Original Research Paper

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Effect of Non-Surgical Periodontal Therapy on Anemic Status In Chronic Periodontitis Patients: A Randomized Clinical Case Control Study.

Abstract:

Back Ground: The aim of this current study is to identify the changes in haematological values of subjects with periodontal disease as compared to periodontally healthy individuals and to establish the relationship between chronic periodontitis and anemic status of the patients by correlating the periodontal parameters with haematological values following Nonsurgical Periodontal Therapy.

Methodology: The present case-control study included hundred subjects, out of which test group included fifty chronic periodontitis. The control group included fifty periodontally healthy subjects. Both periodontal and haematological parameters were assessed for both the groups at baseline and three monthswho had undergone non-surgical periodontal therapy later for the test group. Ten patients in the test group were excluded from the study and the remaining forty patients completed the study period and hence their results were analysedstatistically by employing Mann-Whitney U Test, Wilcoxon Matched Pairs Test, Karl-Pearson's Correlation Coefficient Method.

Results: The results in this study have shown that by treating the periodontal inflammation, it has led to a significant improvement in erythrocyte count as well as haemoglobin levels. Following NSPT in the test group,out of all the haematological values, ESR has shownmajor statistically significant difference along with TRBC and Hb.Whereas there was much lesssignificant improvement in the remaining values viz. PCV, MCV, MCH, MCHC.

Conclusion: The results of this study have clearly demonstrated that the treatment of periodontitis condition does lead to the improvement in various haematological values. Hence, its proven that chronic generalized periodontitis could lead to anemia of chronic disease in the long run. Thereby it is advisable that we should monitor the anemic status of the patients on a regular basis and advice the patients accordingly.

Keywords: Anemia; Blood; Chronic Periodontitis; Erythrocyte Sedimentation Rate; Haemoglobin

Introduction:

Models of pathogenesis have been presented in which systemic disorders affecting neutrophil, monocyte and/or lymphocyte function result in altered production or activity of cytokines and inflammatory mediators. These changes in the host response to bacterial challenge may manifest clinically as localized or generalized increases in bone and attachment loss.[1]Periodontitis is an infectious disease resulting in inflammation within the supporting tissues of the teeth, progressive attachment and bone loss and is characterized by pocket formation and/or recession of the gingiva.[2] Localized infections characteristic of periodontitis can have a significant effect on the systemic health of humans and animals. Just as the periodontal tissues mount an immune inflammatory response to bacteria and their products, systemic challenges with these agents also induce a major vascular response.[3]

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The association between periodontitis and an increased risk for systemic diseases like atherosclerosis, cardiovascular diseases, preterm low birth weight of infants and cerebrovascular diseases indicates that periodontitis patients may have a subclinical systemic inflammatory reaction.[4,5]

Periodontitis may affect changes in cellular and molecular components of peripheral blood such as leukocytes, erythrocytes, thrombocytes, acute-phase proteins, immunoglobins and immune mediators. Locally produced proinflammatory immune mediators like interleukin-1(IL-1), interleukin-6 (IL-6), tumour necrosis factor- (TNF-) and prostaglandin E2(PGE2) are dumped into the systemic circulation and subsequently may exert effects on distant organ systems. As periodontal tissues mount an immune inflammatory response to bacteria and their products, the systemic challenge with these agents also induces a major vascular response. Such inflammatory cytokines can depress erythropoietin production leading to the development of anemia.[6]

Anemia of chronic disease (ACD) which is the second most prevalent after anemia caused by iron deficiency is a multifactorial anemia often coexistent with iron deficiency.[4,7] Anemia of Chronic Disease is the second most prevalent after anemia caused by iron deficiency.[7,8,9]. It is a mild- to- moderate anemia associated with chronic infections, chronic non-infectious inflammatory diseases etc., that are not due to marrow deficiencies or other diseases and occurring despite the presence of adequate iron stores and vitamins. It is a cytokine-mediated anemia and is a frequent complication of many chronic inflammatory conditions.[9]

The hallmark of anemia of chronic disease is the development of disturbances of iron homeostasis with increased uptake and retention of iron within cells of reticuloendothelial system. This leads to limited availability of iron for erythroid progenitor cells. Further, the proliferation and differentiation of erythroid precursors is impaired due to blunted response to erythropoietin.

