Original Article

SALIVARY ELECTROLYTE AS A BIOMARKER IN CARIES ACTIVE TYPE II DIABETES - A COMPARATIVE STUDY

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Abstract:

Background : Diabetes Mellitus and dental caries association has been determined successfully but the electrolyte imbalance responsible is still under investigation.

Aims : This study aims to evaluate the salivary electrolyte concentration in non-diabetic and diabetic patients with active dental caries.

Methods and Material : 60 diabetic individuals with active dental caries were selected fulfilling the inclusion criteria as the study group with equal number as controls. 5ml of saliva was collected from the patient, centrifuged and the supernatant obtained was subjected to subsequent analysis for Na⁺, K⁺ and Cl⁻ ions concentration in saliva.

Statistical analysis: The intergroup comparison correlating the salivary electrolyte concentration was done using student's 't' test. A 'p' value of 0.05 or less was considered significant. Results are presented as mean $\pm$ standard deviation ($\bar{X} \pm SD$).

Results: The mean value of sodium, potassium and chloride ions in diabetic patients was 0.97 ± 0.14 , 10.40 ± 0.9 and 135.4 ± 3.67 respectively ($p < 0.05$). However, in non-diabetics it was 0.23 ± 0.07 , 6.87 ± 1.86 and 96.24 ± 4.85 respectively ($p < 0.05$).

Conclusions: From our study it is evident that salivary levels of electrolytes show a positive relationship between diabetics and non-diabetics with active dental caries. However, a longitudinal data might help in better understanding of this association.

Keywords : Diabetes mellitus, sodium, chloride, potassium, electrolytes, active dental caries

Introduction :

Diabetes mellitus is the most routinely encountered disease among the various systemic diseases¹. Diabetes is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. There are three major types of diabetes: type 1 diabetes, type 2 diabetes, and gestational diabetes¹

Type II diabetes mellitus is characterized by high blood

glucose in the context of insulin resistance and relative insulin deficiency. This is in contrast to diabetes mellitus type I, in which there is an absolute insulin deficiency due to

destruction of islet cells in the pancreas. Type I diabetes accounts for 5-10% and Type II diabetes accounts for 90-95% while gestational diabetes occurs in about 2%-5% of all pregnancies and may improve or disappear after delivery¹. Gestational diabetes is fully treatable, but requires careful medical supervision throughout the pregnancy². The World Health Organization has estimated that in 1995, 19.4 million individuals were affected by diabetes in India and these numbers are expected to increase to 57.2 million by the year 2025 i.e. one sixth of the world total¹. The revised figures are 80.9 million by the year 2030³⁻⁴.

Dental caries has been more prevalent and even severe in diabetic patients than non-diabetics⁵. Diabetic patients are prone to complications such as periodontal disease (gingivitis, periodontitis), dental caries, salivary

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dysfunction, dry mouth (Xerostomia), oral mucosal diseases, oral infections (candidiasis)⁶⁻⁷. Approximately 5% of all patients seen in dental clinics are reported to have diabetes⁸.

Dental caries is a complex disease process that afflicts a large proportion of the world's population, regardless of gender, age and ethnicity, although it does tend to affect more individuals with a low socioeconomic status to a greater extent⁹. Caries is said to be a multifactorial disease. Different individuals of the same age, sex, race and geographic area sustaining on the similar diets under the same the living conditions accentuate the complexity of the caries problem¹⁰.

Saliva plays an important role in maintaining the equilibrium of the oral ecosystem. Saliva is often referred to as the "mirror of the body" as it is the indicator of health not just in the oral cavity but also throughout the body¹¹. Whole saliva contains locally produced as well as serum-derived markers that have been found to be useful in the diagnosis of a variety of systemic disorders. Analysis of saliva can offer a cost-effective approach for the screening of large populations, and may represent an alternative for patients in whom blood drawing is difficult, or when compliance is a problem¹².

Various trace elements like sodium, magnesium, potassium, chloride, zinc etc., are present in our body fluids. These trace elements are also referred to as electrolytes since they carry electric charges. It is important to maintain the balance of electrolytes in our body. They are what our cells use to maintain voltage across their cell membranes and carry electrical impulses across themselves and to other cells¹³.

A limited amount of study is undertaken on salivary electrolyte concentration in non-diabetic and diabetic patients. Considering this present study aims to evaluate the salivary electrolyte concentration in non-diabetic and diabetic patients with dental caries.

