C-Reactive Protein Levels and Periodontal Diseases During Pregnancy in Malaysian Women

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Purpose: To investigate the association between plasma CRP levels and periodontal disease during pregnancy.

Materials and Methods: Fifty-six pregnant women attending the Antenatal Clinic, UMMC for their first antenatal check-up consented and were recruited for this study: 28 subjects with diseased periodontium (test group) and 28 subjects with healthy periodontium (control). The test group underwent nonsurgical periodontal therapy and the control group was given oral hygiene education. Periodontal parameters and CRP levels were evaluated at baseline and 6 weeks. Pregnancy outcome data were recorded from the Antenatal Clinic, UMMC.

Results: Plasma CRP levels in the test group were statistically significantly elevated compared to the control group (8.55 ± 5.28 mg/l vs 5.66 ± 2.91 mg/l). After nonsurgical periodontal therapy, a statistically significant reduction in the CRP level in the test group (2.06 mg/l) along with statistically significant improvement in periodontal status in both groups was observed. The mean birth weight for infants of both groups showed no statistically significant difference.

Conclusions: Plasma CRP levels in pregnant women with diseased periodontium were statistically significantly reduced after nonsurgical periodontal therapy. However, no association between CRP levels and adverse pregnancy outcome was observed.

Key words: C-reactive protein, nonsurgical periodontal therapy, periodontal disease, pregnancy

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Periodontal diseases are usually chronic infections that cause inflammation and destruction of periodontium, initiated by bacterial plaque accumulation on the tooth.[45] The severity of the destructive processes associated with these diseases is due to an excessive host response to the bacterial challenge, which is immune mediated. Therefore, periodontal disease is a multifactorial and complex disease[2] with several risk factors that modify the host response towards bacterial assault, such as smoking,[7] diabetes mellitus,[37] obesity,[41] genetic predisposition[14] and conditions that change estrogen levels, such as puberty, pregnancy and menopause.[13] In periodontal disease, bacteria and their virulence factors found in the periodontium directly stimulate various host cells residing in this area.[30] These cells release pro-inflammatory cytokines (tumour necrosis factor alpha (TNF-α), interleukin-1β (IL-1β), IL-6, IL-12, chemo-attractants and prostaglandin (PGE2) in the connective tissue, thus amplifying the inflammatory cascade.[30] Pro-inflammatory cytokines enter the circulation and reach the liver. There they activate hepatocytes, which leads to the synthesis of plasma proteins known as acute-phase proteins, including lipopolysaccharide (LPS), lipopolysaccharide-binding protein (LBP), CD14, complement protein and C-reactive protein (CRP). CRPs opsonise the bacteria, thereby facilitating recognition for phagocytosis.[42]

Increased CRP levels indicate active systemic inflammation in the body. Periodontal disease and the systemic bacteraemia caused by periodontal pathogens are significant contributors to a systemic maternal inflammatory response during pregnancy. In turn, inflammation can serve as an independent biochemical threat to the foetal-placental unit by inducing labour, rupture of membranes and early parturition. Studies have shown that increased CRP levels have been associated with adverse pregnancy outcomes, including preterm birth (PTB),[33] preeclampsia[44] and intrauterine growth restriction.[46]

Therefore, it is postulated that CRP may be a plausible mediator of the relationship between periodontal disease

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and adverse pregnancy outcome. A possible association between periodontal disease in pregnancy and adverse pregnancy outcomes, e.g. PTB and/or low-birth weight (LBW) infant, has been reported.[4] Preterm birth can be defined as the babies born before the completion of 37 weeks of pregnancy.[8] Studies have reported that periodontal therapy which reduces the bacterial load in the oral cavity may reduce the incidence of PTB and LBW infants.[11,38] However, the exact mechanism involved is still unknown. Other studies have reported no effect of periodontal therapy on adverse pregnancy outcomes, but there was improvement in periodontal health.[16,24]

Therefore, the aims of this study were 1. to investigate the association between plasma CRP levels and periodontal disease during pregnancy by evaluating the plasma CRP levels in pregnant women with diseased periodontium and those with healthy periodontium; 2. then to compare the plasma CRP levels in pregnant women with diseased periodontium before and after nonsurgical periodontal treatment; and 3. to determine whether the CRP levels were associated with pregnancy outcomes in both the test and control groups.

