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Periodontitis, a marker of risk in pregnancy for preterm birth

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Abstract

Background: Why chronic periodontitis may induce an inflammatory response with premature pregnancy termination is unclear.

Aims: (1) To assess if periodontitis predicts premature gestation; (2) to study amniotic fluid cytokines and periodontitis variables in early-stage pregnancy.

Material and Methods: A periodontal examination and collection of amniotic fluid was performed (weeks 15–20) of pregnancy in 36 women at risk for pregnancy complications. Amniotic fluid (bacteria), vaginal smears and intra-oral plaque samples were studied. Cytokine levels in amniotic fluid were studied in relation to other study variables.

Results: Periodontitis was diagnosed in 20% of normal and in 83% of preterm birth cases ($p<0.01$). Bacteria were never found in the amniotic fluids studied. Sub-gingival plaque samples including bacteria in the orange and red complexes were found in 18% of full-term 100% of preterm cases ($p<0.001$) and total colony-forming units (CFUs) were higher in preterm birth ($p<0.01$). Amniotic levels of interleukin (IL)-6 and prostaglandin-E_2 (PGE_2) were higher in preterm cases ($p<0.001$). Amniotic IL-6 ($r=0.56$, $p<0.01$) and PGE_2 ($r=0.50$, $p<0.01$) cytokine levels were correlated with CFU from sub-gingival plaque samples ($r^2=0.44$).The odds ratio of preterm delivery and having periodontitis was 20.0 (95% confidence interval (CI): 2.0–201.7, $p<0.01$). The odds of >60 CFU in sub-gingival plaque and preterm birth was 32.5:1 (95% CI: 3.0–335.1, $p<01$).

Conclusions: Pregnant women with findings of elevated amniotic fluid levels of PGE_2, IL-6 and IL-8 in the 15–20 weeks of pregnancy and with periodontitis are at high risk for premature birth. The implication of this is that periodontitis can induce a primary host response in the chorioamnion leading to preterm birth.

Key words: amniotic fluid; bacteria; cytokine; orange complex; periodontitis; preterm delivery; red complex

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In a recent systematic review on the associations between periodontitis and increased risk for coronary heart disease and preterm pregnancy outcomes it was concluded that there is limited evidence of an association between periodontitis and these diseases and a need for additional observational and intervention studies (Madianos et al. 2002). In order to perform intervention studies, the causal relationship between preterm gestation and periodontitis must first be investigated and understood. Studies have suggested that women with periodontitis are at greater risk of having a preterm gestation with the result of a low birth weight (Offenbacher et al. 1996). Contradictory conclusions, however, have also been published (Davenport et al. 2002). The differences in conclusions from these two studies might originate in socioeconomic and ethnic factors that may explain 50% or more of the risk for premature gestation (Aveyard et al. 2002, Ferguson et al. 2002, Alexander et al. 2003). Intervention studies, however, have suggested that non-surgical periodontal therapy in pregnant women with periodontitis may reduce the risk for preterm delivery and low-birth-weight births (Lopez et al. 2002, Jeffcoat et al. 2003).

The pathological mechanisms by which chronic periodontitis may cause, or exaggerate, an inflammatory response resulting in premature termination of a pregnancy remains unclear. There is evidence that patients with untreated periodontitis are at greater risk for bacteremia (Daly et al. 2001). It has been postulated that periodontal infection may cause bacteremia including the circulation of endotoxins (i.e. lipopolysaccharides (LPS), hydrolytic enzymes and peptid-glycans) that might trigger a host immune response exaggerating the effects of low-grade inflammation in other organs. Pro-inflammatory cytokines and chemokines (e.g. tumor necrosis factor (TNF)-α, interleukin (IL)-1, IL-6 and IL-8) are involved in

preterm labor (Hitti et al. 2001). Studies of human myometrium have also shown that cytokine release is stimulated by LPS (Sehringer et al. 2000). Furthermore, gingival fluid prostaglandin E_2 (PGE_2) levels can be used as an indirect estimate of the amniotic fluid levels of PGE_2 (Damare et al. 1997). Thus, an elevated PGE_2 level in gingival fluid has been reported to identify periodontitis activity and has also been associated with low birth weight and preterm labor (Offenbacher et al. 1998).

Offenbacher et al. (1998) provided two possible explanations why periodontitis might be a factor in premature gestation. LPS from pathogens associated with periodontitis may activate placental release of TNF-α and IL-1. The local production of these cytokines in the periodontal pocket caused by periodontitis may also result in an elevated serum concentration of such cytokines and therefore also be present in the amniotic fluid. Animal studies have supported the hypothesis of a significant relationship between an increase in serum PGE_2 induced by experimental *Porphyromonas gingivalis* and *Escherichia coli* infections in pregnant hamsters resulting growth deficit and fetal mortality (Collins et al. 1994).

