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RESEARCH ARTICLE

CARRIAGE RATE OF STREPTOCOCCUS MUTANS AMONG TYPE I & TYPE II DIABETIC PATIENTS WITH DENTAL CARIES AND COMPARED WITH NON – DIABETIC POPULATION – A CROSS SECTIONAL STUDY

***,[1]Umadevi, [1]Dr. Santha Devy, [2]Dr. Udhaya Visvanathan, [1]Dr. Vezhavendhan, [1]Dr. K. R. Prem Lal and [1]Dr. S. Vidyalakshmi**

[1]Department of Oral Pathology and Microbiology, Indira Gandhi Institute of Dental Sciences, Puducherry, India
[2]Department of Microbiology, Rajah Muthiah Medical College, Chithambaram, India

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ABSTRACT

Background: Oral cavity reflects the general health status of an individual and is colonized by more than seven hundred species living in its own domain and balances the normal oral ecology. This ecosystem tends to g*et al*tered when there is a local or systemic disease. One among the systemic diseases, Diabetes Mellitus a common life style disease has an influence on the microbiota which in turn tends to shift the ecosystem. Thus the aim & objective of the study is to identify the Carriage rate of Streptococcus mutans from five anatomical surfaces of oral cavity of Type I and Type II Diabetes Mellitus and Non Diabetic population & to compare carriage rate Streptococcus mutans of Type I and Type II Diabetic patient with Non Diabetic population.

Materials and Methods: Swabs were collected from five different anatomical sites of the oral cavity from Type I & Type II and Non Diabetic patients of 30 each. Samples were confirmed for the species and then inoculated in Mitis Salivaris Agar to assess the Frequency of isolation and Colony Forming Units (CFU) of Streptococcus mutans.

Results: Observation of the study revealed that the Frequency of Isolation and Colony Forming Units were high in patients with Type II Diabetes Mellitus when compared to that of Type I Diabetes Mellitus and the Non Diabetes Mellitus subjects.

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INTRODUCTION

Diabetes Mellitus a major public health concern is considered to be the most common endocrine disorder, where the metabolism of carbohydrate, fat and protein are deranged caused by the autoimmune destruction of the beta cells of the Islets of Langerhans of the pancreas, and other multifactorial factor that leads to a total lack of insulin which further progress to acute and chronic complications to set the disease process. Diabetes Mellitus affects approximately 2% of the population and traditionally it has been classified as Type 1 and Type II (Greenberg *et al.,* 2008; Samuel, 2006) where classically Type-1 is referred to Insulin Dependent Diabetes Mellitus and Type II as Non-Insulin Dependent Diabetes Mellitus with the archetypal signs and symptoms that includes the classic triad of frequent urination (polyuria), increased thirst (polydipsia), and increased hunger (polyphagia) and is characterized by hyperglycemia due to insulin deficiency, a resistance to its peripheral action or both. Diabetes Mellitus adversely affects almost all organs and tissues of the body including the oral cavity and it is the ability of oral infections that profoundly affects the metabolic control of the Diabetic state. The oral cavity is sterile at birth and the colonization of first microorganism the *Streptococcus salivarius*, *Streptococcus mutans* etc. starts shortly after birth and these organisms are referred to as pioneer species. Gradually the oral cavity is predominated by the other aerobic and anaerobic microorganisms that colonize on the oral surfaces where they form a consortium referred to as Oral biofilm. As such over 700 microbial species are found in the typical adolescent human mouth and are collectively referred to as Oral Microflora, Oral Microbiota, or Oral Microbiome. These microorganism harbour in several distinct microbial habitats such in its own ecological niche, on the varied surfaces found within the human mouth, such as on the distinct non shedding surface of the teeth and to an extent on the mucosal surface. These microbial species are the commensals considered to be

****Corresponding author: Umadevi***
Department of Oral pathology and Microbiology, Indira Gandhi Institute of Dental Sciences, Puducherry, India.