MATERIALS AND METHODS

This was a prospective, interventional study where pregnant women with diseased and healthy periodontium were selected. Ethical clearance was obtained from the Medical Ethics Committee of University Malaya Medical Centre (UMMC) (MEC Ref No: 890.91) and the Ethics Committee Board of The Faculty of Dentistry, University of Malaya (No: UM.D/PD/211/11). The pregnant women attending the Antenatal Clinic, UMMC for their first antenatal check-up were selected using the convenience sampling method and invited to participate in the study following informed and written consent. Those subjects who met the inclusion and exclusion criteria were referred to the Periodontal Postgraduate Clinic, Faculty of Dentistry, University of Malaya for further periodontal examination, blood collection and periodontal treatment. The inclusion criteria were (i) pregnant women with gestational age between 12 and 28 weeks, (ii) patients exhibited at least 20 teeth excluding third molars and (iii) patients ages 18 to 40 years. The exclusion criteria were: (i) smoking or a drug abuse history; (ii) presence of medical or systemic conditions such as cardiovascular disease, respiratory problems, diabetes mellitus, kidney disease, haematological disorders, autoimmune diseases, inflammatory disorders or other chronic diseases; (iii) women with multiple fetuses, gestational diabetes and pregnancy-induced hypertension; (iv) periodontal treatment had been done in the past 3 months; (v) course of topical or systemic antibiotics had been administered in the past 3 months.

Sample Size

Power and Sample Size Calculation (PS) Software Version 3.0.43 was used to calculate the sample size for this study. Based on the expected mean difference of 0.5 in the CRP level between the two groups,[39] it was calculated that at least 22 pairs of subjects would be needed to be able to reject the null hypothesis that the response difference is zero with a probability (power) 0.8 (80%). The Type I error probability associated with this test of the null hypothesis is 0.05 ($p = 0.05$).

Fifty-six pregnant women consented and were recruited for this study; 28 subjects with diseased periodontium and 28 subjects with healthy periodontium. All subjects underwent full periodontal assessment at baseline and after 6 weeks. Blood samples were collected for all subjects at baseline and after 6 weeks. The medical records of all subjects were gathered from the Antenatal Clinic of UMMC. The information included: history of any previous pregnancy, expected due date (EDD), patient's medical status, presence of gestational diabetes, pregnancy-induced hypertension in the current pregnancy and other serious medical or systemic conditions. The selected subjects were then asked to complete a questionnaire which included information on age, ethnicity, EDD, household income, occupation, dental history and pregnancy history.

Periodontal Parameters

Periodontal examinations were conducted by one examiner (AA). All dental variables were assessed at six different sites (mesiobuccal, midbuccal, distobuccal, mesiopalatal, midpalatal and distopalatal) of each tooth present, excluding third molars. The periodontal assessments were carried out at the baseline visit and at a recall visit after 6 weeks. Intra-examiner calibration for reproducibility of probing depth, clinical attachment loss, bleeding on probing score and plaque score was performed prior to the study. Clinical measurements of periodontal parameters included: (i) plaque score,[28] where the surfaces with visible plaque were calculated and scored as percentage; (ii) bleeding on probing score,[1] where the surfaces with bleeding after 10 s of probing with an electronic constant-force probe (Florida Probe; Gainsville, FL, USA) were calculated and scored as percentage; (iii) probing depth (PD) was measured as the distance from the gingival margin to the location of the tip of the constant-force periodontal probe inserted into the periodontal pocket with a force of 15 g; (iv) clinical attachment level (CAL) was measured as the distance from the cementoenamel junction to the location of the inserted probe tip. Loss of periodontal tissue support was assessed by measurement of pocket depth and gingival recession using the electronic constant-force probe.