Assuming that the association between periodontitis and premature birth/low birth weight has a shared pathogenesis, early periodontal screening could predict/identify women at risk for premature delivery as a low-cost examination procedure. This would allow measures for the prevention of premature birth that is a high cost condition both to the subjects and to the society.

Therefore, the objectives of the present study were as follows:

- To assess if periodontitis is predictive of premature gestation in an identified population of women at risk for birth complications as a consequence of medical factors (specifically age).
- To assess if there is an association between cytokines in amniotic fluid and periodontitis in the early stage of pregnancy.
- To assess if there is a relationship between bacterial counts in periodontal pockets and cytokines in amniotic fluid.

Material and Methods

The study was ethically approved by the University of Vienna, Austria. A total of 36 women undergoing amniocentesis consented to participate in the present study. These women were scheduled to have the procedure performed for medical reasons. Between the 15th and 20th weeks of pregnancy, amniocentesis was performed by a gynecologist. A total of 2 ml additional amniotic fluid was collected for the purpose of the present study. Until processed the amniotic fluid supernatant was stored at $-80^{\circ}C$. Routine vaginal smears were also collected and studied for ascending bacterial infection. Standard clinical information regarding medical conditions including diabetes mellitus, medications, as well as behavioral factors (smoking history and alcohol consumption) was collected from medical records. The subjects were also asked about their dental habits including whether they had access to dental care and had received routine care. The women were monitored and cared for by their obstetricians consistent with best practice until the end of pregnancy. The study did not include any dental procedures. A preterm birth was defined as a pregnancy of less than 37-week duration and/or a birth weight of less than 2500 g.

During the 15th through the 20th week of pregnancy a full-mouth clinical periodontal examination was performed. This examination included assessment of gingival bleeding and probing depths. Clinical probing depths were measured at four sites per tooth using periodontal mm graded probes. Subjects were defined as having periodontitis, if they presented with at least one site with a clinical probing depth $\geqslant 5.0$ mm in each quadrant not accounting for the probing depths at the distal aspects of the most posterior tooth in the quadrant and if the combination of pathogens from the red and orange clusters in the bacterial samples exceeded 60 colony-forming units (CFUs) ($>$mean value plus $2 \times$ SD among women with no clinical evidence of periodontitis). Radiographic exposures were avoided because of the pregnancy status of the women.

In each subject, the two periodontal sites with the deepest periodontal pockets were selected for microbial sampling. Supra-gingival plaque was removed and the sample sites were isolated from saliva. Sterile endodontic paper points were placed in the periodontal pockets until resistance was felt and left in place for 10 s. The samples were then placed in vials with transport media. Routine aerobic and anaerobic bacteriological cultures were studied from amniotic fluid and from the periodontal plaque samples. Quantitative determinations of bacterial growth (red and orange clusters) were accounted for in CFUs. The quantitative estimations of the total amount of bacteria were made with a spiral plater. Polymer chain reaction (PCR) and bacteriological culturing methods were processed to determine the presence of micro-organisms of the red and orange clusters (Socransky & Haffajee 2003).

The assays for cytokines (IL-1, IL-4 IL-6, IL-8, IL-10, TNF-α, interferon-γ and PGE_2) were performed using enzyme-linked-immuno-absorbent assays (ELISAs) (R&D Systems Minneapolis, MN, USA).

Statistical methods

Descriptive statistics were used to describe the study population. To test the null hypothesis of no difference by grouping variables the Mann–Whitney *U*-test was used because the data were either non-parametric or lacking normal distribution characteristics. As appropriate, Pearson's or Spearman's rank correlation coefficients between parameters were also studied. The utility of CFU and preterm gestation was studied with a receiver-operating curve (ROC). The Mantel–Haentszel common odds ratio estimate was used to assess the odds of agreement between the two conditions (preterm/low birth weight and periodontitis). The SPSS 11.5 statistical software was used for data analysis (SPSS Inc., Chicago, IL, USA).

Results

Subject characteristics for the women and their newborn children are presented (Table 1). In this study of population of women at risk for miscarriage, or preterm delivery only one woman was identified with a smoking habit and she neither presented with evidence of periodontitis nor did she deliver preterm. The 10-scale classification system used at the Department of Obstetrics at the University of Vienna, Austria, to define the number of medical conditions among the newborns with a score of 10 being completely normal, was employed to assess the health of the newborn. Thus, 76.6% of the newborn

Table 1. Mean values and standard deviation (SD) for age, length (cm), weight (kg) and gestation period in full-term and preterm birth for women and newborn children

	Mother full-term Mean ± SD	Mother preterm Mean ± SD	Sign	Child full-term Mean ± SD	Child preterm Mean ± SD	Sign
age	33.9 ± 5.3	30.5 ± 7.2	NS			
length	165.1 ± 6.0	160.5 ± 5.5	NS	51.0 ± 2.6	40.5 ± 2.9	$p<0.001$
weight	67.1 ± 10.8	64.8 ± 10.3	NS	3.39 ± 0.45	2.31 ± 0.28	$p<0.001$
gestation (weeks)	39.5 ± 1.4	33.5 ± 3.8	$p<0.001$			

Sign, significance.

children in the full-term gestation group presented with a score of 10. In the preterm gestation group, 32.3% of the newborn presented with a score of 10. The difference in the number of signs of medical complications between the two groups was statistically significant ($p<0.001$ Mann–Whitney *U*-test).