the resident flora which associate with each other and along with the host factors thereby maintaining the homeostasis within the ecosystem. According to the ecological plaque hypothesis certain factors exist, may be the systemic or local that triggers the shift in the proportion of the resident microbiota to a more or less pathogenic state and predisposes to disease process. One among the systemic cause is Diabetes Mellitus, which may influence certain changes affecting the oral micro biota especially pertaining to *Streptococcus mutans* that invariably alters and affects the soft and hard tissue of the oral cavity. The hard tissue manifestations include variations in tooth development, periodontal manifestations and prevalence of dental caries. Whereas the soft tissue attributes towards an increased prevalence of oral mucosal lesions, infections, burning mouth syndrome, taste disorders and other oral changes are the result of salivary gland abnormalities. Though there are different types of oral microbiota the study of and their load in the oral cavity of the Type I Diabetes Mellitus needs more emphasis as there is only very few literatures available. Therefore the present study is intended to search the inter link for the role of *Streptococcus mutans* in Type I and Type II Diabetes Mellitus and thereby the study is aimed at the Frequency of isolation and the Colony forming units of *Streptococcus mutans* in various surface of the oral cavity and to correlate out the probable reason for the ecological change in Type I and Type II Diabetic patients.

Aims and Objectives

- To study the Carriage rate of *Streptococcus mutans* from the different anatomical site of the oral cavity of Diabetes Mellitus and Non-Diabetes Mellitus population.
- To compare Carriage rate of *Streptococcus mutans* of Type I and Type II Diabetes Mellitus with Non-Diabetes Mellitus population.

MATERIALS AND METHODS

A cross sectional study was conducted from January 2014-May 2015 .The study protocol has been approved by the Institutional Review and Institutional Ethics Board of Indira Gandhi Institute of Dental sciences, Sri Balaji Vidyapeeth (SBV). A total of 90 subjects participated in the study. The study population was divided into three categories as follows:

Group I: Type I Diabetes mellitus patients with Dental caries (30samples)

Group II: Type II Diabetes mellitus patients with Dental caries (30 samples)

Group III: Non-diabetes mellitus patients with Dental caries. (30 samples)

The subjects selected which met the following criteria:

Inclusion criteria

Clinically proven Diabetes mellitus patients of Type I and Type II disease with Dental caries formed the test groups. Age and sex matched Non - Diabetes mellitus patients with Dental Caries were included as controls to compare the results with test groups.

Exclusion criteria

The following individuals / patients were excluded in our study.

- Normal healthy individuals without Dental Caries.
- Normal healthy individuals with other oral complaints.
- Diabetic patients with other oral/ Dental complaints except Dental Caries (Type II)
- Diabetic patients with other oral/ Dental complaints except Dental Caries (Type I)

Swabs were separately collected from five anatomical sites such as Tongue dorsal surface, Tongue ventral surface, Buccal mucosa, Gingiva and Caries tooth surface of the Oral cavity. Initial confirmation for the presence of Gram positive cocci was done by Gram stain and viewed under microscope as shown [Figure 1]. The collected oral samples were serially diluted and 50 microliters of 1/1000 diluted samples (each) were subjected to anaerobic culture on Mitis Salivarius Agar (MSA) and incubated at 37 degree centigrade for 48 hours. Bacterial Colony forming units (CFU) was assessed by counting the typical *Streptococcus mutans* colonies and recorded for further statistical analysis. The colonies are recognized by their colony morphology based on Raised, convex, undulate, opaque, pale-blue colonies that are granular (i.e., "frosted glass") in appearance. [Figure 2] Colonies may exhibit a glistening bubble on the surface due to excessive synthesis of glucan from sucrose. Colonies identified as *Streptococcus mutans* are further confirmed of their genus by Catalase negativity and by their hemolytic pattern (α and γ). [Figure 3, 4].

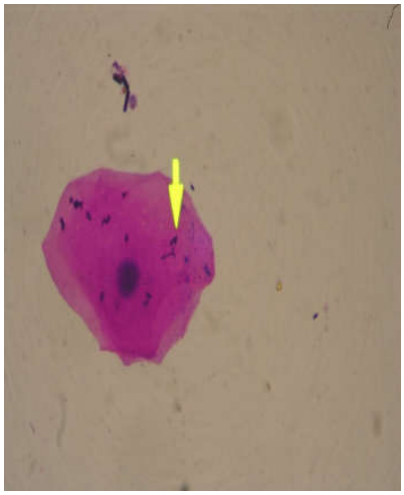

Figure 1.

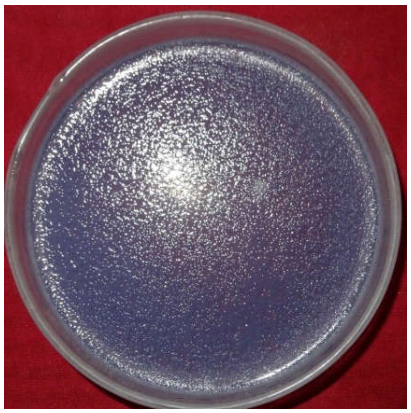

Figure 2.