After the periodontal measurements were taken, the subjects were diagnosed as: healthy periodontium (control group) and diseased periodontium (test group). The diseased periodontium group included those subjects with gingivitis and chronic periodontitis. The diagnosis was based on the clinical criteria stated and described in the 1999 Consensus Classification of Periodontal Diseases[3] as healthy periodontium when bleeding on probing was $\leq 25\%$ and there were no sites of attachment loss, or diseased periodontium when bleeding on probing was $\geq 25\%$ with no

Table 1 Sociodemographic characteristics of the study sample

Variables		Healthy periodontium (control group) (n = 28)	Diseased periodontium (test group) (n = 28)	p
Age (years) (mean ± SD)		28.25 ± 3.15	29.86 ± 4.31	0.12[a]
Gestational age (week) at baseline (mean ± SD)		18.75 ± 2.18	19.75 ± 2.35	0.12[b]
Gestational age (week) at 6 weeks (mean ± SD)		24.96 ± 2.09	25.78 ± 2.18	0.12[b]
Primigravida	Yes n (%)	12 (43%)	14 (50%)	
	No n (%)	16 (47%)	14 (50%)	
Ethnicity				
Malay		26 (92%)	24 (85%)	
Chinese		1 (4%)	2 (7%)	
Indian		0 (0%)	1 (4%)	
Others		1 (4%)	1 (4%)	
Educational level				
Secondary		5 (18%)	12 (43%)	0.04[c]
Tertiary		23 (82%)	16 (57%)	
Frequency of toothbrushing				
Twice daily		12 (43%)	17 (61%)	0.18[c]
≥ 2 times daily		16 (57%)	11 (39%)	
Mouthwash usage				
Yes		19 (68%)	16 (57%)	0.41[c]
No		9 (32%)	12 (43%)	

[a]Independent t-test; [b]Mann-Whitney test; [c]chi-squared test.

sites of attachment loss (gingivitis), as well as when bleeding on probing was ≥ 25% with the presence of at least 5 teeth with PD ≥ 4 mm and with CAL ≥ 2 mm at the same site (chronic periodontitis).

Systemic Parameters

Three ml of venous blood was collected from each patient at baseline prior to treatment and at the recall visit 6 weeks post-treatment. The venous blood was then assessed for CRP using the immunoturbidimetry assay test for high sensitivity CRP (hs-CRP). All blood investigations were done by a private laboratory.

Clinical Intervention

Motivation and oral hygiene education (test and control groups): All patients first received an explanation of their periodontal conditions. Information on the association of periodontal disease and the pregnancy outcomes was explained to the patients. Later, patients were shown how to improve their oral hygiene technique. To standardise oral hygiene, all patients were given the same soft bristle toothbrush, toothpaste and dental floss, as well as standardised orgal hygiene instructions.

Scaling and root planing (SRP) (test group only): SRP was carried out in a single visit for all subjects in the test group. Supragingival and subgingival scaling was done using ultrasonic scalers. Root planing was done at PD ≥ 5 mm using Gracey curettes. Then, full-mouth polishing was done using a rubber cup and prophylaxis paste. All patients were instructed to rinse three times daily using 15 ml of 0.12% chlorhexidine mouthrinse for 14 days.

All patients were contacted by phone, and the data on gestational week of delivery and birth weight were self-reported by the subjects after their delivery and discharge. The self-reported pregnancy outcome data was then confirmed with their record in the Antenatal clinic, UMMC.

Statistical Analysis

The intra-examiner reproducibility was assessed by carrying out clinical periodontal parameter data collection on four subjects. Each subject was assessed twice in one visit, over a 2-h interval. The data collected were analysed for reproducibility using the intra-class correlation coefficient (ICC) for PD and CAL, and kappa analysis for bleeding and plaque score. The reproducibility agreement for PD was 0.84, 0.85 for CAL, 0.85 for bleeding score and 0.88 for plaque score. In accordance with Landis and Koch,[20] the following ICC interpretation scale and kappa scale was used: poor to fair (below 0.4), moderate (0.41–0.60), excellent (0.61–0.80), and almost perfect (0.81–1).