Since anemia of chronic disease is immune driven, cytokines and cells of the reticuloendothelial system induce these changes resulting in erythropoietin deficiency, reduced life span of red cells contributing to the pathogenesis of anemia.[9], 10Often patients are unaware of the existence of such anemia and likely to go undiagnosed and do not consult physicians for treatment.

The prevalence and severity of chronic periodontitis is very high in India. It is estimated that more than eighty percent of adult Indians have some form of periodontitis, out of which about fifty percent are moderate to severe in nature. Therefore, it is quite likely that many of the periodontitis patients may either being suffering from undiagnosed anemia or have haematological values that are subnormal. Therefore, it is important that this association is evaluated in order that timely intervention can be made, thereby preventing the complications of anemia of chronic disease.

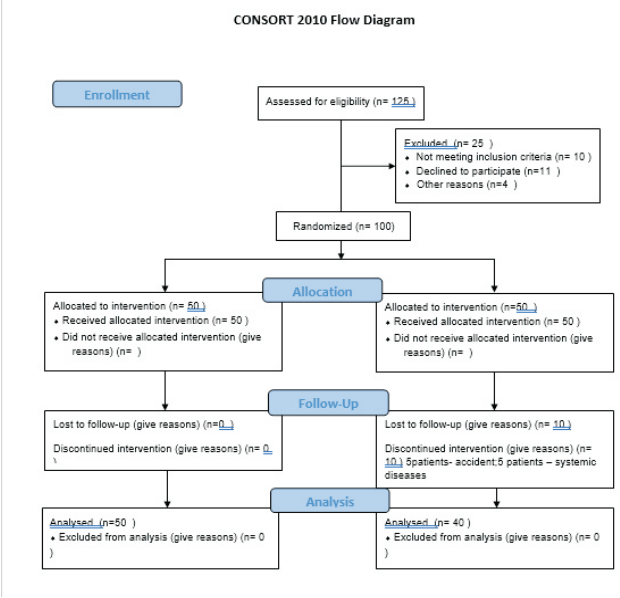
This study envisages not only to evaluate the haematological values in patients with chronic periodontitis but also assessing the benefit of non-surgical periodontal therapy in improving the stated values.

Materials and methods:

In these three months follow up of case-control study, a total number of 100 systemically healthy patients aged between 35 to 50 years of both sexes were recruited. 50 of the patients who were diagnosed with moderate to severe chronic periodontitis formed the test group whereas the remaining 50 who were free of any demonstrable periodontal disease served as controls. All these subjects were selected from those who attended the Department of Periodontics, Vishnu Dental College, Bhimavaramduring the period of 2012-[13]. Sample size was taken according to previous study done by Pradeep A R6. Institutional ethical committee has approved the study protocol and all patients signed an informed consent explaining all procedures involved in the protocol and the possible benefits associated with the proposed study. No

external funding, apart from the support of the authors' institution, was available for this study. Informed consent was taken from all the patients after explaining all the procedures involved in the protocol and the possible benefits/problems associated with the proposed study and study was performed according to Helsinki declaration 1975 which was modified in 2008. Study was performed according to CONSORT guidelines (Figure 01) wherever possible.

Figure 01: The Consort diagram showing the study design



Inclusion Criteria included aTest Groupin whichpatients with chronic periodontitis with minimum of 20 remaining teeth in the dentition showing clinical attachment level of ≥4mm in at least six teeth of dentition, confirmed by the radiographic evidence of bone loss. Control Groupin whichperiodontally healthy subjects with clinically normal tissues as evidenced by lack of bleeding upon probing, uniformed probing depth of 3mm or below, with no evidence of loss of attachment.

