Original Article

Effect of gestational diabetes on plaque micro flora and periodontal health

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ABSTRACT

Purpose: Increased sex hormones during pregnancy exaggerate gingival tissue response to plaque. Diabetes can cause change in plaque micro-flora and modify the host response. Gestational diabetes mellitus (GDM) affects approximately 4-10% of all pregnancies and is associated with significantly increased risks of maternal and infant morbidity and developing diabetes in later life, but its effect on maternal periodontal health is not well documented. Hence this study was performed to assess effect of gestational diabetes on plaque micro flora and periodontal health. Methods: The study was conducted in pregnant women and consisted of two groups: Gestational diabetes mellitus patients (GDM) and non gestational diabetes mellitus patients (NGDM). The periodontal parameters measured were Plaque Index (PI), Gingival Index (GI), Gingival Recession (GR), Probing Pocket Depth (PPD) and Clinical Attachment Loss (CAL). Bacteriological sampling and culture were done to check the microbial flora of the plaque. The results were analyzed and depicted statistically. Results: There was a significant difference in the three of periodontal parameters of both the groups i.e. PI, GI and GR. After bacteriological culture and sampling the GDM patients showed a significant difference in the number of colony count as well as gm-ve rods. Conclusions: Gestational Diabetes resulted in anaerobic rod dominated plaque micro-flora which could have resulted in increased severity of periodontal disease in such patients.

Introduction

Gestational diabetes mellitus (GDM) is a condition defined as any degree of glucose intolerance that starts or is first recognized during pregnancy, and it is characterized by recent hyperglycemia as а consequence of an association between insulin resistance and inadequate insulin secretion (Buchanan et al, 2007) [1]. Gestational diabetes mellitus appears to result from the same broad spectrum of physiological and genetic abnormalities that characterize diabetes outside of pregnancy. Gestational diabetes mellitus is the most common complication and metabolic disorder observed during pregnancy. Its

prevalence varies between 1 and 14% depending upon the diagnostic criteria and population.

Periodontal disease is an infectious disease characterized by the destruction of the periodontal tissues leading to loss of tooth support. It has a multifactor etiology and pathogenesis, resulting from interaction between environmental, acquired, and genetic risk factors (Nishihara and Koseki, 2004) [2]. The development of periodontal disease is a highly

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Pair	Mean difference	t value	P value	Remark
GDM- NGDM	-0.07533	4.665	0.0001	Significant

TABLE 1.Comparision of plaque index

Pair	Mean	t value	P value	Remark
	difference			
GDM-	-0.04067	2 477	0.0196	Significant
NGDM	0.0+007	2.777	0.0170	Significant

TABLE 2.Comparision of gingival index

communicative and interactive process between pathogenic components in the dental plaque, the host tissues (including epithelium), the vasculature, immune systems, the connective tissue cells and their matrix.

Diabetes has been associated with increased prevalence and severity of gingivitis. Poorly controlled diabetes mellitus subjects had significantly greater bone loss and attachment loss, than did well control diabetes mellitus subjects. Periodontal diseases and diabetes mellitus are closely associated and the understanding of the relationship between periodontitis and diabetes including the factors associated with coexisting synergies has been established.

Bacterial plaque has been established as the primary etiological factor for the initiation of periodontal disease. Hormonal changes during Pregnancy and changing micro flora in Diabetes have been suggested as important modifying factors that may influence the pathogenesis of periodontal diseases [3].

A few studies have suggested that pregnancy is a modifying factor of periodontal disease (Laine, 2002). Increased vascularization and gingival inflammation have been reported as a result of an increase in estrogen and progesterone levels during pregnancy which also leads to changes in the oral micro flora (Kornman and Loesche)[4,5].

MATERIAL AND METHODS:

Patient Selection: The patients who had taken part in the present study were chosen from among the patients who attended the Dr. Ganorkar Maternity Hospital, Nashik, Maharashtra. Periodontal assessment was done in all the patients who visited to the hospital, for routine check-up. Patients with chronic periodontitis were selected and glucose tolerance test (GTT) was carried out. After that patients were divided into 2 groups, 15 patients in gestational diabetes mellitus (GDM) group, and 15 patients in non gestational diabetes mellitus (NGDM) group. (Fig 1 a).