Table 2 Plaque score (PS) and bleeding on probing (BOP) score, mean probing depth (PD) and mean clinical attachment level (CAL) at baseline and at 6 weeks (follow-up visit) for the control and test groups

Variables	Healthy periodontium (control group) (n = 28)	Diseased periodontium (test group) (n = 28)	p
Plaque score (PS) % (mean ± SD)			
Baseline	21.04 ± 7.93 95% CI [17.96, 24.11]	45.25 ± 8.12 95% CI [42.10, 48.40]	0.000[b]
6 weeks	11.39 ± 3.85 95% CI [9.89, 12.89]	17.50 ± 5.98 95% CI [15.18, 19.82]	0.000[b]
Difference (baseline – 6 weeks)	9.65%	27.75%	
p (intragroup)	0.000[c]	0.000[c]	
Bleeding on probing score (BOP) % (mean ± SD)			
Baseline	11.71 ± 4.63 95% CI [9.92, 13.51]	47.12 ± 7.27 95% CI [44.29, 49.93]	0.000[b]
6 weeks	5.86 ± 3.12 95% CI [4.65, 7.07]	13.36 ± 5.40 95% CI [11.26, 15.45]	0.000[a]
Difference (baseline – 6 weeks)	5.85%	33.76%	
p (intragroup)	0.000[d]	0.000[d]	
Probing depth (PD) % (mean ± SD)			
Baseline	1.80 ± 0.27 95% CI [1.69, 1.90]	2.91 ± 0.23 95% CI [2.82, 3.00]	0.000[b]
Sites with PD 1-3 mm (%)	99.6%	72.6%	
Sites with PD 4-5 mm (%)	0.4%	23.1%	
Sites with PD 6-7 mm (%)	0%	4.3%	
6 weeks	1.66 ± 0.26 95% CI [1.56, 1.76]	2.06 ± 0.25 95% CI [1.97, 2.16]	0.000[b]
Sites with PD 1-3 mm (%)	99.8%	95.4%	
Sites with PD 4-5 mm (%)	0.2%	4.6%	
Sites with PD 6-7 mm (%)	0%	0%	
Difference (baseline – 6 weeks)	0.14	0.85	
p (intragroup)	0.000[d]	0.000[d]	
Clinical attachment level (CAL) % (mean ± SD)			
Baseline	0.12 ± 0.06 95% CI [0.09, 0.14]	0.37 ± 0.13 95% CI [0.31, 0.42]	0.000[b]
Sites with CAL 0 mm (%)	91.7%	79.4%	
Sites with CAL 1-2 mm (%)	8.3%	17.3%	
Sites with CAL 3-4 mm (%)	0%	2.9%	
Sites with CAL 5-6 mm (%)	0%	0.4%	
6 weeks	0.09 ± 0.02 95% CI [0.08, 0.10]	0.29 ± 0.07 95% CI [0.26, 0.31]	0.000[b]
Sites with CAL 0 mm (%)	93.1%	82.8%	
Sites with CAL 1-2 mm (%)	6.9%	15.5%	
Sites with CAL 3-4 mm (%)	0%	1.3%	
Sites with CAL 5-6 mm (%)	0%	0.2%	
Difference (baseline – 6 weeks)	0.03	0.08	
p (intragroup)	0.012[d]	0.008d	

[a]Independent t-test; [b]Mann-Whitney test; [c]paired t-test; [d]Wilcoxon Signed Ranks test.

SPSS version 12.0.1 was used to perform the data analyses. Statistical significance was set at $p \leq 0.05$. Descriptive statistics was used to describe the demographics data of the subjects, and numerical data analysis was used to describe the clinical parameters. The descriptive statistics and numerical data were expressed as % ± SD and means ± SD. The independent samples test, paired-sample test, Mann-Whitney test and Wilcoxon signed ranks test were used to analyse statistically significant differences between the test and control group, before and after the treatment. Effect size was calculated using Cohen's d formula.[10]

RESULTS

Sociodemographic Characteristics of the Study Sample

The average age of the subjects in the test and control group was 28.25 (± 3.15) years and 29.86 (± 4.31) years respectively. The mean gestational age at the baseline was 18.75 (± 2.18) weeks for the control group and 19.75 (± 2.35) weeks for the test group. The mean gestational age at the follow-up visit was 24.96 (± 2.09) weeks for the control group and 25.78 (± 2.18) for the test group (Table 1). The independent samples t-test and Mann-Whitney test confirmed that the mean age of the subjects and the mean gestational age at baseline and follow-up visit were similarly distributed between the test and control groups. The ethnicity distribution of both groups is given in Table 1. About 82% (n = 23) of the subjects in the control group and 57% (n = 16) of the subject in the test group had undergone tertiary education, which showed statistically significant differences ($p = 0.04$) between the groups (Table 1). A higher percentage of subjects in the control group brush their teeth more than twice daily (57% vs 39%) and a higher percentage of the subjects in the control group used mouthwash (Colgate Plax and Listerine) (68% vs 57%) when compared to test group. However, there were no statistically significant differences in the frequency of toothbrushing and the use of mouthwash between the groups.