Exclusion Criteria were applied for both groups in which the patients with systemic diseases including anemia and also already diagnosed with any form of anemia, Smokers, alcoholics, pregnant women,female patients with irregular menstrual cycles, patients who underwent periodontal therapy or used antibiotics, or oral supplementations in the previous six months period prior enrolment,patients who are taking iron supplements and haematinics.

Clinical Parameters:

The gingivaland periodontal parameters were assessed by using Plaque Index11 ,Gingival Index11, Probing Pocket Depth (PD)3 Clinical Attachment Level (CAL)3. All assessments were carried out with a UNC-15 periodontal probe. PD and CAL measurements were obtained on six sites of tooth. Haematological parameterswere recordedunder strict aseptic conditions in which venous blood was drawn using 5ml syringe(Figure 02)from antecubital fossa and was subjected to theinvestigations which includedTotal RBC Count (TRBC), Haemoglobin Concentration (Hb), Erythrocyte Sedimentation Rate (ESR), Packed Cell Volume (PCV), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC).

Figure 02: Armamentarium

Primary and Secondary Outcomes:

PD,CAL,Hb, ESR, TRBC – primary outcomes

GI, PI,PCV,MCH,MCHC,MCV – secondary outcomes

Under Treatment Protocol all the patients in the test group were given oral hygiene instructions and educated and motivated for optimal plaque control. A meticulous phase-I therapy was performed which included thorough scaling and rootplaning, restoration of any carious teeth and extraction of tooth with any hopeless prognosis. Subjects were recalled at monthly intervals for supportive periodontal therapy and to reinforce oral hygiene. All the patients were instructed to inform immediately for any change in their medical status,

any severe injuries resulting in excess blood loss. All the periodontal and haematological parameters were again assessed at the end of three months.Patients in the control group were given oral hygiene instructions and oral prophylaxis was performed and did not have any further participation in the study.

Statistical analysis:

The results were evaluated statistically by employing Mann-Whitney U Test, Wilcoxon Matched Pairs Test, Karl-Pearson's Correlation Coefficient Method for assessing that inter and intra group relations and also to establish correlations between the periodontal and haematological parameters. A level of significance of 5% was assumed ($p < 0.05$). The statistical software namely SPSS 20.0, Stata8.0, MedCalc 9.0.1, Systat 12.0 were used for analysis of the data and Microsoft Word and Excel have been used to generate graphs and tables.

Results:

The present case-control study included 100 subjects, out of which 50 had chronic periodontitis who formed the test group. The remaining 50 who were periodontally healthy subjects formed the controls. Both periodontal and haematological parameters were assessed for both the groups at baseline and 3months later for the test group who had undergone non-surgical periodontal therapy. 10 patients out of which 5 patients had met with accidents and lost substantial blood/ or had to undergo surgeries and 5 patients who developed systemic diseases requiring medications including haematinics were excluded from the study. The remaining 40 patients completed the study period and their results were analysed.

Demographic variables:

Age: The age of the patients ranged between 35-50 years with the mean age of 42.5years for control group and 44.5years for test group.

Gender:There were 29 males and 21 females in control group and 25 males and 15 females in test group respectively. (Table: 01)

Table 01 : Distribution of male and females in control and test groups

Sex	Control group	%	Test group	%
Male	29	58.00	25	62.50
Female	21	42.00	15	37.50
Total	50	100.00	40	100.00

Periodontal variables

Plaque index:

The mean plaque score at baseline in the control group was 0.09±0.10 and 1.17±0.47 in the test group. The mean plaque score reduced to 0.83 at 3months in the testgroup. This decrease in the mean plaque score was statistically significant($p<0.05$).

Gingival index:

The baseline mean gingival score in the control group was 0.31±0.18 and 1.16±0.50 in test group. The score in the latter group reduced to 0.90±0.45 at the end of 3 months. Thisreduction of mean gingival scores was statistically significant ($p<0.05$) in test group.

Probing depth:

The mean probing pocket depth at baseline was 3.62±0.49mm. At the end of 3 months, the mean probing depth reduced to 2.97±0.35mm. This reduction was statistically significant ($p<0.05$).(Figure 03,04)

Figure 03: Test Group: Preoperative left lateral view is shown where pocket depth of 5mm is seen on the distobuccal aspect of 36 by using CPITN probe at baseline.