Inclusion Criteria:

- 1. Pregnant patients in the age range of 20-30 year.
- 2. Diagnosed with Chronic Periodontitis with at least ≥ 2 sites with pocket depth ≥ 5 mm.
- 3. GDM group
 - a. Positive GTT
- 4. NGDM group
 - a. Negative GTT

Exclusion criteria:

- 1. Patients who had received periodontal therapy in the last 6 months.
- Patients who had received antibiotics in the last 6 months.
- 3. Patients with other systemic disorders.
- 4. Alcoholics and smokers.

Criteria for assessment of gestational diabetes: Gestational diabetes was assessed based on the laboratory report obtained from the hospital. Patients were given 100mg of oral glucose and their blood sugar levels were evaluated at baseline (fasting), 1

Pair	Mean difference	t value	P value	Remark
GDM-	0.2800	1 912	0.0906	Not Quite
NGDM	-0.2800	1.815	0.0800	Significant

TABLE 3.Comparision of probing pocket depth

Pair	Mean	t value	P value	Remark
	difference			
GDM-	-0.3733	3.047	0.0050	Significant
NGDM				

TABLE 4.Comparision of gingival recession

hour, 2 hour and 3 hours. If 2 of the 4 values in the Glucose Tolerance Test were at or above the cut-off levels (fasting glucose >95 mg/dl, one-hour glucose > 180 mg/dl, two hour glucose > 155 mg/dl, or three-hour glucose > 140 mg/dl), then it was diagnosed as gestational diabetes mellitus.[6]

Periodontal status evaluation: Clinical parameters assessed for the study were Plaque Index (PI), Gingival Index (GI), Gingival Recession (GR), Probing Pocket Depth (PPD) and Clinical Attachment Loss (CAL).

Criteria for site selection: Sites for plaque collection in both the groups were based on the following criteria: presence of bleeding on probing, and probing pocket depth \geq 5mm.

Procedure for sample collection: Subgingival plaque samples were collected from the sites of teeth with the deepest pocket by means of a sterile paper points. They were transferred into a carrier medium containing 5ml of Robertson's cooked meat medium and transferred to the microbiology laboratory.

Processing of samples: Agar plates used in the study were divided into four sections. Collected samples were spread evenly on the blood agar with the help of spreader in a semi-quantitative method. After plating,

blood agar plates were transported into anaerobic gas jar (volume 3.5 Liter), anaerobic gas pack was placed into the jar. (Anaerobic gas pack is a disposable oxygen-absorbing and carbon dioxide-generating agent). 1 sachet of anaerobic gas pack per jar of volume 3.5 liters was required. Anaerobic jar was closed tightly after placing gas pack because oxygen absorption and carbon dioxide generation starts immediately on contact with air. Subsequently the jar was placed into the incubator for 48hrs at 37^{0} c. After 48hrs, the plates were removed from the incubator and colonies of bacteria were counted from both plates, and gram staining of bacterial culture was done for identification of bacteria.

Statistical analysis: Mean and standard deviation was estimated from the sample for each study group. Mean values were compared among different study groups by using independent "t" test and p<0.001 was considered as the level of statistical significance. The statistical software SPSS was used for the analysis of the data.

RESULTS:

The results of the comparison of periodontal parameters of the two groups are depicted in tables 1, 2, 3, 4, 5 and graph 1. The GDM group showed higher values for all periodontal parameters as compared to the NGDM group The difference was statistically significant in three parameters i.e. Plaque Index (p=0.0001), Gingival Index (p=0.0196), and Gingival Recession (p=0.0050). The difference of Probing Pocket Depth was not so significant (p=0.0806) and that of Clinical Attachment Loss was not significant (p=0.1627). The results of the bacteriologic culture and sampling are depicted in fig. 1 & 2 and fig. 3 & 4.

Pair	Mean difference	t value	P value	Remark
GDM- NGDM	-0.1467	1.434	0.1627	Not Significant

TABLE 5.Comparision of clinical attachment loss

Graph 1: Comparison of periodontal parameters between test and control group at baseline

There was an increase in the bacterial count as well as the number of gm –ve rods in the GDM group as compared to the NGDM group.