Periodontal Disease Status of Subjects in Test Group

About 75% of the subjects in the test group were diagnosed as having chronic gingivitis, whereas 14% of the subjects had generalised early to moderate chronic periodontitis and 11% had localised moderate to advanced chronic periodontitis.

Periodontal Parameters

The baseline and 6-week follow-up plaque score (PS), bleeding on probing (BOP), mean PD and mean CAL for both groups are given in Table 2 and Fig 1. There were statistically significant differences for PS and BOP score between the control and test groups at baseline ($p = 0.000$; 0.000) and at follow-up ($p = 0.000$; 0.000). There were also statistically significant reductions in PS (9.65% reduction in the control group and 27.75% reduction in the test group), and BOP score (5.85% reduction in the control group and 33.76% reduction in the test group). Statistical analysis showed that there was a statistically significant difference between control and test groups for the mean PD at baseline and the follow-up visit. Furthermore, statistically significant intragroup reductions were found in the PD from baseline to the follow-up visit for both groups. A statistically significant difference was also observed between control and test groups for the mean CAL at baseline and follow-up visit. Both groups showed a statistically significant CAL gain from baseline to follow-up: 0.03 mm CAL gain in the control group and 0.08 mm CAL gain in the test group.

Systemic Parameters

The baseline and follow-up CRP levels for both groups are given in Table 3 and Fig 2, along with the mean gestational age at delivery and mean infant birth weight for both groups. There was a statistically significant difference between the two groups with a large effect size for this difference ($d = 0.7$) at baseline, whereas there was no statistically significant difference between the groups at the follow-up visit. However, there was an intermediate effect size between the groups ($d = 0.41$). There were statistically significant reductions of CRP levels for both groups. The reduction in control group was 0.99 mg/l with an intermediate effect size ($d = 0.3$). The reduction in the test group was greater, with a 2.06-mg/l reduction and intermediate effect size ($d = 0.4$). Mean gestational age at delivery was 38.96 weeks for the control group and 37.47 weeks for the test group. In the test group, 85.7% (n = 24) of the subjects delivered at > 37 weeks and 14.3% (n = 4) of the subjects delivered at < 37 weeks. All subjects in the control group delivered their baby at > 37 weeks of gestational age. There was a statistically non-significant difference in the mean gestational age at delivery between groups. Mean infant birth weight was 3.09 kg for the control group and 3.03 kg for the test group. All babies in the control group weighed ≥ 2.5 kg at delivery, whereas only 89.3% (n = 25) of the babies in the test group weighed ≥ 2.5 kg and 10.7% (n = 3) weighed < 2.5 kg. However, there was no statistically significant difference between groups.

DISCUSSION

Numerous studies have revealed a positive association between periodontal disease and circulating CRP levels,[26] while a meta-analysis confirmed that plasma CRP is elevated in patients with periodontitis compared to healthy individuals.[32] Periodontal disease, especially chronic periodontitis, is an inflammatory disease caused by bacterial biofilm, and leads to the destruction of the attachment apparatus of the teeth. Pregnancy and other systemic conditions can worsen the severity of periodontal disease.[22] The high percentage of subjects with periodontal disease observed in this study may be attributed to the ethnicity of the subjects, socioeconomic status and to the altered immune response due to hormonal imbalance which occurs in pregnancy.

Being an inflammatory reaction to a microbial infection, the host produces inflammatory mediators such as cyto-

