



(RESEARCH ARTICLE)



## Comparison of salivary calcium, dental caries, and oral health status among healthy and diabetes mellitus patients: A cross-sectional study

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### Abstract

Diabetes mellitus is a disease characterized by increased blood glucose levels. Saliva can often be viewed as a reflection of the physiological state of the body. The sixth recognized complication of diabetes mellitus is periodontitis. Diabetes patients also have a higher risk of getting dental caries and gingivitis. This short-term clinical study was performed to assess salivary calcium levels, dental caries, and oral health status in diabetes mellitus patients. Our results showed that there is a statistically significant difference in salivary calcium level, Simplified Oral Hygiene index (OHI S), and the DMFT (Decayed, Missing, Filled teeth index) among the healthy and diabetes mellitus patients.

**Keywords:** Saliva; Diabetes Mellitus; Oral Hygiene Index; DMFT index; Salivary Calcium

### 1. Introduction

Diabetes mellitus (DM) is a disease characterized by increased blood glucose levels. This increase in the levels is the result of insufficient insulin secretion or increased cellular resistance to insulin. Some common clinical consequences of diabetes mellitus are retinopathy, nephropathy, neuropathy, macrovascular disease, and slow wound healing. [1] Type 1 diabetes mellitus (T1DM) and Type 2 diabetes mellitus (T2DM) are the two primary subtypes of DM, and both are typically brought on by faulty insulin secretion (T1DM) and/or action (T2DM). 90% of persons worldwide with DM have T2DM. [2]

Diabetes mellitus has been linked to a number of oral conditions including Dry mouth, dental caries, periodontal disease, gingivitis, oral candidiasis or thrush, burning mouth syndrome, taste disturbances, rhinocerebral zygomycosis (mucormycosis), aspergillosis, oral lichen planus, geographic tongue among others. There are numerous studies on the connections between diabetes and periodontal health that demonstrate how periodontal disease can impair diabetes control and how diabetes can result in poor periodontal health.[3] Periodontal attachment loss in patients with poorly controlled diabetes is also greater in comparison to that with well-controlled diabetes or healthy individuals.

Dental caries is a multifactorial disease. Diabetes patients have been reported to experience it more frequently and severely than non-diabetic individuals.[4] Dental caries is caused by demineralization which is triggered by the accumulation of microbial plaque flora, an increase in salivary glucose levels, diminished levels of salivary calcium, and a decrease in the flow rate of saliva. Remineralization favoring factors namely calcium, phosphates, and fluorides are known to maintain the integrity of enamel surfaces and intraoral remineralization capacity of saliva.[5] Hence, Estimation of salivary calcium levels may provide information regarding the extent or prevalence of caries in the individual.

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Saliva, the properties of which are mainly determined by secretions from the major and minor salivary glands, performs a large number of functions that are indispensable for both oral and general health. Saliva is principally composed of water (>99%) but has several minor components including cytokines, digestive enzymes, growth factors, immunoglobulins, mucus, antibacterial peptides, bacterial cells, salts, and low molecular weight metabolites. [6]

It has been shown that many inflammatory biomarkers associated with oral diseases have been detected in saliva, such as interleukins-1b, -6, -8 (IL-6, IL-1, IL-8), tumor necrosis factor-alpha (TNF-a), and matrix metalloproteinases (MMP)-8 and -9 and substances like salivary glucose, amylase, calcium, and phosphorus have been shown to have a direct link with the oral health of the individual.[4]

Hence, this short-term clinical study was performed to assess salivary calcium levels, dental caries prevalence, and oral health status in healthy and diabetes mellitus patients.

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## 2. Material and methods

A total of 60 study subjects were recruited from the patients reporting to the Outpatient department. Subjects were grouped under two categories.

- GROUP 1 - HEALTHY PATIENTS
- GROUP 2 - DIABETIC PATIENTS

### 2.1. Inclusion Criteria

- Group I: 30 – 70 Yrs, Any gender, Healthy subjects without any systemic diseases, HbA1c values: < 6.5%
- Group II: 30 – 70 Yrs, Any gender, with a history of diabetes, HbA1c values : ≥6.5%

### 2.2. Exclusion Criteria

- Group I - Participants with a history of diabetes including gestational diabetes and other metabolic diseases are excluded.
- Group II – Participants without a history of diabetes and the presence of any other systemic diseases are excluded.

Common exclusion criteria: Participants who are under medications other than diabetics, and ongoing dental treatments.