Table 1 [a-e] The number of samples that showed isolation of *Streptococcus mutans* in a group from each surface that is investigated & their level of significance between groups

		Tongue Dorsal		
a		Negative	Positive	
GROUP	I	29	1	chi - 15.659
	II	18	12	p <0.001
	III	27	3	
b		Tongue Ventral		
		Negative	Positive	
GROUP	I	30	0	Chi - 16.410
	II	19	11	p <0.001
	III	27	3	
c		Buccal Mucosa		
		Negative	Positive	
GROUP	I	27	3	Chi - 28.151
	II	12	18	p <0.001
	III	28	2	
d		Gingiva		
		Negative	Positive	
GROUP	I	21	9	Chi - 20.633
	II	7	23	p<0.001
	III	23	7	
e		Carious Tooth		
		Positive		No comparison
GROUP	I	30		
	II	30		
	III	30		

G1 – Group I, G2 – Group II, G3 – Group III plotted on the X – axis

Table 2. The Mean and Standard Deviations of colony forming units of *Streptococcus mutans* isolated from the three subsets of patients in various surfaces

	Number	*Dorsal Tongue*		*Ventral Tongue*		*Buccal Mucosa*		*Gingiva*		*Carious tooth*	
		Mean	*SD*	*Mean*	*SD*	*Mean*	*SD*	*Mean*	*SD*	*Mean*	*SD*
GROUP I	30	8.33	2.339	3.13	1.525	9.13	2.177	53.43	12.558	181.23	44.829
GROUP II	30	52.90	5.486	11.10	4.894	30.50	5.231	178.87	45.580	396.00	76.283
GROUP III	30	.83	.913	2.67	1.826	1.53	.973	22.67	8.438	37.70	15.038
Total	90	20.69	23.366	5.63	4.980	13.72	12.761	84.99	73.261	204.98	156.673

Table 3 (a , b). The significance level of the Colony forming units analysed for the study groups from the Dorsal surface of tongue

Table 3a.

Site taken	Comparison groups	Sum of squares	Degree of freedom	Mean square	F value	Significance p value
Dorsal surface of tongue	Between Groups	47533.756	2	23766.878	1958.932	*.000*
	Within Groups	1055.533	87	12.133		
	Total	48589.289	89			

Table 3b.

Group	Group	Mean difference	Std. error	Significance
I	II	-44.567*	.899	*.000*
	III	7.500*	.899	*.000*
II	I	44.567*	.899	*.000*
	III	52.067*	.899	*.000*
III	I	-7.500*	.899	*.000*
	II	-52.067*	.899	*.000*

Table 4(a , b). The significance level of the Colony forming units analysed for the study groups from the Ventral surface of tongue

Table 4a.

Site taken	Comparison groups	Sum of squares	Degree of freedom	Mean square	F value	Significance p value
Ventral surface of tongue	Between Groups	1348.067	2	674.033	68.280	.000
	Within Groups	858.833	87	9.872		
	Total	2206.900	89			

Table 4b.

Group	Group	Mean difference	Std. error	Significance
I	II	-7.967*	.811	.000
	III	.467	.811	.834
II	I	7.967*	.811	.000
	III	8.433*	.811	.000
III	I	-.467	.811	.834
	III	-8.433*	.811	.000

Table 5 (a , b). The significance level of the Colony forming units analyzed for the study groups from the Buccal Mucosa

Table 5a.

Site taken	Comparison groups	Sum of squares	Degree of freedom	Mean square	F value	Significance p value
Buccal Mucosa	Between Groups	13533.622	2	6766.811	614.245	.000
	Within Groups	958.433	87	11.016		
	Total	14492.056	89			

Table 5b.

Group	Group	Mean difference	Std. error	Significance
I	II	-21.367*	.857	.000
	III	7.600*	.857	.000
II	I	21.367*	.857	.000
	III	28.967*	.857	.000
III	I	-7.600*	.857	.000
	II	-28.967*	.857	.000

Table 6(a , b). The significance level of the Colony forming units analyzed for the study groups from the Gingiva

Table 6a.

Site taken	Comparison groups	Sum of squares	Degree of freedom	Mean square	F value	Significance p value
Gingiva	Between Groups	410785.5	2	205392.74	267.153	.000
	Within Groups	66887.500	87	768.822		
	Total	477673	89			

Table 6b.