Clinical periodontal findings

The dental examination performed between weeks 15 and 20 identified that 5/6 (83%) of the women who later had a preterm delivery with low birth weight had a diagnosis of chronic periodontitis as defined by the joint presence of probing depth $\geqslant 5.0$ mm, clinical evidence of gingival inflammation and the presence of pathogens in the orange/red clusters (>60 CFU). None of the subjects presented with clinical signs of aggressive periodontitis. In the group of women, delivering within the normal gestation period, only six subjects (20%) presented with such signs of periodontitis. This difference between this groups was statistically significant ($p<0.01$, Mann–Whitney *U*-test).

No difference in reported access to dentistry was found between the two groups. In fact, all women with a preterm gestation period reported that they had been users of regular dental care. The distribution of clinical probing depths from the periodontal sites (two sites with deepest probing depth in each subject) selected for microbial sampling is presented (Fig. 1). The difference in probing depth was significantly different between women delivering preterm from those delivering full-term ($p<0.001$, Mann–Whitney *U*-test).

Presence of bacteria from amniotic fluid, vaginal smears and from intra-oral plaque samples

Microbial culture studies of amniotic fluid samples taken between weeks 15 and 20 failed to show evidence of microbial growth. Thus, the amniotic fluid was not infected in any of the cases. In addition, at this time period none of the women presented with an ascending bacterial infection as defined by the vaginal bacterial culture results.

Intra-oral sub-gingival plaque samples collected at the dental examination within the 15–20-week period yielded a significantly higher proportion of positive findings of pathogens in the orange and red clusters among women delivering preterm ($p<0.01$). In fact positive findings of pathogens in the orange/red clusters were found in 16.7% of the women with full-time gestation, whereas 83.3% (5/6) in the group delivering preterm had positive findings of those pathogens. The total counts of CFU from dental plaque samples demonstrated that women delivering preterm had significantly higher counts of these pathogens ($p<0.01$) (Fig. 2).

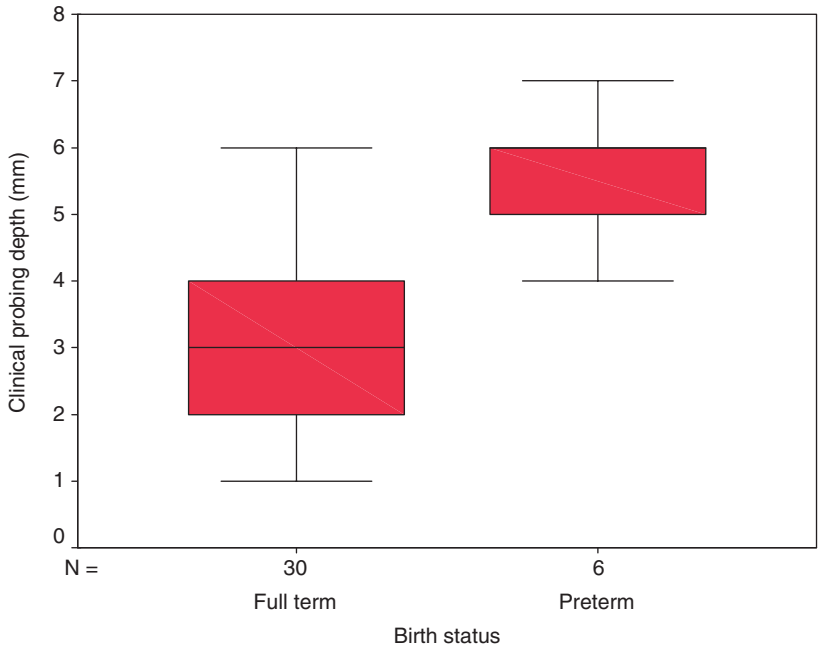


Fig. 1. Distribution of clinical probing depths at selected microbial test sites in women who gave birth at full- or preterm.

Amniotic markers of inflammation

Analysis of amniotic fluid collected between weeks 15 and 20 demonstrated that the levels of TNF- α, IL-1, IL-4, IL-10 and IFN-γ did not differ by gestation period (preterm or not).