### 2.3. Clinical Examination

The oral hygiene status of the individual is measured by employing the oral hygiene simplified index(OHI-S). The dental caries will be recorded by employing the Decayed – Missing- Filled tooth (DMFT) index.

### 2.4. Sample Collection

Method for collection of Saliva sample - The participants will be asked to wash their mouths with tap water and to spit two or three times, after which they will be asked to spit the saliva pooled in their mouths for the following 10 minutes into the sterile sample collection container.

### 2.5. Estimation Of Salivary Calcium Level

The salivary calcium estimation is performed using the OCPC method using a commercially available OCPC kit. Calcium, in an alkaline medium, combines with OCPC to form a purple-colored complex. The intensity of the color formed is directly proportional to the amount of calcium present in the salivary sample. Three clean and dry test tubes will be labeled as blank (B), standard (S), and test (T).0.02 ml of Distilled water, standard solution, and test solution were pipetted out into the test tubes B, S, T respectively. Then, 0.5 ml each of buffer and color reagent will be added to all three test tubes and the contents will be mixed thoroughly and incubated at 37°C for 5 minutes. The optical density of standard (S) and test (T) is measured against blank (B) on a colorimeter at 570 nm and the values are recorded.

### 2.6. Statistical Analysis

Descriptive statistics were used to assess the mean and standard deviation among the study variables. Inferential statistics were obtained using an unpaired t-test to compare the mean values between diabetic and healthy patients. To

analyze the data SPSS software was used. The significance level is fixed at 5% ( $\alpha = 0.05$ ). P-value  $<0.05$  is considered to be statistically significant.

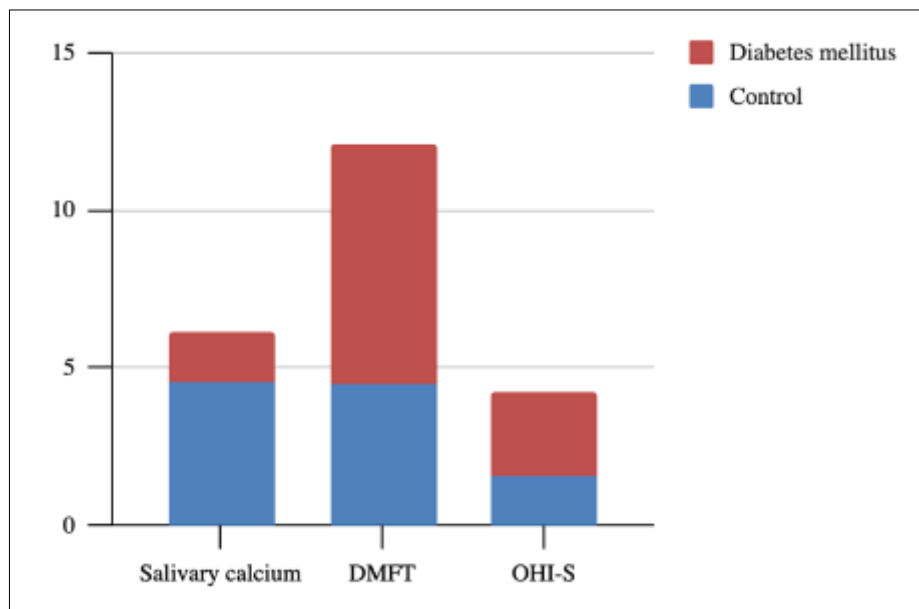
### 3. Results

The mean and the standard deviation of the salivary calcium levels of healthy and diabetes patients were found to be  $4.5222 \pm 3.88789$ ,  $1.6069 \pm 1.20726$  respectively. The mean and the standard deviation of the OHI-S index of healthy and diabetes patients were found to be  $1.5973 \pm 0.80413$ ,  $2.6597 \pm 0.71437$  respectively. The mean and the standard deviation of the DMFT index of healthy and diabetes patients were found to be  $4.5000 \pm 2.75118$ ,  $7.6000 \pm 2.81131$  respectively.

Statistical analysis showed that a statistically significant difference was noted for salivary calcium levels among healthy and diabetes mellitus patients with a p-value of 0.0018. The Simplified Oral Hygiene Index (OHI S) and the DMFT values among healthy and diabetes mellitus patients were also statistically significant with a p-value of 0.0011 and 0.0016 respectively [Table 1].

