EVALUATION OF ORAL HEALTH AND SALIVARY STATUS IN JUVENILE DIABETIC CHILDREN"

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Abstract

Background: Diabetes causes a slew of issues for children throughout their life. Children with poor glycaemic control had a greater prevalence of caries and gingivitis, as well as a decrease in salivary flow. So an in vivo study was carried out to correlate oral health, dental caries and salivary microbial status of 5 to 15 years old children with Type I Diabetes Mellitus (T1DM).
Materials and Methods: A total of 60 children: Group A (30 known diabetic children) and Group B (30 healthy children) in age group of 5 to 15 years were included. Oral health was assessed by using Simplified Oral Hygiene Index (OHIS) and dental caries was assessed using DMFT and deft index. The salivary microbial examination was done by evaluating streptococcus mutans (SM) and lactobacillus count (LB).
Results: The mean OHIS value (0.73± 0.35) in well-to-moderately controlled diabetic children was lower than the poorly controlled TIDM (1.21 ± 0.33) children which was statistically significant. Comparison of the caries experience showed that the children with T1DM had higher caries as compared to their matched non-diabetic controls. Salivary parameters showed higher streptococcus mutans in Group AI (2.84 ± 1.13) as compared to Group AII (2.02 ± 1.06) which was statistically significant (p=0.006).
Conclusion: Poorly controlled diabetics had poor oral hygiene, increased caries rate and salivary microbial count of SM and LB as compared to well-to-moderately controlled diabetics.
Keywords: Type 1 Diabetes Mellitus Children, OHIS, DMFT, Streptococcus mutans and Lactobacillus counts.
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Introduction: Diabetes mellitus (DM) is a group of chronic, metabolic diseases characterized by elevated levels of blood glucose. Type 1 diabetes mellitus (T1DM) also known as juvenile diabetes, is the most common type in children and adolescents.[1]T1DM is also on increase like type 2 diabetes with a trend of 3–5% increase/year.[2]The onset of T1DMoccurs predominantly in childhood, with median age of 7–15 years, but may present at any age. Studies have shown high ratio of gingival inflammation and periodontal diseases with subsequent loss of teeth in diabetic patients, and the degree of inflammation and caries incidence depends on the metabolic control.[3]

Dental caries, gingivitis, periodontitis, salivary dysfunction are the major oral infections of DM.[4] However, awareness of these complications is deficient worldwide. Therefore, diabetic oral complications need to be recognized and included in the definitive care of diabetes to fight this disease efficiently.[5]

Following study was designed to investigate the relationship among T1DM, oral health status, and streptococcus mutans (SM) and lactobacillus (LB) conditions in children.

Material and Methods:

Study population: Sample size was calculated to be 60 so as to achieve 80% power, which has a significance level of 0.05.A total of 60 subjects were divided into two groups: Group A- 30 juvenile diabetic children and Group B- 30 non-diabetic children aged between 5 to 15 years. Group A children were selected from the Paediatric Endocrinologist's hospital in Ahmedabad, who have been diagnosed for the T1DM. Further Group A was subdivided into two groups according to their metabolic control. Group AI in which diabetic subjects had HbA1c < 9.0% and Group AII where HbA1c ≥ 9.0%. The level of metabolic control of diabetes mellitus was determined by the glycosylated haemoglobin, HbA1c (analyzator DCA- 2000; Bayer, Diagnostics, Eckhart, USA) reflecting levels of glycemia over the preceding 6–12 week.[6] According to the guidelines for the management of T1DM[6], diabetic subjects with HbA1c <9.0% were considered as well-to moderately controlled patients(Group AI), whereas diabetics with HbA1c ≥ 9.0% were assigned to be poorly controlled diabetes patients(Group AII). Group B or the control group was established by matching each of the diabetic child to non-diabetic child, who did not have any systemic diseases and did not receive medications.

The study protocol was approved by the Ethical Committee of Karnavati School of Dentistry. Prior oral or written consent was obtained from the respective hospital authorities and from the patient's parents/guardians through the hospital to conduct the study.

Children who were diagnosed only with juvenile diabetes and not having or suffering from any systemic disease and those who either themselves or their parents or guardians gave consent were included in the study while those who have juvenile diabetes due to underlying systemic disease or due to drug therapy were excluded from the study.