The amniotic levels of cytokines IL-6 and PGE_2 were significantly higher in women delivering pre-term ($p<0.001$), whereas IL-8 were significantly higher in those who delivered with a normal course of pregnancy ($p<0.05$) (Fig. 3). These differences with higher IL-6 and PGE_2 levels remained statistically significant for IL-6 and PGE_2 in the pregnant women with a diagnosis of periodontitis (Fig. 4). Furthermore, amniotic IL-6 ($r=0.56$, $p<0.01$) and PGE_2 ($r=0.50$, $p<0.01$) cytokine levels were significantly correlated with CFU from sub-gingival plaque samples ($r^2=0.44$, $p<001$) (Fig. 5). Thus, higher levels of IL-6 ($p<0.01$) and PGE_2 ($p<0.01$) were found in amniotic fluid based on the CFU >60 CFU threshold form high versus low levels of CFU in sub-

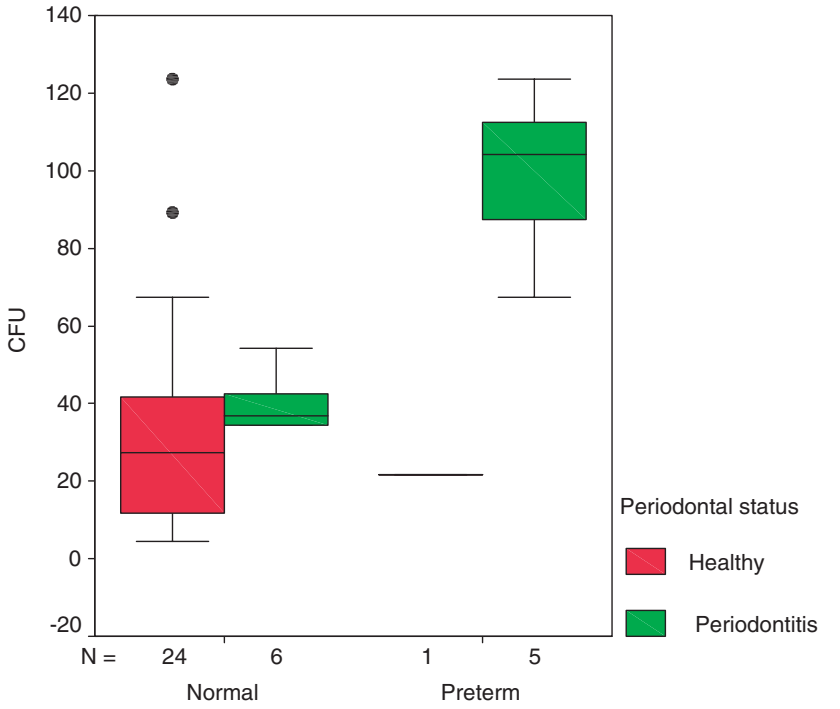


Fig. 2. Colony-forming units (CFUs) for pathogens in the red and orange clusters from dental plaque samples in pre- and full-term birth in women with and without a diagnosis of periodontitis. Dots mark outliers.

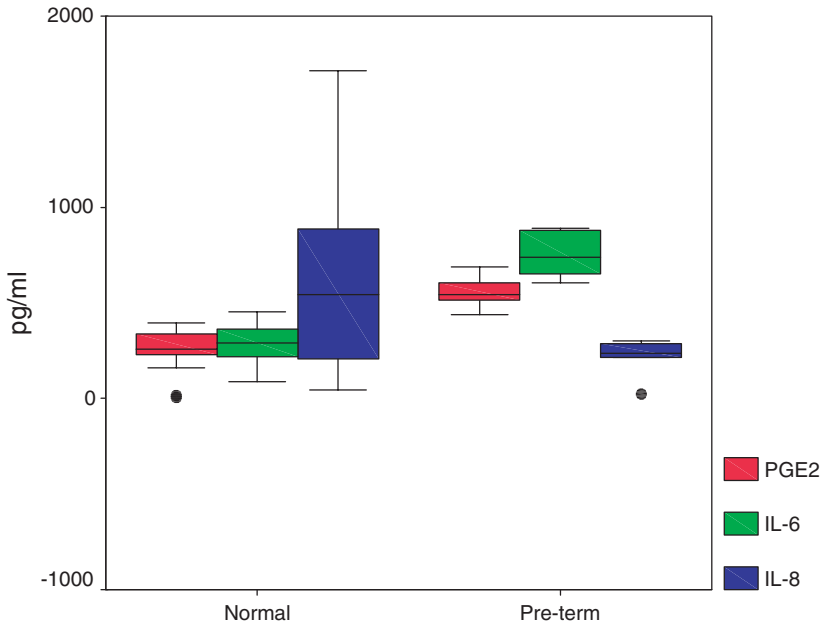


Fig. 3. Boxplot diagram showing the distribution of cytokines interleukin (IL)-6 and prostaglandin-E_2 (PGE_2), IL-6 and IL-8 levels in women delivering on time or as pre-term birth. Dots mark outliers.

gingival plaque samples. When the subjects were identified by periodontal status excluding delivery status the amniotic fluid mean levels of IL-6 was 1.8 times and PGE_2 was 1.5 times higher in those subjects with periodontitis (Table 2).