Figure 04 : Test group:Three months postoperative left lateral view is shown where pocket depth of 2mm is seen on the distobuccal aspect of 36 by using CPITN probe.

Clinical attachment level:

The mean clinical attachment level which was 4.29±0.77mm at baseline reduced to 3.51±0.73mm at end of study period. This gain in clinical attachment level was statistically significant from baseline to 3months.

Haematological Variables:

The mean haemoglobin concentration at baseline was 13.35±1.64gm% in periodontally healthy group and 13.05±1.52gm% in periodontitis group. This difference was not statistically significant.At the end of3months following periodontal therapy, thehaemoglobin concentration increased to 13.50±1.53gm% in the test group and the difference between the baseline value and 3 months wasstatistically significant.

TRBC:

The mean total red blood cell count was 4.64±0.73millions/µlin the control group and 4.49±0.62millions/µl in test group at baseline. This difference in mean total red blood cell count was also not statistically significant. However, by the end of 3 months, the mean TRBC score increased to 4.69±0.53millions/µl and this was statistically significant.

ESR:

In the control group, the mean ESR was 5.63±2.60mm/hr. However, in the test group this count was 11.15±6.90mm/hrat baseline. This difference was statistically significant. By the end of 3 months following therapy, the ESR in periodontitis group reduced to 7.88±4.54mm/hr. This reduction in mean ESR score was statistically significant from baseline to 3months in test group.

PCV :

The mean PCV in the patients of control group at baseline was 43.20±5.83% and 42.68±4.37% in test group. This difference was not statistically significant. This increased to 43.43±4.27% at the end of 3months. This increase was also not statistically significant.

MCVI:

The mean MCV in the patients of control group at baseline was 92.80±6.63Fl and 94.33±6.91Fl in test group. This difference was not statistically significant. This reduced to 92.93±5.87Fl by the end of 3 months. This decrease was not statistically significant.

MCH:

The mean MCH score at baseline was 28.28±1.06pg and 28.45±0.50pg in test group. This difference was not statistically significant. By the end of 3months there was no significant change in the MCH score in the test group.

MCHC:

The mean MCHC score was 30.38±2.07% and 29.78±3.93% in control and test group respectively. This difference was not statistically significant. In test group, the mean MCHC score increased to 30.75±1.82% by the end of 3 months although this was not statistically significant.

Correlation Between Clinical andHaematological Parameters:

There was no statistically significant correlation found between clinical and haematological parameters at baseline with any of the values. However, at 3months, the decrease in the plaque index score was significantly correlated to the decrease in the ESR score (Table: 02)

TABLE 02 : Correlation between clinical and haematological parameters difference scores of baseline and 3 months by Karl Pearson's correlation coefficient method.

Parameters	PI	GI	PD	CAL
HB	r=0.1942	r=0.2195	r=0.3597*	r=0.2160
T RBC	r=0.3108	r=0.2967	r=0.0880	r=-0.0620
ESR	r=-0.2662	r=-0.2495	r=0.1917	r=0.4377*
PCV	r=0.1089	r=0.0523	r=0.1229	r=-0.0119
MCV	r=-0.0430	r=-0.1098	r=-0.1886	r=-0.1985
MCH	r=0.0950	r=0.0654	r=-0.0649	r=0.0027
MCHC	r=-0.0411	r=0.0069	r=0.1571	r=0.0772

*p<0.05

The correlation between clinical and haematological parameters at baseline and 3months was statistically significant for the decrease in the probing depth and increase in the haemoglobin concentration and gain in the clinical attachment and decrease in the ESR score.

Comparison of the Male and Female with Haematological Parameters:

In control group, when comparison was made betweenmale and female with mean haematological parameters. Males were found to have better values of haemoglobin concentration, TRBC, PCV and MCH than females and these differences were statistically significant. Nevertheless, there was no difference in the values of ESR,MCVand MCHC between the sexes. (Table: 03,04)

TABLE 03 : Comparison of male and female with haematological parameters scores in control group by t test.