Group	Group	Mean difference	Std. error	Significance
I	II	-125.433*	7.159	.000
	III	30.767*	7.159	.000
II	I	125.433*	7.159	.000
	III	156.200*	7.159	.000
III	I	-30.767*	7.159	.000
	II	-156.200*	7.159	.000

Table 7(a, b). The significance level of the Colony forming units analyzed for the study groups from the Carious tooth surface

Table 7a.

Site taken	Comparison groups	Sum of squares	Degree of freedom	Mean square	F value	Significance p value
Carious tooth	Between Groups	1951054	2	975527.14	363.33001	.000
	Within Groups	233591.7	87	2684.9617		
	Total	2184646	89			

Table 7b.

Group	Group	Mean difference	Std. error	Significance
I	II	-214.767*	13.379	.000
	III	143.533*	13.379	.000
II	I	214.767*	13.379	.000
	III	358.300*	13.379	.000
III	I	-143.533*	13.379	.000
	II	-358.300*	13.379	.000

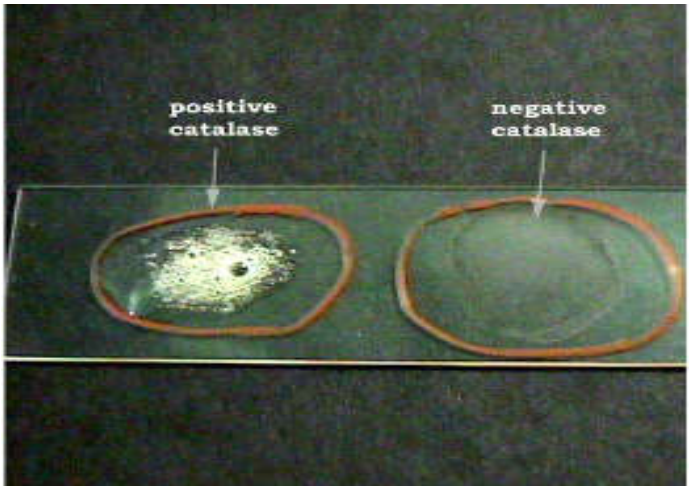

Figure 3.

Figure 4.

The observational results from Table 1 and Table 7 exhibited, The Frequency of isolation of *Streptococcus mutans* was more in Type I (43/150) & Non diabetic (39/150). The Frequency of isolation was significantly different between three groups from the Dorsal surface of Tongue ($p < 0.001$), Ventral surface of Tongue ($p < 0.001$), Buccal mucosa ($p < 0.001$) and Gingiva ($p < 0.001$).The Colony forming units of *Streptococcus mutans* was more in Type I (Group I) and Non diabetic (Group III) in each of the five surfaces investigated following similar pattern as that of Frequency of isolation. The Colony forming units was significantly differently between two given study groups in each of the surfaces investigated except the Ventral surface of Tongue between TypeI (Group I) & Non diabetic (Group II) samples. ($p = 0.834$).The Frequency of isolation & Colony forming units of *Streptococcus mutans* was highest in carious tooth followed by Gingiva in two study groups. The *Streptococcus mutans* was isolated in all the samples from Caries tooth surface in each of the study group as expected. The Frequency of isolation and Colony forming units of *Streptococcus mutans* was the least from Ventral surface of tongue in the Type I diabetic group patients. (Group I). The Frequency of isolation & Colony forming units of *Streptococcus mutans* was the least from Buccal mucosa & Dorsal surface of Tongue respectively in Non diabetic (Group II). *Streptococcus mutans* have been isolated on the surfaces where they are not generally isolated with more number colony yield. We know that if caries is not the factor, Diabetes Mellitus itself should be the influencing cause. The reason for this could be due to the difference in pathophysiology of disease in itself and in the way patient responds to insulin.