Assessment of the utility that dental study parameters in predicting the gestation period

The odds ratio of agreements between gestation pre-term status and periodontitis and for gestation preterm status and the odds of having >60 CFU from gingival plaque samples or the presence of pathogens in the orange/red complex in subgingival plaque samples are presented (Table 3). The utility of the CFU in subgingival plaque samples to identify subjects who later would have a low birth weight/preterm delivery is presented in an ROC diagram illustrating a very high level of predictive utility (Fig. 6).

Discussion

The fact that several of the newborn had one or more medical complications independent of preterm/normal delivery time was anticipated because the mothers were all identified as being at elevated risk for a birth complication although not necessarily a preterm birth as such. Obviously the preterm birth in itself was associated with one or more health complications in a majority of the newborns. It is well established that a preterm birth results in expensive postnatal care for the management of birth defects and other diseases common in children born preterm (Petrou et al. 2003). Whether maternal periodontitis may predict future elevated risk for periodontitis in the offspring has not yet been considered.

Different studies on the association between periodontitis and preterm/low birth weight have used different criteria for the definition of periodontitis (Offenbacher et al. 1996, 1998, Davenport et al. 2002, Lopez et al. 2002). It is well established that severe pregnancy gingivitis and pseudo-pockets may be present during pregnancy. Therefore, in the present study both clinical and microbiological data were used for the diagnosis of periodontitis requiring at least four sites with a probing depth $\geqslant 5.0$ mm and with the presence of key pathogens associated with periodontitis. In addition, elevated bacterial counts (CFU) were required for the diagnosis. The microbiological cut-off for a diagnosis of periodontitis set at >60 CFU was chosen based on the approximation of the mean value plus $2 \times$ SD of CFU (CFU mean value 23, SD+19) for the orange/red clusters in the pregnant women ($n = 19$) with no evidence of probing depth >4.0 mm and with no bleeding on probing at the selected sites with the ''deepest'' probing depth. There appears to be no studies that have used CFU to define periodontits in relation to disease status. Although

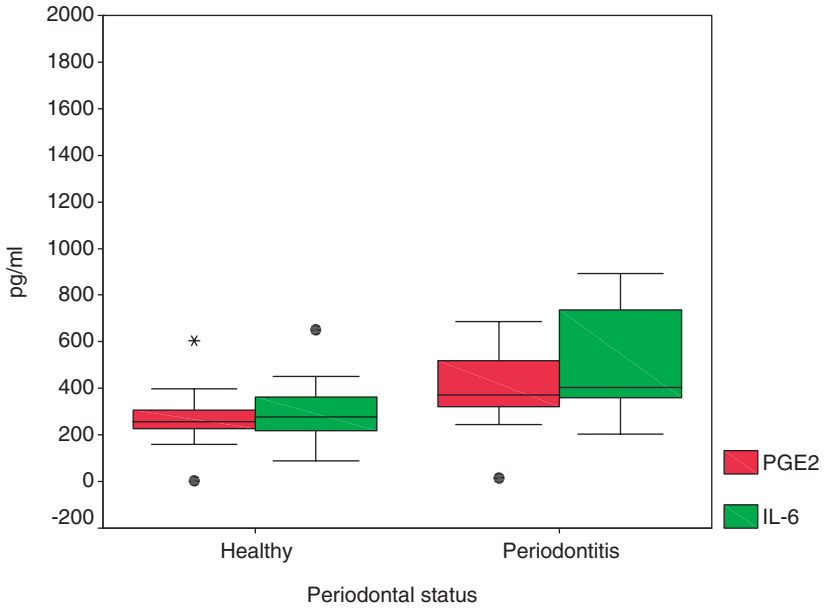


Fig. 4. Boxplot diagram showing the distribution of interleukin (IL)-6 and prostaglandin-E_2 (PGE_2) in pregnant women with or without a diagnosis of periodontitis. Dots mark outliers. Star marks extreme outlier.

