Influence of Salivary pH and Urea Level on Calculus Formation - A Clinical Study

Abstract

Aim: To evaluate the influence of salivary pH and urea level on calculus formation in heavy and mild calculus formers

Materials and methods: pH of saliva was recorded, and urea level of unstimulated whole saliva was estimated, Calculus and plaque scores were recorded.

Results: Statistically significant correlation was evident between salivary urea concentrations and the corresponding pH values in heavy calculus formers, but not in mild calculus formers

Conclusion: Salivary urea has a major influence on calculus formation where as the influence of salivary pH was insignificant.
INTRODUCTION

One of the major oral disease processes is the formation of calculus. Dental calculus is composed of inorganic components and organic matrix. Supragingival and subgingival calculus contain 37% and 58% mineral content by volume, respectively. The matrix of supragingival calculus constitutes 15.7% of the calculus dry weight and contains 54.9% protein and 10.2% lipid.

Calculus is mineralized dental plaque and mineralization can only occur if the fluid phase of plaque is supersaturated with the components of calculus. Saliva and plaque fluid are normally supersaturated with respect to various calcium phosphates, except when fermentable carbohydrates are being consumed, and thus most people are susceptible to calculus deposition, albeit at different rates. Although much research has been carried out to determine possible mechanisms for formation and methods for inhibiting the process, no complete correlations exist. The formation, development, and dissolution of hard deposits such as calculus are complex processes that involve numerous calcium phosphate phases as well as the interaction of these ions with organic molecules.

The saliva is a complex fluid containing a variety of mucosal host defense factors from the different salivary glands and the crevicular fluid. There are glucose and nitrogenous products, such as urea and ammonia. The pH of saliva has a wide range of 5-8. Studies had reported the importance of alkaline pH for deposition of calcium phosphate, thereby promoting plaque mineralization. The variable pH conditions in plaque, expressed in terms of free ionic concentrations, will markedly alter the supersaturations with respect to typical calcium phosphate precursor phases such as dicalcium phosphate dihydrate (DCPD) and octacalcium phosphate (OCP). An early explanation for calculus formations suggested that chemical changes occurred in the mouth which reduced the so-called "solvent power" of the saliva for calcium salts. Much of the work in this area has tended to be contradictory. In several studies, elevations in calcium and phosphorus concentration have been reported in heavy calculus formers in other studies no differences were noted.

Urea is a buffer present in total salivary fluid which is a product of aminoacid and protein catabolism that causes a rapid increase in biofilm pH by releasing ammonia and carbon dioxide when hydrolyzed by bacterial ureases. Ammonia produced from ureolysis of urea contributes to an increased plaque pH that is an essential factor in natural calculus formation. A urea-induced pH response, which was the inverse of the Stephan pH curve induced by sucrose, has been observed in an in vitro biofilm culture system called "artificial mouth". The ureolytic pH response (an increase in plaque pH by the production of ammonia from urea) promotes calculus formation by increasing the saturation degree of calcium phosphate in plaque fluid.

Thus, there is general agreement that supragingival deposits derive most of their mineral content and part of their matrix from saliva. It would, therefore, seem logical to examine salivary composition in heavy and light calculus formers as a means of identifying factors responsible for individual susceptibility. The present study was, therefore, designed to evaluate the effect of salivary pH and urea level on calculus formation in heavy as well as mild calculus formers.

MATERIALS AND METHODS

The subjects constituted the volunteers taken from among the patients coming to the out-patient unit of Department of Periodontics, Mamata Dental College, Khammam. The inclusion criteria followed were:

1. Age group 18-55 years
2. Systemically healthy
3. Not undergoing any drug therapy
4. Not had any periodontal therapy in the past 3 months, and no discernible calculus formation for at least a year
5. Not had any periodontal therapy in the past 3 months, but forming calculus at a rate requiring frequent oral prophylaxis over a year
6. Willingness to participate in the study

Based on objective evaluation of the oral hygiene condition, subjects were divided into two groups:
Group A- Mild calculus formers (mild discernible calculus formation for at least a year)
Group B- Heavy calculus formers (forming calculus at a rate requiring frequent oral prophylaxis over a year)

15 subjects were assigned to each group. Informed consent was obtained from each subject.

The subject refrained from oral hygiene, eating, drinking, or smoking for at least 2 hours before clinical parameters were recorded.

The subject was asked to sit still and relaxed, with head slightly tilted down, and minimal oral musculature activity, including swallowing. Salivary pH was recorded by using pH indicator strips [Dental Saliva pH indicator strips pH 5.0 - 8.0; gradation 0.2; color coded (Global Care—Made in Japan), (Figure. 1)] placed near the opening of the salivary duct at the floor of the mouth. Unstimulated whole saliva was allowed to pool in the floor of the mouth over 5 minutes. The subject was asked to expectorate in a sterile container as passively as possible.
and close the lid (Figure 2). Thereafter, biochemical estimation of salivary urea was performed with the help of manual modified Berthelot method using Urea Kit* [Coral Clinical Systems- CREST Biosystems, (Made in India)].