DISCUSSION

Microorganism's constantly is present on body surfaces and body cavities such as gut, nasopharynx and oral cavity .Within this oral cavity like any other body cavities the microbial community living in a defined habitat ,stabilizes the ecosystem by the concept of homeostasis. In oral cavity the ecosystem is stabilized by the resident flora, such as *Mutans Streptococcus* and *Streptococcus sanguis* are mostly seen on the teeth surfaces whereas *Streptococcus salivarius*, *Actinomycosis* and *Porphyromonas* habitat in the tongue , supra gingival and sub gingival areas respectively. In the mucosal surface the colonization of microorganism are relatively less and it dominated by the *Streptococcus salivaris and Streptococcus mitis* (Allcock *et al.,* 2001). The first parameter from the results of the study were looked in for the Frequency of Isolation of *Streptococcus mutants* from the specified surface of the oral cavity. On comparing the three groups it is found that the Frequency of isolation of *Streptococcus mutans* was the highest in Group II samples from all the five surfaces [Table number: 1, a-e] ie. one hundred and twelve patients out of one hundred and fifty samples showed frequency of isolation for *Streptococcal mutants*(112/150), followed by Group I where only forty three out of one hundred and fifty (43/150)& Group III which showed only thirty nine out of one hundred and fifty samples (39/150). Chi square test was used for the analysis of the data between the three groups and five surfaces, and found that the Frequency of isolation was significantly different between three groups from the Dorsal surface of Tongue ($p < 0.001$), Ventral surface ($p < 0.001$), Buccal mucosa ($p < 0.001$) & Gingiva ($p < 0.001$), [Table number1a-1d]. Where in teeth surface the frequency of isolation was noticed in all the groups ie. out of ninety frequency of isolation was noticed in all ninety samples.

Related to the Colony Forming Units in our study the presence of *Streptococcus mutans* was evaluated in Group I, Group II and Group III respectively and inferred that the mean and standard deviation for colony forming units of *Streptococcus mutans* was the highest in Group II samples followed by Group I and Group III in each of the five surfaces investigated as in [Table 2]. The Colony Forming Unit in dorsal surface of the Tongue, Buccal mucosa, Gingiva and in carious tooth surface showed statistically significant results with the value $p< 0.001$) between the three groups. [Table number 3: a, b], [Table 5: a, b] [Table 6:a,b] [Table 7:a ,b] respectively. Except the ventral surface of the tongue between Group I & Group III [Table 4 a, b] there was no significance as the value is ($p= 0.834$.) Frequency Of Isolation and Colony Forming Units were comparatively more in Group II (Type II Diabetic Mellitus) and the results were concordant with study conducted by Sharma *et al.,* 1989 where there was Colony Forming Units for streptococcus was comparatively more in the Diabetic group than the nondiabetic group. It is a known fact that one of the factor for the progression of dental caries is *Streptococcus mutans* and as in our study all the groups had caries we wanted to insist that the role of diabetes as the cause for the streptococcal count rather than the Dental caries. Therefore the probable reason to substantiate this is that in diabetes there is hypofunction of the salivary gland which leads to low salivary flow, reduction in pH of saliva and alteration in its composition (Piattelli *et al.,* 1989) or may be the reduction in the circulating insulin level in diabetes which leads to compensatory hyperplasia of the salivary glands thereby altering the salivary flow (Terry, 1994) this reason is also supported with the findings in literature (Kuo *et al.,* 2008) stating that poorly controlled diseases has been associated with lower stimulated parotid flow. Other reason could be also that changes in the hormonal, microvascular and neuronal in poorly controlled diabetes may lead to salivary gland hypofunction that alters the micro environment favourable for the growth of the organism (Murrah, 1985). Whereas in Group I there was comparatively less Frequency of isolation and CFU than Group II in a study conducted showed that there is lack of correlation between Type I diabetic and the *Streptococcal mutans* (Gamboa *et al.,* 2008), one more study was concordant with our results where salivary samples showed only 6% of *Streptococcus mutans* (Scully *et al.,* 2010), on contradictory a quantitative study of *Streptococcal mutans* in saliva of Type I diabetes and healthy childrens found that no difference in the number of *Streptococcus* mutans (Sakeenabi *et al.,* 2011). From this we infer that the probable reason is in type I diabetes the cause could be that auto immunity state could disrupt the normal commensals due to poor glycol metabolic control which is also supported by an author (Davie, 2003).

In group III the mean and standard deviation was almost less [Table 2] when compared to Group I and II the probable reason could be that the factor normal salivary flow makes the environment selfcleansing or the immunoglobulin's present in saliva and gingival crevicular fluid neutralizes the bacterial toxins by inhibiting the bacterial adhesions (Salonel *et al.,* 1990). Considering the fact that fundamentals of Diabetes Mellitus and Dental caries remained common amongst our both diabetic groups, a higher yield in Type II Diabetes Mellitus would not be possibly substantiated by many reason that we know of. This made us to probe into the pathophysiology of Diabetes mellitus in itself so as to retrieve an explanation. Type II diabetes mellitus being a chronic low grade inflammatory metabolic disorder (Dandona *et al.,* 2004)