Table 2. Cytokine levels (pg/ml) in subjects who delivered on time (1) or preterm (2)

Birth status	IL-6	IL-8	PGE_2
1	205.1	74.29	234.58
1	107	93.5	393.62
1	373.9	1715.41	234.62
1	202.8	160.28	306.33
1	451.5	208.16	163.7
1	97.4	96.49	392.5
1	409.4	43.52	257.82
1	341.7	391.2	253.06
1	246.2	888.76	298.86
1	244.8	1014.58	344.69
1	314.9	743.51	258.23
1	255.8	1036.65	352.68
1	304.7	2047.67	188.91
1	278.3	1323.44	159.41
1	259.2	259.76	292.69
1	212.3	468.74	188.91
1	363.8	560.4	248.45
1	372.8	277.15	218.91
1	218.4	143.08	227.72
1	268.8	203.04	246.21
1	352.1	450.1	396.43
1	204.9	302.59	246.13
1	301.3	631.97	13.89
1	88.97	544.13	3.48
1	277.7	571.02	273.24
1	355.7	540.26	305.67
1	405	885	335.78
1	449.9	951.92	276.88
1	368.3	605.72	363.63
1	362.8	717.62	371.51
2	605.9	21.3	568.79
2	767.5	214.34	521.47
2	651.4	259.76	605.21
2	879.6	286.19	515.57
2	707.4	301.47	687.05
2	891.9	214.34	439.97

IL, interleukin; PGE_2, prostaglandin-E_2.

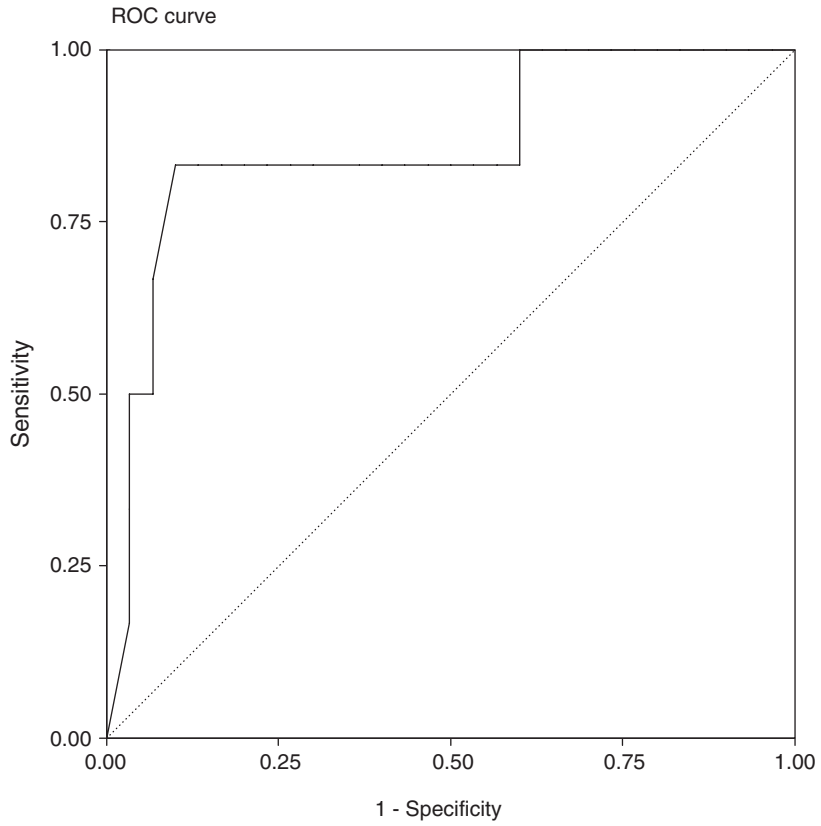
Mitchell-Lewis et al. (2001) have presented data on the average sub-gingival microbial load of single micro-organisms of the orange/red complexes in pregnancy, but there are no data or assessment of the role of total amount of these pathogen clusters in relation to peridontitis.

In the present study, the diagnosis whether the pregnant women had periodontitis or not was made prospectively and by a periodontist who was, obviously, masked to study outcomes. This was in difference to other studies that have made a retrospective diagnosis of periodontitis post-partum (i.e. Offenbacher et al. 1996, Davenport et al. 2002).

The proportion of women who delivered preterm was approximately twice as high as what had been expected or had been reported elsewhere (Romero et al. 1989). This was most likely the consequence of the case selection strategy used in the present study. Between 30% and 70% of all cases of preterm births are associated with an ascending intrauterine infection (Hillier et al. 1988). At the time of study examination, however, none of the women in the present study presented with evidence of ascending intrauterine infection. Furthermore, all women participating in the present study had comprehensive access to medical care within the socio-medical Austrian health insurance program. This program is a dedicated prevention-care program for pregnant women beginning at the time of their knowledge of pregnancy (Mutter-Kind Pass). In difference to other studies all the women in the present study were of European descent (Offenbacher et al. 1996, Davenport et al. 2002). Thus, in the present study ethnic elevated risk factors were not confounding. Furthermore, such a study population is considered to be at lower risk for preterm complication (Alexander et al. 2003).