Subjects and Methods:

This study was approved by the Institutional Ethical

Committee with Ethical Certificate number ABSM/EC/81/2011. 12,500 healthy adult patients coming to the OPD of Department of Conservative Dentistry and Endodontics, A.B. Shetty Memorial Institute of Dental Sciences under the age group of 25-50 years from December 2012- June 2013 were screened for active dental caries and out of which 60 patients who fulfilled the inclusion criteria with Fasting Blood Glucose >126 mg/dl were the study group and compared to 60 age and gender matched control group (non-diabetic) with a fasting blood glucose <125mg/dl. Hence, a total of 120 subjects with active dental caries were selected for the study. Their caries status was assessed according to World Health Organization "W.H.O recommendations 1997" to calculate dental caries index¹⁴.

Inclusion criteria

- | A known Type II Diabetic mellitus patient yielding positive results for fasting blood glucose (>126 mg/dl) under the age group of 25-50 years.
- | Patients with active dental caries in DMFT index yielding decayed teeth > 10

Exclusion Criteria

- | Patient with systemic condition other than diabetes.
- | Patient under any reported Xerostomia, anti-psychotic, anti-cholinergic, anti-hypertensives, anti-secretoagogues and thyroid medication
- | Patients who have consumed alcohol or smoked in last 24 hours
- | Pregnant women
- | Patient on radiotherapy
- | Patients with kidney malfunction or on dialysis.
- | Patients taking any caries preventive regimen like fluoride tooth paste, fluoride rinses or NaF/calcium tablets.

Calculation of DMFT

The smooth and occlusal surfaces of teeth were cleaned with soft bristle brush, dried and examined. DMFT score was calculated.

Collection saliva for salivary analysis

Unstimulated whole saliva (Resting Saliva) from each

subject was expectorated, into sterile tubes, 2 hours after breakfast, after a single mouth rinse with 15 ml of distilled water to wash out exfoliated cells. 5ml of saliva was collected from the patient, centrifuged and the supernatant obtained was stored at 4° C for subsequent analysis.

Estimation of electrolytes in saliva

Na⁺, K⁺ and Cl⁻ were determined using Elyte 3 Kit (Crest Biosystems, India).

Estimation of sodium levels in saliva

Sodium is precipitated as a triple salt with magnesium and uranyl acetate. The excess of uranyl ions are reacted with ferrocyanide in an acidic medium to develop a brownish color. The intensity of the colour produced is inversely proportional to the concentration of sodium in the sample.

Estimation of potassium levels in saliva

Potassium reacts with sodium tetraphenyl boron in a specifically prepared buffer to form a colloidal suspension. The amount of turbidity produced is directly proportional to the concentration of potassium in the sample.

Estimation of chloride levels in saliva

Chloride ions combine with free mercuric ions and release thiocyanate from mercuric thiocyanate. The thiocyanate released combines with the ferric ions to form a red brown ferric thiocyanate complex. Intensity of the colour formed is directly proportional to the amount of chloride present in the sample.

Statistical analysis :

Student 't' test was used to correlate the salivary electrolyte concentration in non- diabetic and diabetic patients with active dental caries. A 'p' value of 0.05 or less

Table 1: Comparison of salivary sodium, potassium and chloride levels in non-diabetic and diabetic adults with dental caries

Groups	Non-diabetic	Diabetic	'P' Value
Parameters			
Sodium (mmol/L)	0.23±0.07	0.97±0.14	P<0.0001
Potassium (mmol/L)	6.87±1.86	10.40±0.98	P<0.0001
Chloride (mmol/L)	96.24±4.85	135.4±3.67	P<0.0001

was considered significant. Results are presented as mean ± standard deviation (X ± SD). Prism 3.0 software was used to analyse the data.

Figure 1: Comparison of salivary sodium levels in non-diabetic and diabetic adults with dental caries

Figure 2: Comparison of salivary potassium levels in non-diabetic and diabetic adults with dental caries

Figure 3 : Comparison of salivary chloride levels in non-diabetic and diabetic adults with dental caries

Results:

The concentration of sodium (Na⁺), potassium (K⁺) and chloride (Cl⁻) ions in saliva was higher in diabetic patients when compared to that of non- diabetic patients with active dental caries. (Table 1) The mean value of Na⁺ ions in saliva of non- diabetic patients was 0.23±0.07 and in

diabetic patients was found to be 0.97 ± 0.14 . 'p' value is statistically significant ($p < 0.05$) (Figure 1). The mean value of K^+ ions in saliva of non-diabetic patients was 6.87 ± 1.86 and in diabetic patients was found to be 10.40 ± 0.98 . 'p' value is statistically significant ($p < 0.05$) (Figure 2). The mean value of Cl^- ions in saliva of non-diabetic patients was 96.24 ± 4.85 and in diabetic patients was found to be 135.4 ± 3.67 . 'p' value is statistically significant ($p < 0.05$) (Figure 3).