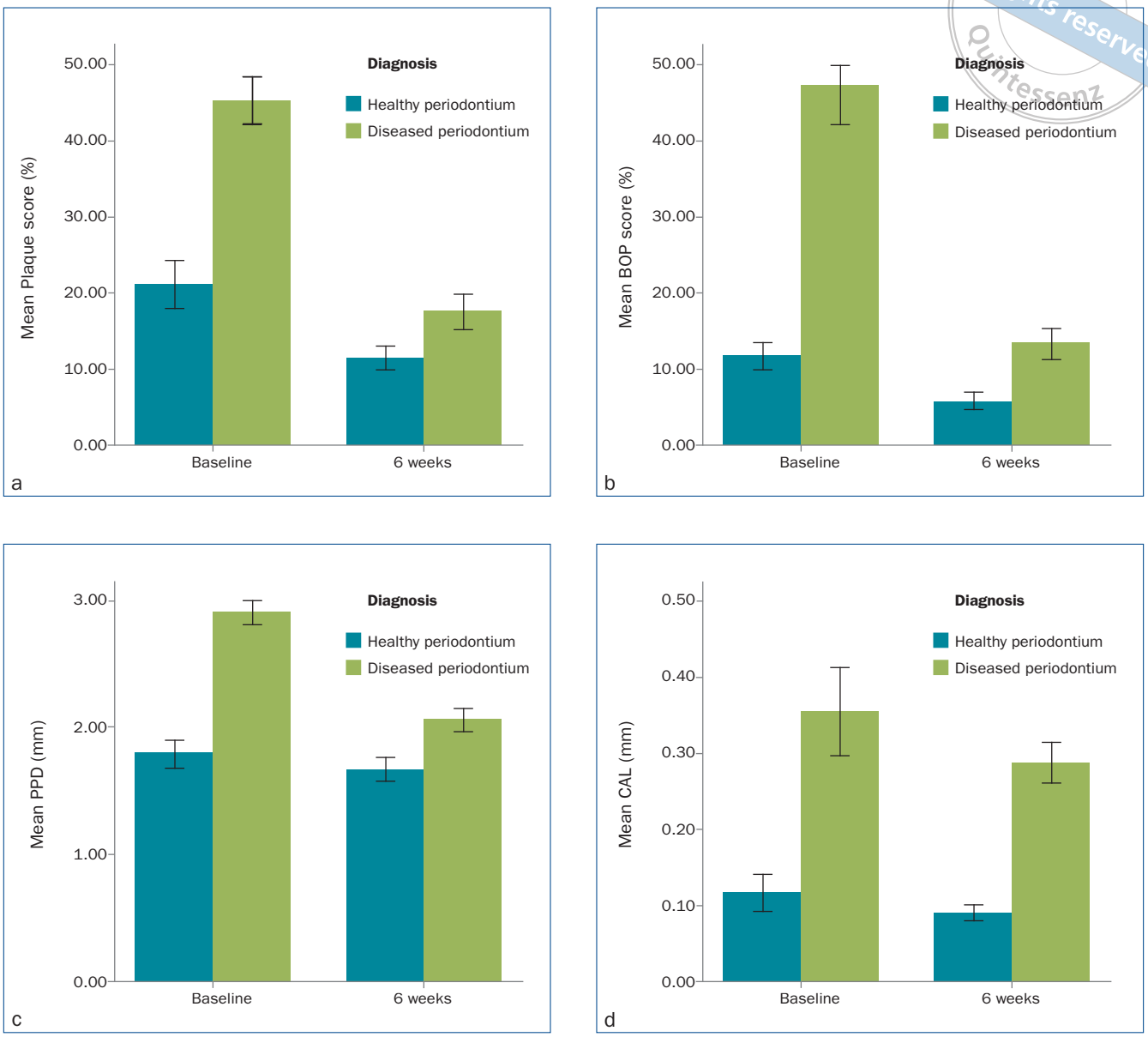

Fig 1 a. Mean plaque score (PS); b. mean bleeding on probing (BOP) score; c. mean probing pocket depth (PD); d. mean clinical attachment level (CAL) at baseline and 6 weeks between the healthy periodontium (control group) and diseased periodontium (test group).

kines and plasma CRP. Studies have shown that CRP produced by patients with periodontal disease, e.g. periodontitis, has an adverse effect in pregnant women, with outcomes such as low birth weight and preterm delivery. Periodontal disease during pregnancy is also associated with an increase in CRP.[29,33] Plasma CRP is a nonspecific marker of inflammation and is known to be elevated in both in persons with periodontal disease[19,26,27] and also during normal pregnancies.[6,21,35] This result conforms to other studies on plasma CRP levels and periodontal disease during pregnancy.[15,23,33,34,39,43] Possibly, several underlying pathogenic mechanisms may play a role in elevated plasma CRP levels in pregnant women with periodontal diseases. One such mechanism is that the periodontal pathogens not only induce local inflammation and tissue destruction, but they are also involved in systemic increases in inflammatory and immune responses.[5] Low levels of bacteraemia, LPS and other bacterial components may provide the stimulus for systemic inflammatory responses such as increased production of CRP, due to activation of the cascade of inflammatory cytokines production by monocytes and other cells in periodontal tissues.[27]