The inclusion criteria for the pregnant women in the present study were defined by their obstetricians who consistent with the best practices ordered diagnostic amniocentesis. Thus, conditions that were considered for this medical clinical included, age, nullipara and preceding abortions. The amniocentesis results showed no signs of chromosomal defects in the amniotic fluid of the pregnant women studied. Thus, the women participating in the present study were representative of women considered to be at high risk for a birth complication not necessarily associated with preterm but to other conditions including birth of a child with trisomy 21. In difference to other studies (Offenbacher et al. 1996, 1998, Davenport et al. 2002) a majority of the women in the present study (75%) were 31 years of age or older. This may, in part, explain the strong relationship between periodontitis and preterm birth in that the prevalence of periodontitis increases with age. In younger women, periodontitis is rather uncommon. Young women may have several other and non-dental risk factors that may contribute to giving birth premature (Eure et al. 2002). The youngest women (age 18) who delivered preterm also had periodontitis. The other women who delivered preterm were 28 years of age or older.

Vaginal infection is described as a cause for preterm gestation (Gravett et al. 1986). Any infection that may cause chorioamnionitis resulting in elevated cytokine and PG levels in the amnionic fluid including high levels of IL-6 may induce premature rupture and the initiation of birth (Yoon et al. 1998, Lewis et al. 2001, Keelan et al. 2003). At weeks 15–20 of the pregnancies there were no medical indications of a preterm birth condition. At the time of delivery, no non-dental medical causes for preterm delivery could be identified (data not shown). Thus, periodontal infection remained as a plausible cause for a preterm condition. Bacterial infec-



Area under the curve

Test result variable(s): CFU

Area	Std. error [a]	Asymptotic sig. [b]	Asymptotic 95% confidence interval	
			Lower bound	Upper bound
0.861	0.092	0.006	0.680	1.042

a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

Fig. 5. Graphic illustration presented for the three-dimensional relationships between prostaglandin-E_2 (PGE_2), interleukin-6 (IL-6) and colony-forming units (CFU) in the study population. The solid line represents women who delivered preterm birth and the dotted line represents those who delivered full-term birth.

tions such as periodontal infections with a chronic persistent inflammatory production of toxins and metabolites could therefore be considered as causative in inducing an inflammatory process with production of cytokines in the placental area. At the 15th–20th week pregnancy period, the placental barrier would normally prevent the passing of proteins with a molecular weight higher than 10,000 kDa. Thus, only proteins with a molecular weight $< 10{,}000$ kDa could pass the placental barrier. Consistent with this, the present study failed to demonstrate the presence of periodontal bacteria in amniotic fluid. However, cytokines produced as a consequence of the infected periodontium may transfer through intact fetal membranes (Kent et al. 1994).

There are several published review articles on the role of pathogens associated with periodontitis and the potential effect on pregnancy. It has been suggested from an experimental gingivitis study in pregnant women that there is an increase in total bacterial load during pregnancy (Raber-Durlacher et al. 1994). This has also been suggested from the interpretation of serological data from pregnant women (Lopatin et al. 1980). In the present study, high counts of CFU were found in 5/6 of the women who delivered preterm but only in 4/30 of the women who delivered full-term. This finding suggests that the periodontal bacterial load might be a significant factor in the outcome of pregnancy independent of the fact that there might be changes in sub-gingival bacterial counts during pregnancy. In one cohort study of pregnant women it was demonstrated that maternal periodontal infection in the absence of protective serum IgG antibodies and a translocation of a fetal antibody IgM response to pathogens in the orange/red clusters may elicit prematurity (Madianos et al. 2001). In the present study, the data demonstrated that both, CFU and the presence of pathogens in the orange and red clusters from sub-gingival plaque samples were highly discriminatory between women delivering preterm or not. Having such elevated numbers of pathogens associated with periodontitis at an early stage of pregnancy suggests that the oral bacterial load and constant infusing of pathogens in the blood stream could trigger a placental response leading to preterm birth (Hill 1998). It is well known that hormonal changes will trigger a host response to infection. However, the development and changes of the sub-gingival biofilm during the course of pregnancy is poorly described. Further studies of the microbiota in association with pregnancy are therefore needed.

At the 15–20-week period when amniotic cytokines were assayed in the present study, only minor amounts of TNF-α, IL-1β, IL-2, IL-4 and IL-10 were found in amniotic fluid and not discriminatory to pregnancy termination. The amniotic cytokine levels change during pregnancy and others have shown that prior to week 32 also TNF-α and IL-1β can be found in higher concentrations in chorioamnionitis (Baud et al. 1999). Because of the fact that the host responses to a lifetime exposure of pathogens associated with periodontitis is not well documented it is not possible to further explore the levels or the impact of these cytokines identified above in the course of pregnancy with or without evidence of periodontitis.