are resultant of altered gut microbiota, leading to endoplasmic reticulum stress (Maximilian Zeyda Thomas, 2009) and altered metabolism of glucose in the target organs (Maximilian Zeyda Thomas, 2009) and in the metabolism of short chain fatty acids in the gut (Davie , 2003; Gao *et al.,* 2009). This process leads to impairment and the release of mediators and endotoxins (Wellen *et al.,* 2005). In our study we considered that the altered microbiota could be a phenomenon that affects the whole of alimentary canal including oral cavity leading to yields which are unexpectedly high and different in Type II Diabetes Mellitus. Such a change could also be a reflection of endotoxins and mediators possibly due to their entry into systemic circulation. The Frequency of Isolation & Colony Forming Units of *Streptococcus mutans* was highest in carious tooth in type II diabetics [Table Number (1- e) and Table 7-(a& b)] respectively, followed by Gingiva in type II diabetes [Table Number 1 (d) and Table 6(a, b)] respectively. In carious teeth it is evident that the fact is the *Streptococcus mutans* will be definitely present because the food impaction or debilitated oral health will promote the resident flora to create an acidic environment, enhances fermentation, reduces the local pH rapid production of lactic acid from the dietary sugars aids in adhesion on the tooth surface causing dental caries. But as the samples in our study are related to diabetic we need to substantiate the role of diabetes and streptococcus and not the streptococcus and dental caries. Therefore it is the factor is that increased glucose level in saliva and gingival crevicular fluids, alters the plaque micro flora, and reduced salivary flow associated with the poor metabolic control could be the cause (Terry, 1994; Kuo *et al.,* 1998; Ryan *et al.,* 1999; Tagelsir *et al.,* 2011). One more reason could be that the tooth surface is a non-shedding hard surface which selectively adsorbs various acidic glycoproteins such as mucins from the saliva which forms the acquired enamel pellicle which contains a high number of sulphate and carboxyl groups which directly increases the net negative charge of the tooth surface and its virulence factors that includes adhesins, glucan producing and binding exoenzymes proteases and cytokines stimulating molecules that helps in attachment (Marcotte and Lavoie, 1998; Zaura *et al.,* 1999).

In diabetic the persistent pH drop after exposure to fermentable dietary carbohydrates can be due to the metabolic activity of increased numbers of bacteria on the tooth surfaces which leads to demineralization of the tooth which progress to cavitation of tooth. One more factor is the saliva which is the principal defence factor of the mouth is important for maintaining good oral health which is compromised by the prevalence of hypo salivation and xerostomia in Type I and type II diabetes that are predisposed to dental caries and growth of *streptococcus mutans* (Terry, 1994; Kuo *et al.,* 1998; Murrah, 1985; Rao *et al.,* 2010). In contradictory there is a study stating that there is no effect of diabetes on the prevalence of caries where the caries protective effect of saliva was partially lost in the patient with Non –Insulin dependent diabetes mellitus. Apart from this one more study states that in early gingivitis an ecological niches may be created for the growth of *Streptococcus mutans* along with other higher counts of cariogenic species and also may be due to the lower oxygen tension within the periodontal pockets that favours the growth of micro aerophilic species such as *Streptococcus mutans* (Canepari *et al.,* 1919). One more study states that periodontal infections caused by chronic gram negative bacteria may lead to poor glycaemic control and greater insulin resistance and it is also postulated that periodontitis induced bacteraemia causes rise in serum pro inflammatory cytokines resulting in hyperlipidaemia that finally leads to an insulin resistance syndrome contributing to destruction of pancreatic beta cells (Leach *et al.,* 1968; Kuo *et al.,* 1998). One more study stated that there is numerous contributing factors that are responsible for increased susceptibility of diabetics to periodontal diseases such as compromised polymorph nuclear leukocyte function resulting from impaired neutrophil adherence, chemotaxis and phagocytosis prevent destruction of bacteria in the periodontal pocket and also markedly enhance the periodontal destruction (Taylor, 2003).