Discussion:

Saliva represents an increasingly useful auxiliary means of diagnosis.¹⁵ It is a body fluid with complex composition and specific roles. Human saliva is a unique secretion of major and minor salivary glands and helps in maintaining the normal physiologic functions of orobiological structures. Dawes et al stated the importance of accuracy in saliva measurements. Presence of circadian rhythm and fasting has been reported to influence salivary flow rate, which makes time-point of the test critical. Hence saliva was collected between 9:00am and 11:00 am.¹⁶ The oral hygiene status and periodontium status were checked before their enrollment into the study. The patients were referred to Department of Periodontics for scaling and curettage and recruited into the study only after the reversal of their periodontal condition.

The analysis of biochemical constituents in saliva is of great help in diagnosis of diseases in oral cavity and also in monitoring general health of an organism. Dodds et al studied to determine if there were any difference in parotid saliva output and composition related to caries activity in healthy individuals and concluded that caries activity is related to salivary electrolyte alterations, but not to protein composition.¹⁷ Various trace elements are present in biological substances or fluids in very minute amounts i.e., in microgram per gram or less in concentration. Saliva shows the presence of various types of trace elements and electrolytes.¹⁸

The frequency of dental caries in diabetic patients is correlated to its risk factor potential based on the quality and quantity of saliva¹⁹, which is in turn related to salivary

gland function. Salivary gland hypofunction alters the salivary composition in terms of increased protein, increased or decreased electrolytes, which reflects the salivary flow rate and acinar and ductal function. A hypofunction of salivary gland poses one to increased risk of caries.¹⁹

It has been reported that there is no statistically significant difference between diabetic and non-diabetic patients regarding dental caries prevalence.²⁰ However some studies show increase caries risk in diabetics and yet other show a higher prevalence in diabetics than non-diabetics. This is probably due to lack of metabolic control, increase salivary glucose, crevicular fluid and decrease salivary flow.²⁰

Potassium is a vital electrolyte. Both high and low levels are already associated with various medical conditions, including hypertension, cardiac arrhythmias, osteoporosis and nephrolithiasis in diabetic individuals. An extensive study on this salivary factor in diabetics has not been conducted yet. However, there are fairly strong associations between low serum potassium and increased diabetes risk.²¹

Chloride is the major ion found in the fluid outside of cells and in the blood. In our study an increased level of chloride ion levels have been found in diabetic individuals than in non-diabetic individuals which may be due to hyperchloremia, one of the complications involved in diabetes.²²

Saliva contains large quantities of potassium and bicarbonate ions. The concentrations of both sodium and chloride ions are several times less in saliva than in plasma. Sodium ions are actively reabsorbed from all the salivary ducts and potassium ions are actively secreted in exchange for the sodium. However, there is excess sodium reabsorption over potassium secretion, and this creates electrical negativity of about -70 millivolts in the salivary ducts; this in turn causes chloride ions to be reabsorbed passively. Therefore, the chloride ion concentration in the salivary fluid falls to a very low level, matching the ductal decrease in sodium ion concentration.²³

In the present study it is seen that salivary electrolytes, sodium, potassium and chloride ion concentrations were significantly higher in diabetic patients with dental caries than in non-diabetic individuals with dental caries.

The mechanism behind the increased level of salivary electrolytes in our study is not fully understood and still necessitates extensive research. Some studies report a significant high level of potassium and chloride ions in caries active groups.²⁴ Moreover, it is well established that diabetic patients have salivary gland hypofunction which makes them prone to decreased mucosal integrity and xerostomia, eventually categorized to high caries risk.²⁴

Sodium controls our body's fluid volume and maintains our acid-base balance or pH, nerve conduction, the passage of nutrients into our cells and our blood pressure. The increased level of sodium as a salivary modulator in diabetics has not yet been extensively studied and still

requires more research. However increased levels of sodium in blood of diabetic patients have been studied, owing its probability to alteration to acid-base balance and its consequences.²⁵

Conclusion :

Diabetes mellitus increases one's susceptibility to dental caries. Hence it is of utmost importance that patients are well educated about their condition and associated risks. Our study is a cross sectional population based data, which gives an ensuing relationship between diabetes diagnosis and subsequent risk of dental caries. It is evident from the present study that a positive relationship exists between salivary electrolyte levels and diabetes mellitus and dental caries. However, a longitudinal data might help in better understanding of this association that may prove valuable for clinical practitioners in identifying individuals at high risk of sub-optimal oral health in diabetic population.

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