Variable	Sex	n	Mean	SD	t-value	P-value
HB	Male	29	14.53	1.09	8.3464	0.00001*
	Female	21	11.91	0.84		
T RBC	Male	29	5.14	0.53	7.3421	0.00001*
	Female	21	4.03	0.39		
ESR	Male	29	5.33	2.23	-0.8060	0.4253
	Female	21	6.00	3.01		
PCV	Male	29	47.18	4.27	7.3229	0.00001*
	Female	21	38.33	3.12		
MCV	Male	29	91.32	6.41	-1.5942	0.1192
	Female	21	94.61	6.61		
MCH	Male	29	27.95	1.29	-2.2130	0.0330*
	Female	21	28.67	0.49		
MCHC	Male	29	30.45	2.06	0.2653	0.7922
	Female	21	30.28	2.14		

*p<0.05

TABLE-04: Comparison of male and female with haematological parameters scores at baseline and 3 months in test group by t test.

Variable	Sex	n	Mean		SD		t-value		P-value	
			Baseline	3months	Baseline	3months	Baseline	3months	Baseline	3months
HB	Male	25	13.76	14.30	1.30	1.34				
	Female	15	11.87	12.18	1.07	0.70	4.7329	5.6553	0.00001*	0.00001*
T RBC	Male	25	4.78	4.97	0.45	0.46				
	Female	15	4.01	4.23	0.55	0.25	4.7732	5.6386	0.00001*	0.00001*
ESR	Male	25	8.32	6.72	4.81	3.35				
	Female	15	15.87	9.80	7.40	5.63	-3.9188	-2.1765	0.0004*	0.0358*
PCV	Male	25	44.84	45.28	3.68	3.81				
	Female	15	39.07	40.33	2.76	3.09	5.2410	4.2511	0.00001*	0.0001*
MCV	Male	25	93.92	91.48	5.75	5.52				
	Female	15	95.00	95.33	8.70	5.80	-0.4734	-2.0964	0.6386	0.0428*
MCH	Male	25	28.44	28.36	0.51	0.49				
	Female	15	28.47	28.53	0.52	0.52	-0.1600	-1.0618	0.8737	0.2950
MCHC	Male	25	29.60	31.16	4.66	1.72				
	Female	15	30.07	30.07	2.37	1.83	-0.3593	1.8974	0.7214	0.0654

*p<0.05

Similarly, in the test group, males showed better values with reference to Hb,TRBC,ESR and PCV as compared to females and these differences being statistically significant. The same was reflected even at the end of 3months. (Table :05)

TABLE –05 : Comparison of baseline and 3 months with respect to ESR(mm/hr)& Hb(gm%) scores in test group by t test.

Variable	Time	Mean	Std.Dv.	Mean Diff.	SD Diff.	% of change	t-value	p-value
ESR (mm/hr)	Baseline	11.15	6.90					
	3months	7.88	4.54	3.28	6.37	29.37	3.2501	0.0024*
Hb(gm%)	Baseline	13.05	1.52					
	3 months	13.50	1.53	-0.45	0.78	-3.47	-3.6497	0.0008*

*p<0.05

Discussion:

Inflammation is the characteristic feature of a living organism and is vital for the maintenance of homeostasis. However, inflammation beyond the acceptable limits or when it is unresolved can cause widespread changes in the body often causing deleterious effects.[12] Inflammations have been known to trigger various events such as production of acute phase reactants, increased formation of low-density lipoproteins, haptoglobin, fibrinogen which could trigger atherosclerotic changes.[13,14] Persisting inflammation due to infection can also cause a profound impact on haemopoiesis and could result in changes in the blood picture as a whole.

Periodontitis is an infection caused by microorganisms, modified by the host response that has been known to trigger inflammatory/ immune activities resulting in the production of variousproinflammatory cytokines and other noxious elements. It is also hypothesized that it can cause disturbance in the haematological picture. Hence this study attempted to identify those changes in haematological values of subjects with periodontal disease as compared to periodontally healthy individuals and in addition to explore if upon the restoration of periodontal health reversed the aforesaid haematological changes.

