Original

Relation of Inorganic Phosphate Concentration to Buffering Capacity in Whole Mouth Resting Saliva

安静唾液における無機磷酸塩濃度と緩衝能との関連性

Itsuo UEDA*

上 田 五 男*

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概要：著者らは先の報告で安静全唾液の緩衝能と流出量との間に唾液 pH を固定した場合、両者間の偏相関関係に統計学的有意な負の相関性のあることを示した（口腔衛生会誌，34；193，1984.）。そこで，安静全唾液の緩衝能発現における無機磷酸塩濃度の役割を明らかにするため本研究を企図した。

男子歯科学学生60名（23～26歳）を対象とし，可及的安静な条件下で全唾液を採取し，いわゆる安静唾液の流出量，緩衝能（Buffer pH）および無機磷酸塩濃度の測定を行った。

唾液流出量は，0.08～0.96ml/min の範囲に分布し，平均値は0.33ml/min であった。全被検者の約70%の流出量が0.1～0.39ml/min の範囲に含まれた。

唾液流出量と無機磷酸塩濃度の間には統計学的有意な負の相関関係が認められた（ $r = -0.46$ ， $p < 0.001$ ）。しかし，唾液流出量と緩衝能値との間には一定の関係はみられなかった。

一方，唾液無機磷酸塩濃度と緩衝能値との間には統計学的有意な正の相関関係がみい出された（ $r = 0.33$ ， $p < 0.01$ ）。

以上の結果は，安静全唾液の緩衝作用発現において唾液無機磷酸塩が一定の積極的な役割を演じていることを強く示唆している。

Key words： Resting whole saliva, Buffer capacity, Flow rate, Inorganic phosphate concentration

索引用語：安静全唾液，緩衝能，流出量，唾液無機磷酸塩濃度

Introduction

The significance of the protective action of saliva from decrease in the plaque pH after glucose ingestion has been reported by Stephan¹⁾ and others^{2,3)}, and they also pointed out its protective effect against dental caries.

It is generally believed that the best established relationship between salivary factors and dental caries is the buffering action of saliva⁴⁾. The role of bicarbonate in the buffering capacity of stimulated whole saliva is well known⁵⁻⁸⁾. However, there are relatively few reports on the buffer effect of the so-called

* Department of Preventive Dentistry, School of Dentistry, Higashi Nippon Gakuen University (Chief: Prof. Nobuyoshi ITO)

* 東日本学園大学歯学部口腔衛生学教室（主任：井藤信義教授）

resting whole saliva.

In the present study, we have examined the relationship between the buffer capacity and inorganic phosphate concentration in the resting whole saliva, and have also investigated the possible contribution of the latter to the buffering capacity of whole saliva.

Materials and Methods

Resting whole saliva samples were collected in the so-called resting state with as little movement as possible for fifteen min by the spitting method⁹⁾ from 60 healthy, unmedicated male dental students between the ages of 23 and 26 years in a quiet room. The collection time was between 10 a.m. and 11 a.m., at least two hr after a meal. The subjects were made to rinse once with 10 ml of distilled water, and then to close the lips with the heads tilted down slightly. The accumulated saliva in the floor of the mouth was swallowed, after which the fresh saliva flowing naturally into the mouth was spit into a graduated test tube fitted with a funnel which was held near the mouth. The flow rate was recorded as ml per min. The measurement of buffering capacity of whole saliva was undertaken with the buffer pH method¹⁰⁾ in which 0.3 ml of 0.1 N lactic acid was added to every 2 ml of the tested saliva and after a thorough mixing the solution's pH was measured with a pH electrode. The coefficient of correlation(*r*) between the buffer pH method and the Ericsson method¹¹⁾ was 0.97.

The concentration of inorganic phosphate in saliva was determined by the method of Bartlett¹²⁾. The samples of saliva were centrifuged at 10,000 r.p.m. for 20 minutes and the proteins were removed by precipitation with cold 10 per cent TCA followed by Millipore filtration.

All summarized results are given as arithmetic means with inter-individual standard deviations. The correlation coefficients were calculated with Peasson's formula for product-moment correlation coefficients.