In the present study, PGE_2, IL-6 and IL-8 were elevated in the amnionic fluid in women who later developed preterm. The fact that TNF-α, IL-1β, IL-2, IL-4 and IL-10 showed low levels in both groups and PGE_2 was high in the preterm birth group suggests that the initial upregulation of PGE_2 had already

Table 3. Odds ratio and 95% confidence intervals (95% CI) between gestation status and clinical diagnosis of periodontitis and microbial findings from sub-gingival plaque samples taken between week 15 and week 20 of pregnancy

Condition	Odds ratio	95% CI	Significance	
Preterm delivery and having periodontitis	20.0	1.95	201.7	$p<0.01$
Preterm delivery and >60 CFU in sub-gingival Plaque samples	32.5	2.97	355.1	$p<0.01$
Preterm delivery and presence of pathogens from the orange/red complexes in subgingival plaque samples	25.0	2.38	262.6	$p<0.01$

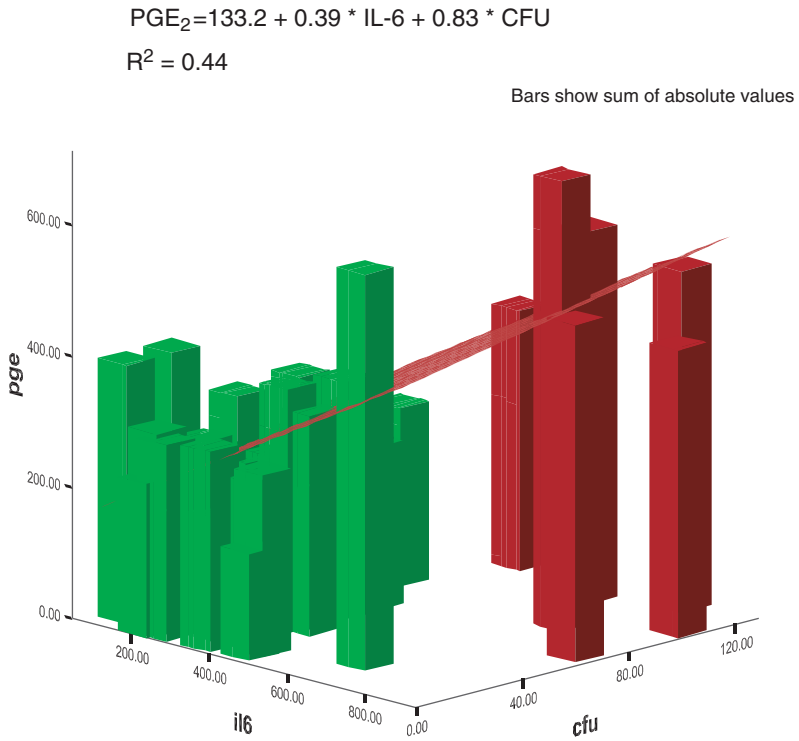


Fig. 6. Graphic receiver-operating curve (ROC) curve presented for the analysis of the relationship between colony-forming units (CFU) >60 CFU as the explanatory variable in distinguishing between pre- and full-term pregnancy outcomes. Area under curve is included demonstrating the significance of predictive value for CFU >60 in early preterm birth prediction.

taken place in the 15–20 week-period and consistent with the periodontal infection burden in these subjects. The fact that the amniotic fluid levels of PGE_2, IL-6 and IL-8 was between 1.5 and 1.8 times higher in subjects with periodontitis suggests that maternal periodontitis may have an important impact on the inflammatory response expressed in the placenta and the fetal conditions. Because the results from the bacterial cultures of the amniotic fluid were negative for all studied cases no other local regulatory mechanisms of increasing cytokine levels could explain the current findings.

The findings forming the present study were based on a relatively small number of subjects ($n = 36$). However, the results were obtained from consecutive consenting women, who in addition to the amniocentesis performed by the obstetrician also accepted the periodontal examination by a periodontist. The analysis of the data demonstrated highly significant differences between those women who delivered full-term from those who delivered preterm. Further studies are needed to confirm the results. The high odds ratio of an association between periodontitis and preterm/low birth weight was unexpectedly high. Prospective multicenter studies should be performed to verify or revise these odds. However, one of the obstacles for such studies is that additional amniotic fluid is necessary for study purposes, beyond what is necessary for routine medical diagnostic uses. It therefore requires that obstetricians are made aware of the needs for such studies. The present study provided such background information.

In conclusion, pregnant women with findings of elevated amniotic fluid levels of PGE_2, IL-6 and IL-8 in the 15–20 weeks of pregnancy and with periodontitis defined by clinical and microbiological parameters are at high risk for premature birth. The implication of this is that periodontitis can induce a primary host response in the chorioamnion leading to preterm birth.

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