**Table 1** Unpaired t-test for Salivary calcium level Simplified oral hygiene index (OHI-S) and DMFT values among Control and Diabetes mellitus patients

	SALIVARY CALCIUM	DMFT	OHI-S
CONTROL	$4.5222 \pm 3.88789$	$4.5000 \pm 2.75118$	$1.5973 \pm 0.80413$
DIABETES MELLITUS	$1.6069 \pm 1.20726$	$7.6000 \pm 2.81131$	$2.6597 \pm 0.71437$
t VALUE	4.492	4.019	5.660
P VALUE	0.0018*	0.0016*	0.0011*



**Figure 1** Comparative analysis of inorganic salivary calcium, DMFT, OHI-S among control and diabetes mellitus patients

### 4. Discussion

Diabetes mellitus (DM) is a collection of long-term conditions characterized by insulin shortage, cellular resistance to insulin action, or both, which leads to hyperglycemia and other related metabolic abnormalities. [7] Diabetes affected 2.8% of people worldwide in 2000 and may affect 4.4% by 2030. [8] Dental caries, gingivitis, periodontitis, impairment

of the salivary glands, altered taste, and infections of the oral mucosa such as lichen planus, recurrent aphthous stomatitis, and candidiasis have all been linked to DM.

The sixth recognized complication of diabetes mellitus is periodontitis.[9]In our study, the oral hygiene (OHI -S) of the patients with diabetes was significantly lower than healthy patients with a p-value of 0.0011 suggesting that diabetic patients with poor oral hygiene are at a higher risk for periodontitis. This is due to a number of harmful side effects that diabetes mellitus has on peripheral blood arteries, such as vascular proliferation or a reduction in blood flow caused by the thickening of the basement membrane. It has been stated that chronic periodontitis may be induced or aggravated as a response to lipid peroxidation in various systemic diseases, including diabetes. [10]

According to some authors, diabetic people have a three times higher chance of developing dental caries than non-diabetics. Even though both diseases are linked to the consumption of dietary carbohydrates, there has been far less focus on the connection between diabetes and dental caries, especially among adults. [11] Our study showed a significantly higher incidence of caries in diabetic patients with a mean value of  $7.600 \pm 2.81131$  to that of  $4.500 \pm 2.75118$  in healthy patients. This may be because diabetes mellitus has been shown to affect salivary composition and function, which ultimately affects the health of the oral cavity and teeth.[11]

There is evidence that saliva from people with low caries has higher levels of the following: pH and buffering, higher levels of calcium and phosphorus, higher concentrations of ammonia, high ATP and fructose diphosphate concentrations, increased bacterial O<sub>2</sub> absorption and aldolase activity increased antibacterial activity overall, increased opsonin activity, increased antimicrobial activity against lactobacilli and streptococci, Higher proportion of intact leukocytes and a different ratio of epithelial to leukocyte cells.[7] Calcium in saliva serves as a key mineral that prevents tooth dissolution by supplying a constant and continuous supply to the affected areas of teeth. Failure to regulate blood glucose in patients with diabetes has a significant negative impact on salivary calcium levels.[11] In the present study, the salivary calcium levels of healthy and diabetes mellitus patients were evaluated. Our study showed that salivary calcium in diabetic patients was lower with a mean of  $1.6069 \pm 1.20726$  to that of  $4.5222 \pm 3.88789$  in healthy patients. From the above two observations it can be concluded that with an increase in diabetes, there is a decrease in salivary calcium level which in turn increases the DMFT value.

Therefore, as diabetes is an adult disease, when uncontrolled it becomes an important risk factor for severe periodontitis and a higher risk for caries.

#### *Limitation*

The primary drawback of the study was the small sample size, which requires a more thorough study with a larger sample size to support these values. Correlation between Serum calcium and salivary calcium with the dental caries status of diabetes mellitus patients is necessary in order to know about the true nature of the disease

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## **5. Conclusion**

Diabetes is one of the most common systemic diseases affecting an individual, and monitoring salivary biomarkers could become an important complement to clinical examinations in diabetes patients. Our findings have also proved that salivary calcium levels are altered in diabetic patients with a concurrent increase in dental caries and poor oral health. Hence, assessing these markers can aid dentists in providing optimal care for their patients.

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## **Compliance with ethical standards**

### *Disclosure of conflict of interest*

All authors declare that they have no conflict of interest.

### *Statement of informed consent*

Informed consent was obtained from all individual participants included in the study.

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