Clinical examination: Clinical examination was performed by a single investigator assisted by a co-investigator for recording the data throughout the study. The children were made to sit on an ordinary chair and under natural daylight and were examined using the mouth mirror and probe. The oral health status was examined based upon the criteria given by W.H.O in 1997. Visual examination was performed with dental mirror and probe.

Oral health was assessed by using Simplified Oral Hygiene Index (OHIS)[7] and dental caries was assessed using DMFT[8] (decayed, missing and filled permanent teeth) and deft[8] (decayed, extracted and filled deciduous teeth) index. The salivary microbial examination was done by

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evaluating streptococcus mutans and lactobacillus count.

For microbial examination, 1ml of unstimulated whole saliva was collected in a sterile saliva collection tube from each of the subject included in the study. It was made sure that patient did not eat or drink anything (except water) at least 2 hours prior to the saliva collection. All the samples were stored in ice box until processed further. Samples were processed within 2 hours of collection.

Preparation of the samples for microbial examination: Samples were shaken well for at least 30 seconds and then were diluted 10 folds with distilled water. Then 5 microliter of the diluted sample was spread over each of the media with a pre-calibrated loop.

For streptococcus mutans, 5 microliter of diluted sample was spread over TYCSB[9] media (Hi-Media) and then it was incubated at 37 degrees Celsius for 48hours.

For lactobacillus, 5 microliters of the undiluted sample were incubated on Rogosaagar[10] (de Man, Rogosa and Sharpe) medium containing sodium acetate for 48 – 72 hours at 35 degree Celsius.

Later colonies growth on both the culture media was identified as specific microbial species by their colonial morphology under stereomicroscope and by various biochemical tests to identify different Streptococcal species and lactobacillus species. Along with that, no of colony forming units (CFU) per ml of saliva was counted with aid of manual colony counter.

List of Biochemical Tests[11,13,15]

- **For Streptococci mutans:**

Biochemical Test	**Streptococcus Mutans**
Catalase	+
Voges - Proskauer (VP)	+
Suger Fermentation Mannitol Sorbotol	 + +
Esculine Hydrolysis	+

(+ = Positive result)

- **For Lactobacillus Spp.:**

- Gram staining Test (L. acidophilus)
- Catalase

Statistical Analysis: All statistical analysis was performed using the IBM SPSS statistics 20.0 (IBM Corporation, Armonk, NY, USA) software version. Microsoft word and Excel were used to generate graphs, tables etc**.** Level of significance was fixed at p=0.05 and any value less than or equal to 0.05 was considered to be statistically significant. Chi square analysis was used to find the significance of study parameters on categorical scale. Student t tests (two tailed, paired & unpaired) were used to find the significance of study parameters on continuous scale between two groups. Mann–Whitney U test was used to find the significance of study parameters between the groups (Inter group analysis).

Results: The variable parameters included in this study were OHIS, DMFT/deft and streptococcus mutans count and lactobacillus count in unstimulated saliva. The distribution of the children according to their age, gender and medical conditions was done as shown in table 1.

The comparison of the oral health status between the two groups as shown in table 2 depicted that the children with T1DM had higher OHIS and DMFT and deft values than their matched non-diabetic controls but there was statistically insignificant difference in mean OHIS value (p= 0.072) and mean DMFT and deft value (p= 0.105) between the two groups.

Salivary parameters in diabetics and their non-diabetic matched pairs (table 3) showed statistically significant difference

in the mean values of SM (p=0.006) and LB (p=0.004) count which was higher in diabetics as compared to non diabetics.

According to the level of metabolic control of type 1 diabetes mellitus as shown in table 4, the difference among the mean value of OHIS (p= 0.001) and DMFT and deft (p=0.006) was found highly significant. The well-to-moderately controlled diabetic children were found to have a statistically significant lower mean OHIS and DMFT and deft values as compared to the poorly controlled type 1 diabetes mellitus children.

When salivary parameters in T1DM children were compared according to the level of metabolic control as shown in table 5, there was statistically significant difference in the mean SM value (p=0.001) and LB value (p=0.020) between Group AI and Group AII with higher SM and LB counts in Group AII.