In our study The *Streptococcus mutans* was isolated in all the samples from Caries tooth surface [Table 1e] Though S. mutants are not good colonizers of the tooth surface there could be mechanism other than high affinity for receptors on the acquired enamel pellicle, like that they have the ability to synthesize adherent glucans from sucrose and the micro environment created by the topography of the tooth surface. In our study as the samples are diabetic the added reason for all the subjects to have caries could be due to the factor related to low salivary flow or altered saliva. The next parameter observed is The Frequency of isolation and Colony forming units of *Streptococcus mutans* was the least from Ventral surface of tongue in the diabetic group patients. (Group I & Group II). Out of thirty samples , in a group I there was zero positive for frequency of isolation and eleven patients was positive for group II [Table 1b] and three positive in Group III and statistically the p value ($p < 0.001$) which was significant , and pertaining to the colony forming units in the ventral surface the Type I and type II diabetes was least with the p value of .834 which was not significant [Table 3(a, b)] the reason could be one is that it is a self - cleansing area with good salivary flow in the floor of the mouth compared to other areas and on more factor is that it is a shedding surface which prevents the colonization of the bacteria. This observation may hold true as the CFU for *Streptococcal mutans* in the floor of the mouth and ventral surface of the tongue in normal individuals are also less (Allcock *et al.,* 2001). Whereas the Frequency of isolation & Colony forming units of *Streptococcus mutans* was the least from Buccal mucosa & Dorsal surface of Tongue respectively in Group III. [Table1c & Table 1a] & [Table 5 (a, b)] & [Table 3 (a, b)]. The oral cavity does have different types of surfaces such as reflective mucosa, specialized mucosa and masticatory mucosa which contribute to diversity of microflora and that the microbial load is relatively low on these mucosal surface of oral cavity which may be due to continuous programmed desquamation process and moreover these areas are highly bathed with saliva which has a self-cleansing properties and the immunoglobulins which are the components of saliva that are responsible for anti- bacterial activity which prevents the colonization of the microorganisms . These could be the reason for the low frequency of isolation and colony forming units of *Streptococcus mutans* in this region .One more point needs to be substantiated for buccal mucosa that in normal individuals the quantity of microorganism is comparatively less in the mucosal surfaces. More over the organism that predominated in these regions are the Streptococcus species especially the *Streptococcus sanguis* and *Streptococcus Oralis* group, where in our study the predominant organism were *streptococcus*. In our study the microorganism isolated from the tongue was very less and they organism that was present was of *Streptococcus mutans*. Whereas the tongue in normal individuals are heavily colonized by high bacterial density such as the S. *salivarius*

and *mitis* group and not *the streptococcus mutans* (Allcock *et al.,* 2001). Careful interpretation of our results are important. Although the raise in values could be as documented, the quantitation as such could be influenced by oral hygiene & dietary practises, DMFS index, harmful habits practised by the patients. These parameters were noted in the history but were not included in the study parameters.

Conclusion

Our study, in an overview shows significant difference in isolation and yield of *Streptococcus mutans* between the Diabetes Mellitus and Non Diabetes Mellitus group of subjects from all the surfaces studied except for Ventral surface of Tongue. The isolation was as expected, the same from Caries tooth, in all the study groups but yield being higher in Diabetes group. Gingiva was the next frequent surface yielding more colonies. The study thus importantly highlights two aspects. Firstly, the yield of *Streptococcus mutans* for reasons is higher in Type II Diabetes Mellitus over Type I Diabetes Mellitus although Dental caries is common in both groups. Secondly, *Streptococcus mutans* have been isolated with higher frequency in the mucosal surfaces where generally isolation is not possible. Although reasons like increased blood glucose correlated to Hypo salivation and Neuropathy can be attributed to this, these are common to both types of Diabetes Mellitus; this cannot substantiate a higher yield in Type II Diabetes Mellitus compared to Type I Diabetes Mellitus. Type II Diabetes Mellitus is now proven to be caused by the chronic low grade inflammation induced by altered gut flora leading further to disturbed energy metabolism. We have studied only a species of Streptococci and a study on wider range of selective groups of organisms will open up our view point in this area.

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REFERENCES

Alaluusua, S., Myllarniemi, S., Kallio, M. 1989. Streptococcus mutans infection level and caries in a group of 5 year old children. *Caries Res.,* 23:190-4.

Allcock, G.H., Allegra, M., Flower, R.J., Perretti, M. 2001. Neutrophil accumulation induced by bacterial lipopolysaccharide: effects of dexamethasone and annexin 1. *Clin. Exp. Immunol.,* 123: 62–67.

Canepari, P., Zerman, N., Cavalleri, G. 1994. Minerva stomatol Lack of correlation between salivary Streptococcus mutans and lactobacilli counts and caries in IDDM children, Nov; 43(11):501-505.

Dandona, P, Aljada, A, Bandyopadhyay, A. 2004. Inflammation: the link between insulin resistance, obesity and diabetes. *Trends Immunol.,* 25: 4–7.

