- [17] B. P. PreethiDodawadReshmaPyatiAnand, Evaluation of Flow Rate, pH, Buffering Capacity,Calcium, Total Proteins and Total Antioxidant Capacity Levels of Saliva in Caries Free and Caries Active Children: An In Vivo Study, *Ind J ClinBiochem*, Oct-Dec 2010; 25(4):425–428.
- [18] D Jayaraj, S Ganesan, Salivary pH and Buffering Capacity as Risk Markers for Early Childhood Caries: A Clinical Study, 10.5005/jp-journals-1005-1307.
- [19] S Dogra, D Bhayya, R Arora, D Singh, D Thakur, Evaluation of physio-chemical properties ofsaliva and comparisonof its relation withdental caries , *Journal of Indian society of pedodontics and preventive dentistry*, Oct-Dec 2013; 31(4): 221-224.
- [20] Ghasempour M, Rajabnia R, Irannejad AHamzeh M, Ferdosi E, Bagheri M. Frequency, biofilmformation and acid susceptibility of streptococcus mutans and streptococcus sobrinus in saliva of preschool children with different levels of caries activity. *Dent Res J (Isfahan)*. 2013;10(4):440-5.