Table 1: Distribution of children according to demographics		
	Group A – Diabetics	**Group B – Non – Diabetics**
Age	0-15 years	0-15 years
Gender (M:F)	14:16	14:16
Medical Comorbidities	Type 1 Diabetes	Healthy
Glycemic Control	Poor	Good

Table 2: Mean (SD) and mean paired differences (SD) of the total OHIS and DMFT /deft in the diabetics and their non-diabetic matched pairs							
	GROUP	**N**	**Mean**	**Std. Deviation**	**Std. Error Mean**	**Mean Difference**	**P VALUE**
OHIS	diabetes	30	1.03	0.403	0.074	0.240	0.072
	non diabetes	30	0.79	0.594	0.109		
DMFT and deft	diabetes	30	2.20	2.340	0.427	0.867	0.105
	non diabetes	30	1.33	1.688	0.308		

Table 3: Salivary parameters in diabetics and their non-diabetic matched pairs							
	GROUP	**N**	**Mean**	**Std. Deviation**	**Std. Error Mean**	**Mean Difference**	**P value**
SM	diabetes	30	2.84	1.133	0.207	0.82	0.006
	non diabetes	30	2.02	1.061	0.194		
LB	diabetes	30	3.07	1.617	0.295	1.20	0.004
	non diabetes	30	1.87	1.474	0.269		

Table 4 : OHIS and DMFT/deft of the diabetic children according to the level of metabolic control of type 1 diabetes mellitus (T1DM)

	HbA1c_group	N	Mean	Std. Deviation	Std. Error Mean	Mean Difference	P VALUE
OHIS	Group AI (HbA1c <9.0)	11	0.73	0.347	0.105	-0.48	0.001
	Group AII (HbA1c ≥ 9.0)	19	1.21	0.326	0.075		
DMFT and deft	Group AI (HbA1c <9.0)	11	0.73	1.348	0.407	-2.33	0.006
	Group AII (HbA1c ≥ 9.0)	19	3.05	2.392	0.549		

Table 5 : Salivary parameters of the diabetic children according to the level of metabolic control of type 1 diabetes mellitus (T1DM)

	HbA1c_group	N	Mean	Std. Deviation	Std. Error Mean	Mean Difference	P VALUE
SM	Group AI (HbA1c <9.0)	11	1.99	0.768	0.231	-1.34	0.001
	Group AII (HbA1c ≥ 9.0)	19	3.33	1.022	0.234		
LB	Group AI (HbA1c <9.0)	11	2.18	1.736	0.523	-1.40	0.020
	Group AII (HbA1c ≥ 9.0)	19	3.58	1.336	0.307		

Discussion: Diabetes mellitus is the term used for a group of metabolic disorders that are clinically and genetically heterogeneous, but share the common characteristic of glucose intolerance.[12] The American Association of Diabetes in 1997 suggested a classification system for diabetes. It is currently classified as type 1 or juvenile diabetes (formerly, insulin-dependent diabetes) and type 2 or acquired (formerly, noninsulin-dependent diabetes).[13] Although the definitive aetiology of diabetes mellitus remains unclear at the present time, certainly with regard to chronic Type I diabetes, experimental findings are in accordance with the classical view that the metabolic disorder in such patients is due to an absolute deficiency of insulin, resulting from a severe deficiency of the insular tissue in the pancreas.[14]

The systemic complications of T1DM include microvascular disease of the eye (retinopathy) and kidney (nephropathy), neuropathies and cardiovascular disease. Neutrophil adherence, chemotaxis, phagocytosis and bactericidal activity, and cell-mediated immunity are all compromised in the hyperglycemic diabetic.[13]

Oral manifestations of T1DM includes higher prevalence of dental diseases like dental caries, xerostomia, gingival inflammation are reported for children with Type I diabetes when compared to systemically healthy children.[15] Orbak R[6] [2008] found that the dental development of diabetic group was fast until the age of

10 and then dental development decreased. Edentulous interval was longer in diabetic children.

Type I diabetic paediatric patients not suffering from any other systemic disease were included in this study because the influence of any other systemic disease as well as the drugs taken for the same could modify the salivary composition. For evaluation of oral health OHI-S and DMFT/deft index was used as OHI-S is simple, sensitive and less time consuming.[7] Lopez ME[8], Miralles L[9] , Rai K[11] have evaluated oral health in Type 1 diabetic children. They concluded that DMFT/deft is simple, rapid, versatile and universally accepted Index. Wolf J[10], Tenovo J[16], Lopez ME[8], Siudikiene J[17,18] and Rai K[11] have also compared caries prevalence in T1DM and healthy children as this study.