This prospective interventional study demonstrated elevated plasma CRP levels in pregnant women with periodontal disease compared to pregnant women with healthy periodontium (Table 3 and Fig 2). This study further

Table 3 CRP levels at baseline and at 6 weeks (follow-up visit) for the control and test groups

Variables	Healthy periodontium (control group) (n = 28)	Diseased periodontium (test group) (n = 28)	p	Effect size [d]
CRP mg/l (mean ± SD)				
Baseline	5.66 ± 2.91 95% CI [4.53, 6.78]	8.55 ± 5.28 95% CI [6.51, 10.60]	0.032[b]	0.7
6 weeks	4.67 ± 3.43 95% CI [3.34, 6.00]	6.49 ± 5.32 95% CI [4.43, 8.56]	0.198[b]	0.41
Difference (Baseline – 6 weeks)	0.99	2.06		
p value (intra group)	0.042d	0.000d		
Effect size d	0.3	0.4		
Gestational age at delivery (week)				
Mean ± SD	38.96 ± 1.23 95% CI [38.9, 39.44]	37.46 ± 7.10 95% CI [34.71, 40.21]	0.637[b]	
≥ 37 weeks (%)	100%	85.7% (n = 24)	0.111[e]	
< 37 weeks (%)	0%	14.3% (n = 4)		
Birth weight (kg)				
Mean ± SD	3.09 ± 0.35 95% CI [2.95, 3.22]	3.03 ± 0.39 95% CI [2.88, 3.18]	0.774[b]	
≥ 2.5 kg (%)	100%	89.3% (n = 25)	0.236[e]	
< 2.5 kg (%)	0%	10.7% (n = 3)		

[b]Mann-Whitney test; [d]Wilcoxon Signed Ranks Test; [e]Fisher's Exact test.

demonstrated decreased plasma CRP levels in pregnant women with diseased periodontium after nonsurgical periodontal treatment when compared to the pre-treatment status (Table 3 and Fig 3). This confirms other randomised clinical trials and systematic reviews on nonsurgical periodontal treatment.[18,40] In this study, all clinical parameters (PS, BOP, PD and CAL) (Table 2 and Fig 2) improved in test group after oral hygiene instruction and nonsurgical treatment, which concurred with other studies.[18,40] However, in the control group, only oral hygiene instructions were given, which nevertheless led to reduction in plasma CRP levels (Table 3 and Fig 2) and improvement in all clinical parameters (PS, BOP, PD and CAL) (Table 2 and Fig 1). The patients in the control group might have had subclinical periodontal lesions at baseline – i.e. the initial lesion that can be observed histologically and is characterised by oedema, an increase in gingival fluid flow, and accumulation of polymorpholeukocytes and loss of connective tissue.[31] Moreover, the Hawthorne effect[17] and the frequent examinations carried out in studies could be a contributory reason for the improvement of all clinical parameters in the control group.

Moreover, this study evaluated the pregnancy outcomes (gestational age at delivery and baby's birth weight) in both the test and control groups, but failed to show any statistically significant differences between the groups. Many studies have shown that periodontal treatment before the 28th week of pregnancy (during the second trimester) may reduce the risk of PTB and LBW.[36] However, other studies reported statistically significant improvement in periodontal health but little effect or no effect on adverse pregnancy outcome.[25,47] Nonsurgical periodontal treatment causes bacteremia and a temporary surge in systemic inflammatory response,[9] causing cytokines and bacteria to flow into the systemic circulation. This surge in systemic inflammatory response may negate any positive effects on pregnancy outcomes otherwise achieved by nonsurgical periodontal treatment method over the long term.[24] Moreover, providing nonsurgical periodontal treatment during pregnancy may be too late to decrease local and systemic inflammatory responses that result in adverse pregnancy outcomes. Therefore, once the inflammatory cascade has started, interventions focusing on inflammatory pathways may have no effect in avoiding PTB.[48] Xiong et al[48] suggest targeting women with non-surgical periodontal treatment even before they conceive.