Results

Table 1 lists the mean values of flow rate, buffering capacity, and inorganic phosphate concentration in the resting whole saliva. The distribution of the flow rates of saliva is given in Fig. 1. The values were in the range of 0.08-0.96 ml/min with a mean value of 0.33 ml/min. The distribution of the results of buffering capacity of saliva is shown in Fig. 2. The buffer effect (buffer pH), expressed as final pH, gives a mean value of pH 4.0 with a range of pH 3.35-5.01. Fig. 3 shows the distribution of inorganic phosphate concentrations in saliva. The concentra-

Table 1 Mean values of flow rate, buffer capacity and inorganic phosphate concentration in resting whole saliva

	Mean $\pm$ SD
Flow rate, ml/min	0.33 $\pm$ 0.17
Buffer capacity, pH	4.00 $\pm$ 0.13
Inorganic phosphate, mg/dl	16.6 $\pm$ 5.6

Fig. 1 Frequency distribution of the flow rates in resting whole saliva.

Fig. 2 Frequency distribution of the buffer capacity values in resting whole saliva.

Fig. 4 The relation between the flow rate and inorganic phosphate concentration of resting whole saliva.

Fig. 3 Frequency distribution of the inorganic phosphate concentrations in resting whole saliva.

Fig. 5 The relation between the inorganic phosphate concentration and buffer capacity of resting whole saliva.

tions varied from 10.1 to 34.9 mg/dl and the average value was 16.6 mg/dl (1.75 mM). 80% of the total was within 10.0-17 mg/dl.

Fig. 4 shows the relationship between the flow rate and inorganic phosphate concentration in resting whole saliva. A statistically significant inverse correlation was found between the two.

Fig. 5 shows the relationship between the buffering capacity and phosphate concentration in the whole saliva. The direct correlation between the two was statistically significant: the higher the former, the higher the latter and vice versa.

There was not a uniform relationship between the buffer capacity and flow rate in the resting whole saliva.

Discussion

The flow rate of resting whole saliva in this study is in good agreement with the findings of Beck and Wainwright¹³⁾ in the age groups of 20-29 years. 70 % of the total samples was in the range of 0.1-0.39 ml/min.

The present study showed a significant inverse relationship between the flow rate and inorganic phosphate concentration in resting whole saliva. Some investigators¹⁴⁻¹⁶⁾ reported that the phosphate concentration was inversely related to the flow rate in human parotid saliva. Likewise, whole resting saliva contained a more inorganic phosphate than stimulated saliva^{17,18)}.

Dawes¹⁴⁾ showed that at flow rates of 0.25, 0.50, and 1.00 ml/min the mean phosphate concentrations of parotid saliva were 4.38, 3.61, and 3.10 mM, respectively. Our previous data¹⁷⁾ showed a significant correlation ($r = -0.58$; $p < 0.01$) between the flow rate and inorganic phosphate concentration in centrifuged resting whole saliva.

The present study showed a significant direct correlation between the buffer capacity and inorganic phosphate concentration in the resting saliva. This clearly shows that the concentration of inorganic phosphate contributes partially to the buffering action of resting whole saliva. Our previous investigation¹⁰⁾ has already shown that in resting whole saliva there is a significant inverse relation between the buffer capacity and flow rate in the partial correlational analysis if the pH of saliva is fixed to remove its effect. This strongly suggests an essential role of the phosphate buffer system in the buffering action of resting whole saliva.

The importance of salivary buffering capacity as endogenous resistant factor against dental caries has already been confirmed^{8,4)}. Wah Leung⁵⁾ showed that bicarbonate was the most important buffer in saliva, and he suggested that the phosphate and proteins make up the remaining buffering activity, particularly at lower pH ranges than 7.2 to 6.8. For the buffer capacity in resting whole saliva Lilienthal⁶⁾ showed that the proportion of total buffering power which is contributed by the bicarbonate system varied from 30 to 85 per cent, suggesting that the only other buffer system having appreciable activity is probably phosphate.

On the other hand, Shannon et al.¹⁹⁾ reported that the concentrations of inorganic phosphate were higher in caries resistant than in caries susceptible individuals.

It seems that the salivary phosphate, especially in the resting saliva, would act as a caries resistant factor to an extent varying with its salivary content.

Finally, it would be of interest to investigate the influence of phosphate concentration and buffering capacity in the resting saliva on the plaque pH changes which are related to the caries process.

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