The data from numerous surveys of various tooth surfaces, of different patient age groups from numerous countries, and populations with different dietary habits, etc., have shown a strong positive association between increased level of mutans streptococci and initiation of demineralization. As Streptococcus mutans are responsible to initiate caries whereas, Lactobaccilus are responsible for the caries progression[19], SM and LB count was selected as salivary parameter in this study. Karjalainen[20], Edblad[21] and Siudikiene J[17,18] also observed Streptococcus mutans and lactobacillus count in children with insulin dependent diabetes mellitus.

In the present study, insignificant difference in oral health status of diabetic children and non diabetic children. Our results is similar to the studies done by Lopez ME[8], Miralles L[9]. Whereas Rai K[11] showed positive correlation of oral hygiene with T1DM and healthy children.

Clinical results of the present study indicate insignificant difference of dental caries in patients with type 1 diabetes compared with non-diabetic individuals. This result is similar with the previously performed studies by Wolf J[10], Lalla E[22]. In contrast Lopez ME[8], Siudikiene J[17,18] and Rai K[11] showed significant difference of dental caries in type 1 diabetes subjects compared with non-diabetics which suggest caries incidence has been attributed partially to decreased salivary flow and increased levels of carbohydrates in the parotid saliva.[23] A close relationship between hyperglycemia and an increased rate of dental caries in rats has been established.[24] Wegener[25] in a study of 700 diabetic children, noted that immediately after onset of the disease, these children evidenced a higher caries incidence than controls. Mattson. L. and Koeh. G.[26] have even noted a lower caries frequency in diabetic children under treatment than in age-matched healthy controls. Dietary restriction, particularly the omission of sucrose, has been suggested as the explanation for this finding. Dubey S. et. al.[27] 2018 also inferred increased prevalence of dental caries in diabetic patients and attributed this due to the leakage of glucose from blood into the oral cavity.

This study shows significant difference in SM and LB count in T1DM and non diabetic patients. According to Tenovuo J[16] and Twetman[28] SM counts were higher in T1DM and in contrast Iughetti[29] and Siudikiene J[18] found no difference in LB count among T1DM and healthy controls. Whereas Collin HL[30] found lower LB count in juvenile diabetics, which are contradictory to our results.

Decreased salivary flow in poorly controlled diabetics may have resulted in less effective cleansing and a decreased supply of antibacterial substance. Harrison [31] in their study concluded that combination of decreased salivary flow, increased salivary glucose, and poor oral hygiene contribute to an increased accumulation of plaque in poorly controlled diabetics. Diabetic patients with reduced salivary flow rate had mean DMFT to be significantly lower. This could be because in diabetic patients, the good metabolic control prevented the most dangerous salivary changes such as high glucose content and lower pH and a good diabetic

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diet, rich in fiber, and low in simple carbohydrates could slow down the production of plaque and the proliferation of acidogenic bacterial microflora.[[28] By contrast, Edblad E[21] were not able to identify any significant differences in caries experience of diabetics in relation to their metabolic control.

A high HbA1c level reflecting hyperglycemic periods is deleterious for oral health. Tenovuo[16] reported high salivary glucose concentrations concomitantly with high blood glucose concentrations. Twetman S[28] showed that the cariogenicity of SM and LB is especially prominent among the diabetic population with poor glycemic control. Syrjala AMH[32] found that among subjects with poor metabolic control have higher levels of mutans streptococci and lactobacilli when compared to those with better metabolic control. In acidic environment cariogenic bacteria are likely to be thrived. Increased blood glucose levels, reduced salivary flow rate, reduced buffering capacity, poor dietary control are the other risk factors which increases the risk of dental carries in Diabetes Mellitus patients.[33] So it is necessary for the public health authorities and health professionals to provide awareness of oral health for special children.[34]

In the present study, only dental caries and oral hygiene related to debris and calculus accumulation were taken into consideration. So in the future study the occurrence of other oral lesions such as aphthous stomatitis, angular cheilitis, candidiasis and lichen planus and developmental defects of teeth and their relation with the metabolic control should be taken into consideration.

Conclusion: Oral health status and caries prevalence in Diabetic children showed only mild difference then non-diabetic children. Salivary microbial evaluation revealed increased SM and LB count in Diabetic Children. Poorly controlled diabetics had a poor oral hygiene, increased caries rate and salivary microbial count of SM and LB. Thus it can be concluded that metabolic control in type 1 diabetes mellitus plays a major role on overall child's oral health.

Conflict of interest: We declare that there is no conflict of interest between the authors.

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