In this study, all subjects in the test group underwent nonsurgical periodontal treatment. Since there was no untreated group among the pregnant women with periodontal disease, the effect of periodontal treatment on the pregnancy outcomes was inconclusive. The data obtained on the pregnancy outcomes in this study showed no statistically significant difference in gestational age at delivery or infant birth weight between the test and control groups (Table 3). The results of pregnancy outcomes could not be considered the effect of the treatment given, because all subjects in the test group (diseased periodontium) were

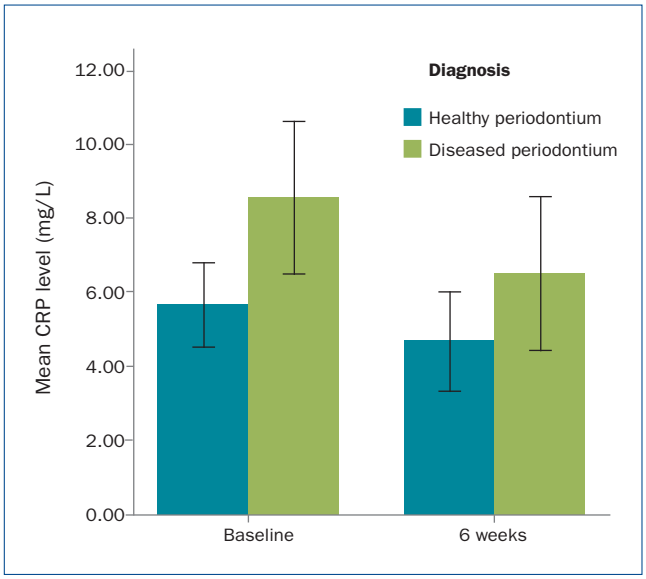

Fig 2 Mean CRP level at baseline and 6 weeks between healthy periodontium (control group) and diseased periodontium (test group).

given nonsurgical periodontal treatment. In order to evaluate the effect of nonsurgical periodontal therapy, the test group could have been divided into: a) treated and b) untreated subgroup. However, ethical regulations did not allow any antenatal subjects with periodontal disease not to receive treatment.

The association observed between CRP levels and periodontal disease in pregnancy may or may not have been causal. Periodontitis may increase CRP levels in pregnancy. CRP could amplify the inflammatory response through complement activation, tissue damage and induction of inflammatory cytokines in monocytes, and therefore may mediate the association between periodontitis and adverse pregnancy outcomes. Periodontal treatment resulted in a decrease of CRP levels and incidence of preterm delivery and LBW infants.[12] Therefore, it raises the possibility that CRP may mediate the association between periodontal disease and adverse pregnancy outcomes, as suggested by previous studies.[33,39] Further studies on maternal and fetal immune response to chronic oral infection and on placental pathology in women with periodontal disease have to be done to determine whether the relationship between periodontal disease and adverse pregnancy outcome is causal or simply associative.

CONCLUSIONS

The plasma CRP levels were significantly elevated in pregnant women with periodontal disease compared to pregnant women with healthy periodontium. Moreover, plasma CRP levels in pregnant women with periodontal disease were significantly reduced after nonsurgical periodontal therapy with intensive oral hygiene instruction/education and motivation, improving the periodontal status of the subjects in both groups. However, there was no association between CRP levels and adverse pregnancy outcome. This may be due to the relatively small sample size and selection bias, as the selected, exclusively urban subjects may not be representative of the general population.

Therefore, it is recommended that multicentre studies of longer duration and larger sample populations with the involvement of the obstetrics personnel include subjects with other confounding factors for adverse pregnancy outcome. In this way, multiple regression analysis could be conducted to determine the strength of correlation of periodontal disease on adverse pregnancy outcome in comparison to other confounding factors. In addition, the test group could be further divided into a treatment test group and a no-treatment test group, as in the study conducted by Sharma et al.[39] The reason for this division would be to evaluate the effect of nonsurgical periodontal therapy on adverse pregnancy outcomes, particularly PTB and LBW infant. However, this would involve ethical issues, since subjects with periodontal disease in the no-treatment group would not receive proper periodontal therapy in period of the study.

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