DISCUSSION:

The state of periodontal disease can be defined as an imbalance between the quality and quantity of bacterial micro flora colonizing the periodontal pocket and the immunological potential of the host, which can be modified by several risk factors. Despite the widespread acceptance of the specific plaque hypothesis, in the etiology of chronic periodontitis, periodontal pathogens are frequently detected in periodontally healthy individuals. Nevertheless, once formed, deep periodontal pockets can provide a suitable environment that further selects specific anaerobic bacterial complexes. Factors that alter this sub gingival environment include inflammation and the myriad of immune and metabolic factors that can influence the composition of the sub gingival biofilm [7].

In this context, factors such as diabetes that alter the nature of the immune/inflammatory response could conceivably influence which bacterial complexes form sub gingivally. While certain bacterial species are more commonly found in diabetic patients, it is more difficult to determine whether this occurs because of direct alterations to the sub gingival microenvironment or whether it occurs indirectly by alterations to the host response. Diabetic individuals may be more susceptible to chronic periodontitis as a result of hyperglycemia altering the sub gingival microenvironment such that bacterial species that are more pathogenic in nature will become dominant.

Clinical periodontal disease has been previously associated with Gestational Diabetes Mellitus in crosssectional studies. Novak et al [8] in a cross-sectional study of 4244 pregnant women found that those with gestational diabetes (113 women) had a much higher prevalence of periodontal disease. Their study further found that this increase in periodontal disease was associated with an increase in the levels of dental plaque.

Xiong et al [9], in another cross-sectional study of 53 pregnant women with gestational diabetes and 106 pregnant women without Gestational Diabetes found that 77.4% women with Gestational Diabetes had periodontitis, whereas only 57.5 % of the women without gestational diabetes had periodontitis. Their results indicated a statistically significant association of periodontitis to Gestational Diabetes mellitus.



Figure 1 a : Flow chart for patient selection

A.P. Dasanayake, tried to correlate the micro flora from sub gingival micro flora procured from pooled samples from first molar teeth, to the overall periodontal status of the patient. He then further tried to correlate sub gingival plaque samples with microbiological samples from the vagina and cervix. The study was proposed to assess if altered periodontal status in a gestational diabetic patient could alter pregnancy outcomes. The Dasanayake study was however unable to make a correlation between oral sub gingival micro flora and cervical or vaginal micro flora.

In present study there was significant difference in periodontal parameters in two groups. Both the group patients, i.e. those with gestational diabetes mellitus and non gestational mellitus showed significant difference in Plaque Index (p=0.0001), Gingival Index (p=0.0196), and Gingival Recession (p=0.0050). There was no significant difference in Probing Pocket Depth (p=0.0806), Clinical Attachment Loss (p=0.1627) in both the groups.

On microbial culturing the samples of the GDM group showed more number of bacterial colony count on blood agar plate as compared to the NGDM group. After gram staining of selected samples of colonies from blood agar plate of the patients with gestational diabetes mellitus, showed more number of gm –ve bacteria while patients with non gestational diabetes mellitus showed less number of gm –ve bacteria.



Figure 1: Blood agar plate of test group showing more bacterial colonies involving all 4 quadrants



Figure 4: On gram staining control group showing less number of gram negative rods



Figure 2: Blood agar plate of control group showing less bacterial colonies involving 2 quadrants



Figure 3: On gram staining test group showing more number of gram negative rods

Results of the present study showed that there was significant difference in periodontal findings of both the group except for probing pocket depth and attachment loss, and significant difference in bacterial colony count with more number gram negative rods in patients with gestational diabetes than non diabetic pregnant patients, indicating that gestational diabetes affect the periodontal health of patients.

Results of the present study showed that gestational diabetes affect the periodontal health of the patients, with change in plaque micro flora, increase in large number of gram negative bacteria.

CONCLUSION:

Gestational diabetes patients showed more gm –ve rod dominated plaque micro flora than normal pregnant patients. This may have resulted in increased severity of periodontal disease is such patients. Thus optimal plaque control in GDM patients could significantly reduce incidence and severity of periodontal disease.

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