The calculus scores were recorded according to Calculus Surface Severity Index\textsuperscript{11}. Thereafter, plaque was disclosed (using Alphaplac\textsuperscript{®} DPI-India) and plaque scores recorded based on Turesky-Gilmore-Glickman modification of Quigley- Hein Plaque Index\textsuperscript{12}.

**STATISTICAL ANALYSIS:**

The information was tabulated and Independent t- test (Student’s) was performed for pH and urea levels with the help of SPSS package (SPSS, Inc, Chicago, IL, USA) version 12.0.

**RESULTS**

At the end of the study, of 30 subjects, the mean and standard deviation for Group A and Group B are shown in table 1 and the independent t- test was shown in table 2, there was no statistically significant difference between pH levels in both the groups (p=0.201) There was a significant difference between salivary urea concentrations in both the groups (A- 0.019, B- 0.023) which was found to be statistically significant.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group A</th>
<th>Group B</th>
<th>t-test</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calculus formation</td>
<td>Mild calculus formers</td>
<td>15</td>
<td>31.0667</td>
<td>9.99619</td>
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<tr>
<td></td>
<td>Heavy calculus formers</td>
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<td>49.8667</td>
<td>27.59365</td>
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<tr>
<td></td>
<td>Mild calculus formers</td>
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<tr>
<td></td>
<td>Heavy calculus formers</td>
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<td>7.3200</td>
<td>.44593</td>
</tr>
</tbody>
</table>

**Table-2 Comparison of salivary urea and pH among group A and B**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Group A</th>
<th>Group B</th>
<th>t-test</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
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<td>Urea</td>
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<td>-2.481</td>
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<td>.023</td>
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<tr>
<td>pH</td>
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<td>-1.310</td>
<td>.201</td>
<td>.201</td>
</tr>
</tbody>
</table>

# Independent t-test, Significance p<0.005

**DISCUSSION**

Considerable effort has been put in by various authors in the past to understand the mechanism of calculus formation, and the factors influencing it.

The present study was carried out to evaluate the influence of salivary pH and urea levels on calculus formation in heavy as well as mild calculus formers. It was evident that there was a significant difference in the salivary urea concentrations of both the groups.

However, only 60% of the subjects with heavy calculus reported with high urea concentration (≥ 36 mg/dL), while the remaining subjects had normal or only marginally high urea levels. It was found that there is a significant correlation between urea...
concentration in saliva and corresponding pH values in heavy calculus formers, which reflected in considerable amount of calculus deposition in these subjects.

The importance of alkaline pH for deposition of calcium phosphate, thereby promoting plaque mineralization was also reported by Wong L et al. The salivary pH in chronic generalized periodontitis was found to be statistically significantly high compared to healthy gingiva.

The present study demonstrated a positive and significant correlation between urea levels and calculus formation in both the groups, which is in accordance with the study conducted by Gupta et al.

Most probably the nitrogenous material in saliva is broken down to smaller units such as the conversion of urea to ammonia, amino acids to ammonia and other products, and even the breakdown of mucin and other proteins to smaller units. While oral ureolytic activity is ubiquitous, it varies quantitatively between individuals' plaque. This variation is probably related to variations in the flora, since common plaque organisms differ greatly in their capacity to degrade urea. Plaque may be regarded as a promotion of a natural process. Focal alkalization has long been considered a factor in the natural mineralization of plaque during its transformation to calculus, with the production of ammonia from salivary urea one of the means by which the pH is raised. Application of urea solutions to plaque in situ causes a rapid rise in supragingival plaque pH. This rise has been attributed to the formation of ammonia by the ureolytic activity of the plaque bacteria. Both the levels of urea in saliva and of ammonia in plaque are sufficient to account for the high pH of fasting plaque.

In contrast to our study, a double-blind, cross over study of three months' frequent use of sugar free chewing gum-with or without urea-neither promotes nor inhibits calculus formation.

However, the variation in calculus formation in different individuals may also be related to variation in salivary flow rate in different regions of the oral cavity. Other components of saliva may also contribute to calculus deposition, such as $Ca^{2+}$ and $P$-super saturation, buffering capacity, protein content, and certain organic acids.

**CONCLUSION:**

It can be concluded that salivary urea has a major influence on calculus formation, and even though evidence shows that pH has a role in calculus formation, the present study did not show significant relation between pH and calculus formation, which could be because of the small sample size. Therefore further investigation using a large sample size may reveal the significance of pH in calculus formation.

**FIGURES:**

![Saliva pH indicator strips](image)
REFERENCES


