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Original Article

Microbiology Section

Variations in the Oral Anaerobic Microbial Flora in Relation to Pregnancy

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ABSTRACT

Introduction: Pregnancy gingivitis is a major oral infection. Periodontium acts as a reservoir of inflammatory mediators and sub gingival biofilms of bacteria.

Aim: To evaluate the anaerobic oral microbial flora in pregnant women before delivery and after delivery by comparing them with control group.

Material and Methods: The study group included fifteen cases of pregnant women before and after delivery and healthy non-pregnant women of same age as control group. Sub gingival plaque samples were collected with the help of dentists. The samples were inoculated immediately into Thioglycollate broth (MV010), transported to the laboratory, inoculated on to selective media for anaerobes (Hi-media laboratories) incubated anaerobically (Gas pack).

Results: Prevotella, Tanerella forsythia, Porphyromonas gingivalis and Fusobacterium nucleatum, Veillonella, Peptostreptococcus were isolated.

Discussion: The anaerobic bacteria in pregnant women were Prevotella, Tanerella forsythia and Porphyromonas gingivalis. Viellonella and Peptostreptococcus were seen in control group and after delivery. Research suggests that periodontal pathogens may travel the blood stream from the oral cavity to the placenta.

Conclusion: Pregnancy has significant effect on periodontal tissue. There is a significant alteration of bacterial flora during and after pregnancy. Oral health has to become a part of antenatal care /check up.

Key Words: Oral anaerobic flora, pregnancy, gingivitis

INTRODUCTION

The numerous physical and the physiological changes that occur during pregnancy [1] affect every major body system and they result in localized physical alterations in many parts of the body, which include the oral cavity [2]. During pregnancy, the inflammatory response to the dental plaque is increased, leading to swollen gingivae, which tend to bleed on brushing. The gingivitis which is caused by the hormonal changes which occur pregnancy is known as pregnancy gingivitis [3]. It is considered to be the most common oral manifestation of pregnancy, as it has been reported to occur in up to 100% of the pregnant women. Pregnancy gingivitis becomes apparent in the second month of gestation and it worsens as the pregnancy progresses, before receiving a peak in the eighth month. In the last month of the gestation, the gingivitis usually decreases and immediately post partum, the gingival tissues are found to be comparable to that of normal women [4].

Robert Durlacher et al., [5] demonstrated that pregnancy gingivitis was not caused by an increase in the dental plaque. It could be due to effect of the pregnancy on the gingival tissues, where both the oestrogen and the progesterone receptors are found. Although the exact mechanism of the inflammation is not known, there are alternations in the immune system and changes in the connective tissues. The key developments are a decrease in the number of neutrophils, decreased chemotaxis and phagocytosis and a depressed antibody response and cell immunity [6]. As a result, there is a sub gingival biofilm formation of the bacteria. They are *Prevotella intermedia (P.intermedia) Porphyromonas gingivalis (P.gingivalis), Treponema denticulate and Actino bacillus actino mycetemcomitans* [7]. There is an increase in the selective growth of the *P.intemedia, P.gingivalis* and *Tanerella* species, which has been demonstrated in sub gingival plaque during the onset of pregnancy gingivitis. This increase in the selective growth may be due to the organisms which utilize the hormone, progesterone as a source of their nutrition, the changes in the immune system, the local changes in the gingival crevices, such as bleeding gingivae, which provide the further nutrients and the increased pocket depths which create a more favourable environment for the anaerobes [8]. The periodontal diseases have potential harmful effects on pregnant women and their developing foetuses. The presence of bacterial infections results in the activation of the cell mediated immunity and the subsequent production of interleukins such as the tumour necrosis factor and prostaglandin- PGF2. These inflammatory mediators may trigger a preterm birth and a low birth weight [9]. More recently, it has been suggested that subclinical infections may also pose a challenge in the developing foetuses.

The aim of the present study was to know the variations in the oral anaerobic microbial flora during pregnancy and after delivery, by comparing them with those in the control group and to know about the pregnancy outcome.

MATERIALS AND METHODS

This study was conducted at Mamata General Hospital, Khammam, Andhra Pradesh, India after obtaining permission from its ethics committee. The study group included 15 antenatal women and they were followed up after 3 weeks of their deliveries. 15 healthy, non pregnant women served as the controls.

After an informed consent was obtained from the women, their sub gingival plaque samples were collected with the help of dental surgeons. Immediately, they were inoculated into the Thioglycollate medium (MV010) and they were transported to the laboratory without any delay. They were inoculated into Brewer's anaerobic agar (M491), Kanamycin bile esculin agar (M1035) and anaerobic blood agar (M1345), which were supplied by Hi – Media Laboratories.(Mumbai, India) The plates were incubated in an anaerobic jar by using an anaerobic gas pack (LE 002A). The jar was opened after 72 hours and the organisms were isolated on the basis of their colony morphologies and biochemical reactions (Flow charts of Barton and Citron MD and Koneman et al.) [10].

RESULTS

The organisms which were most commonly detected in both the groups were: *Vielonella, Tanerella forsythia, Prevotella intermedia, Porphyromonas gingivalis, Peptosreptococcus and Fusobacterium nucleatum.* In the group I specimens, *Propionebacterium, Mobiluncus and Candida sps* were also found in the single samples. [Table/Fig-1].