Davie, J.R. 2003. Inhibition of histone deacetylase activity by butyrate. *J Nutr.,* 133(Suppl.): 2485S–2493S

Gamboa F., Estupinan, M., Chaves, M., Galindo, A. 2008. Bacteriocins in S. mutans strains isolated from children with and without dental caries: biotypes and sensitivity to antibiotics. *Acta Odontol Latinoam,* 21:97-104.

Gao, Z., Yin, J., Zhang, J., *et al*. 2009. Butyrate improves insulin sensitivity and increases energy expenditure in mice. *Diabetes*., 58:1509–1517

Greenberg, M.S., Glick, M., Ship, J.A. 2008. Burket's Oral Medicine, Eleventh edition. Hamilton: BC Decker Inc.

Kuo, L.C., Polson, A.M., Taeheon, K. 2008. Associations between periodontal diseases and systematic diseases: a review of the inter-relationships and interactions with diabetes, respiratory disease, cardiovascular diseases and osteoporosis. *Publ Health,* 122: 17.

Leach, S. A., and M. L. Hayes, 1968. A possible correlation6.. Rolla, G., P. Bonesvoll, and R. Opperman, 1979. Interactionsbetween oral streptococci and salivary proteins, p. 227-241. InI. Kleinberg, S. A. Ellison, and I. D. Mandel (ed.), Proceedings: saliva and dental caries (a special supplement to Microbiology Abstracts). Information Retrieval, Inc., Washington, D.C.

Marcotte, H., Lavoie Mc. 1998. Oral Microbial Ecology and the Role of Salivary Immunoglobulin A. *MicrobiolMolBiol Rev*., 62(1): 71-109.

Maximilian Zeyda Thomas M. StulnigObesity, 2009. Inflammation, and Insulin Resistance – A Mini-Review Gerontology, 55:379–386

Murrah, V. A. 1985. Diabetes mellitus and associated oral manifestations. *J of Oral Pathol.,* 14, 271-281

Piattelli, A., Pelliccia, P., Sabatino, G., Chiarelli, F., Trisi, P. 1989. [Clinical study on the prevalence of dental caries in patients with juvenile diabetes mellitus]. *Minerva Stomatol.,* 38(11):1227–30.

Rao, D.D., Desai, A., Kulkarni, R.D., Gopalkrishnan, K., Rao, C.B. 2010. Comparison of maxillofacial space infection in diabetic and nondiabetic patients. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod.,* 110:7–12.

Ryan, M.E., N.S. Ramamurthy, M. E. Ryan, N. S. Ramamurthy, T. Sorsa, and L. M. Golub, 1999. "MMP-Mediated events in diabetes," Annals of the New York Academy of Sciences, vol. 878, pp. 311–334.

Sakeenabi, B., and Hiremath, S.S. 2011. Dental Caries Experience and Salivary Streptococcus Mutans, Lactobacilli Scores, Salivary Flow Rate, and Salivary Buffering Capacity among 6-Year-Old Indian School Children. *Journal of International Society of Preventive and Community Dentistry,* 1 (2) 45-51

Salonel, L., Allander, D., Bratthall, D., Hellden, L. 1990. Mutans streptococci , oral hygiene and caries in an adult Swedish population, *J Dent Res*., 69 :1469-75

Samuel J McKenna, 2006. Dental management of patients with diabetes. Dent Clin N Am 50, 591-606.

Scully, C., Bagán, J.V., Eveson, J.W., Barnard, N., Turner, F.M. 2008. Sialosis: 35 cases of persistent parotid swelling from two countries. *Br J Oral Maxillofac Surg.,* 46:468–72.

Tagelsir, A., Cauwels, R., van Aken, S., Vanobbergen, J., Martens, L.C. 2011. Dental caries and dental care level (restorative index) in children with diabetes mellitus type 1. *Int J Paediatr Dent.,* 21(1):13–22

Taylor, G. W. 2003. "The effects of periodontal treatment on diabetes," *Journal of the American Dental Association*, vol. 134, pp. 41S–48S.

Terry D Rees, 1994. The diabetic dental patient. *Dent Clin N Am.,* 38(3):447-63.

Wellen, K.E., Hotamisligil, G.S. 2005. Inflammation, stress, and diabetes. *J Clin Invest.,* 115: 1111–1119

Zaura, E., Keijser, B.J., Huse, S.M., Crielaard, W. 2009. Defining the healthy ''core microbiome'' of oral microbial communities. *BMC Microbiol.,* 259: 12.

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