All the patients who were part of the test group had moderate to severe periodontitis with a mean clinical attachment loss of 4.29mm. They exhibited probing depths of a mean of 3.62mm whereas the control group had no evidence of periodontal disease. All the subjects were apparently otherwise healthy. Hence it is reasonable to accept that changes in the blood picture could be attributed to periodontal disease. The results in this study showed that the haematological parameters of the periodontitis subjects were inferior to that of the control group. But other than ESR, none of the values showed any statistically significant difference. Similar results have been reported in these studies.15,16. Out of all the erythrocyte values major differences was seen withTRBC,Hb and ESR. Whereas other values PCV, MCV, MCH, MCHC did not show that appreciable difference. This is quite acceptable as it takes a long time an anemia of a severe nature to bring about appreciable changes in these values. In another recent study, it was found that there is reduction of some of the haematological parameters like RBCs, Haemoglobin, PCV of patients with chronic generalised severe periodontitis comparing with periodontally healthy individuals, where serum ferritin levels has been evaluated and the patients with iron deficiency anemia were excluded in this study.[17]

Erythrocyte sedimentation rate which was shown to be higher in the test group can alter due to various conditions and is no longer considered specific and clinicians do not attach much importance to its values presently. Whereas TRBC and Hb values are much more significant and it is cleared that in this study they were affected by periodontal inflammation although not a large extent. This could be due to the duration and extent of periodontal disease in mouth and also whether the disease was active or in a dormant state. As expected, males showed slightly better values than females in both control and test groups with regard to hemogram.In a recent case control study, it was shown that aggressive periodontitis condition also resulted in a decrease in a number of erythrocytes and levels of haemoglobin. This implies that aggressive periodontitis like other chronic conditions may lead to anemia. This anemia is mild to moderate, and inflammatory cytokines may also play a role in its pathogenesis.[18,19]

The periodontal disease seen in the test group was reversed to a great extent by conscientious NSPT and stress given to maintenance of meticulous oral hygiene. All the periodontal parameters improved to an acceptable level. None of the subjects did anything during the study period like change of diet and living style, any nutritional supplements. It is to be appreciated that the levels of Hb, TRBC, ESR showed statistically significant improvement following NSPT. Whereas there wasn't much difference in the remaining values viz. PCV, MCV, MCH, MCHC. Similar results were observed in the studies[20,21,22] However a surprising result of the study was that there was no correlation between the periodontal scores and haematological values except between plaque scores, probing depths and ESR values and to some extent between probing pocket depth and haematological values.[23,24] Previous standard studies have reported a strong and significant correlation between BOP and Hb level. Overall the results of these studies have clearly demonstrated that chronic periodontitis does lead to reduction in various haematological values which could lead to anemia of chronic disease in the long run.[25,26,27] Further, it also showed that periodontal therapy could help in improving the anemic status although not completely by itself.[28,29,30] Hence it is important that periodontists should monitor the anemic status of their patients on a regular basis and advice their patients accordingly.

One major lacunae in the study is not using the third generation equipment like the Florida probe for obtaining the values which could have demonstrated subtle differences while measuring small changes. Further the sample size also should have been higher since the difference in the values between two groups was small. Further longitudinal studies done without the above stated drawback can through more light into the association between chronic periodontitis and anemia.

Conclusion:

Patients with moderate to severe chronic periodontitis demonstrated lesser haematological values viz. Hb, TRBC, PCV, MCV, MCH and MCHC as compared to healthy subjects. However, the difference was not statistically significant.

Chronic periodontitis subjects showed higher ESR reading and were statistically significant. Following NSPT they showed significant improvement in the periodontal parameters and haematological values viz. HB, TRBC and significant reduction in the ESR. There was positive correlation between probing pocket depth, Hb and ESR levels. No correlation could be established between other periodontal and haematological values.