[Table/Fig-2] shows the total number of organisms in the antenatal, post-natal, and the control groups. *Vielonella and Peptostreptococcus* were present in higher numbers in both the groups. *Tanerella forsythia* was present in two patients out of fifteen in the pregnant group, which was reduced to only 1 during the post-partum visit. P*revotella intermedia* was present in four patients out of 15 in the pregnant-group as compared to 2 in the non-pregnant group, and the count was reduced to 2 during their post-partum visits. *Porphyromonas gingivalis* was present in 5 patients out of 15 in the pregnant-group as compared to only 1 in the non pregnant group and the count was reduced to 3 during the 2 month post-partum visit. *Fusobacterium nucleatum* was present in only 2 patients out of 15 in the pregnant group, which was reduced to 1 during the post-partum visit, and it was not detected in the non pregnant group.

Organism	Antenatal (n=15)	Post-natal (n=15)	Control (n=15)
1. Vielonella	10 (66%)	9 (60%)	10 (66%)
2. T. forsythia	2 (13%)	1 (6%)	0
3. P. intermedia	4 (27%)	2 (13%)	2 (13%)
4. P.gingivalis	5 (33%)	3 (20%)	1 (6%)
5. Peptostreptococcus	8 (53%)	8 (53%)	8 (53%)
6. F. Nucleatum	2 (13%)	1 (6%)	0

[Table/Fig-2]: Comparison of mean % of major micro-organisms detected between Group I and Group II

There was no adverse pregnancy outcome in all the postnatal cases in our study.

DISCUSSION

Pregnancy can influence the gingival health. For the colonization of bacteria at the sub gingival site, the host immune response is important. In pregnancy, the immune response in the periodontium is affected, with the overall effect being one of the decreased activity and the efficiency [8,11]. Although the casual role of the specific bacteria in pregnancy associated gingivitis has been difficult to

Sl. No.	Group I		GRUP II
	Antenatal Specimen	Postnatal Specimen	
1	Veilonella, Prevotella intermedia	Peptostreptococcus, Mobiluncus	Veilonella, Peptostreptococcus
2	Tanerella forsythia, Porporomonas gingivalis	Veilonella Propionibacterium Acne	Peptostreptococcus
3	Peptostreptococcus	Prevotella intermedia	Prevotella Intermedia,Veilonella
4	Porporomonas gingivalis, Peptostreptococcus, Veilonella	Fusobacterium nucleatum Peptostreptococcus	Peptostreptococcus
5	Tanerella forsythia, Veilonella	Porporomonas gingivalis Veilonella	Peptostreptococcus
6	Porporomonas gingivalis, Peptostreptococcus, Veilonella	Porporomonas gingivalis Veilonella	Veilonella
7	Porporomonas gingivalis Veilonella	Tanerella forsythia, Veilonella	Veilonella, Peptostreptococcus
8	Prevotella intermedia, Fusobacterium nucleatum Peptostreptococcus, Candida Spp	Veilonella, Peptostreptococcus	Veilonella, Peptostreptococcus
9	Veilonella, Peptostreptococcus	Veilonella	Veilonella, Peptostreptococcus
10	Porporomonas gingivalis Propionibacterium acne	Veilonella, Peptostreptococcus	Porporomonas gingivalis,Veilonella
11	Fusobacterium nucleatum, Veilonella	Veilonella Prevotella intermedia	Porporomonas gingivalis,Veilonella
12	Veilonella, Peptostreptococcus	Peptostreptococcus	Veilonella, Peptostreptococcus
13	Veilonella, Prevotella intermedia	Porporomonas gingivalis Peptostreptococcus	Veilonella, Peptostreptococcus
14	Veilonella, Peptostreptococcus	Veilonella, Peptostreptococcus	Veilonella, Peptostreptococcus
15	Porporomonas gingivalis Peptostreptococcus	Peptostreptococcus	Peptostreptococcus

[Table/Fig-1]: Microbiological results for subjects of Group I and Group II

establish, the gingival bleeding and the inflammation appear to be associated with a rise in the number of gram negative rods which are present [8]. However, an increase in the selective growth of *P.intermedia* [5,12], *P gingivalis* [11] and *Tanerella* species [13] has been demonstrated in the sub gingival plaque during the onset of pregnancy gingivitis. In our study, the anaerobic bacteria in the pregnant woman were mainly *Prevotella intermedia, Taneralla forsythia* and *Porphyromonas gingivalis,* which were mainly responsible for the periodontitis. After the delivery, the anaerobes which were isolated were the same as that in the control group, except in one case.

A recent case control study which was done by Offenbacher et al., suggested that there were increased levels of inflammatory mediators in the gingival crevicular fluid and that the organisms which were isolated were *Tanerella forsythia, P.gingivalis* and *A.actinomycetemocomitans* in the mothers of the preterm low birth weight infants than in the controls [14].

Although the periodontits during pregnancy was associated with a pre term birth in many studies, in our study, none of the mothers had pre term births.

A recent review which was written by Xinong et al concluded that although the periodontal disease may be adversely associated with the pregnancy outcome, a further methodologically rigorous research was required, as the isolation depended on various factors like the probing depth, etc. [15].

CONCLUSION

Pregnancy has significant effects on the periodontal tissues and pregnancy gingivitis is a common manifestation. There is a significant alteration in the bacterial flora during and after pregnancy. Further research is required to establish the association between the periodontal health and the adverse pregnancy outcome. There is a need for an effective communication between the dental and the medical disciplines to ensure that pregnant woman receive the best possible oral, obstetric and general health.

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