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Anemia and Periodontitis: An Enigma?

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Abstract:
Introduction: *Periodontitis is a chronic infectious as well as inflammatory condition that has been vastly associated with various systemic conditions. There has been a lot of conflicting data regarding the relationship between anemia and periodontitis. The objective of the present study is to investigate the relationship between anemia and periodontal disease.*
Materials And Methods: *A total of sixty subjects were taken from both sexes (age group 35–65 years; mean age 46 years) were included into three groups : group I – Healthy, group II- Gingivitis, group III- Periodontitis ; 20 subjects in each group. Periodontal parameters and orthopantamographs were taken for all the groups and then 5 mL venous blood samples were sent for heamatological investigations. Inter-group and intra-group comparisons were performed for all the assessed parameters.*
Results: *Comparison of hematocrit values using paired sample test showed that in Periodontitis and Healthy group ;Hematocrit values (Hb%,MCHC%) was lower in periodontitis compared to healthy (p= 0.039, 0.015) .Comparing the Periodontitis and Gingivitis group results showed Hematocrit values (MCHC%) was significantly lower (p=0.001) in periodontitis compared to gingivitis .Gingivitis and Healthy group values did not show any statistical significant difference except for ESR (p=0.010) values that were higher in gingivitis group . MCH and RBC values showed no statistical difference among the three groups.Comparing the Pearsons correlation values showed that in the Gingivitis group :Negative corelation between GI vs Hb % and GI vs RBC was seen. In the periodontitis Group :Negative correlation between CAL vs Hb% was noted.*
Conclusion: *The present study provides evidence that periodontits like other chronic infections , may tend towards anemia. For a more coherent picture on this topic further investigations with larger sample sizes should be conducted.*
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I. Introduction

Oral cavity is a mirror to various underlying disorders.Periodontal diseases is one of the commonest diseases affecting the oral cavity.Periodontal medicine has thrown light on the fact that periodontal diseases can affect underlying systemic conditions[1- 5] As the periodontal tissues mount an immune inflammatory response to bacteria and their products, the systemic challenge with these agents also induces a major vascular response. The bacteria and their products evoke an immunoinflammatory reaction in the host tissue. Although this process is intended to eliminate the microbial challenge, it often results in damage to the host tissue.The sulcular epithelium acts as a protective barrier and prevents entry of microorganisms and other irritants into the systemic circulation. The host-microbial interaction in periodontitis leads to ulceration of sulcular epithelium. The ulcerated epithelium acts as a portal of entry for the bacteria to enter the connective tissue and thus into the systemic circulation. Bacteremia has been observed in patients with periodontitis and has been directly related to the severity of inflammation[1,6].The subgingival microbiota in patients with periodontitis poses a significant and persistent Gram-negative bacterial challenge to the host. Acute-phase proteins, such as C-reactive protein, have been shown to be elevated in patients with periodontitis.[2-4] This suggests a possible influence of periodontitis on systemic status of an individual. Studies have associated periodontitis with atherosclerosis, cardiovascular diseases, and stroke.[5,6] These studies indicate that periodontitis leads to low-grade systemic inflammation.
However, the association of periodontitis with anemia has not been very clear so far. Anemia is defined as the deficiency in the oxygen carrying capacity of blood either due to reduced hemoglobin %(Hb%) or reduced Red Blood Corpuscles(RBC)count. Many studies have seen association of anemia in periodontitis patients and the possible etiology cited for decreased blood counts was the downregulation of erythropoiesis in the bone marrow

by proinflammatory cytokines released due to periodontal disease[6,7]This condition is termed as anemia of chronic disease (ACD) and is defined as the anemia occurring in chronic infections, inflammatory conditions or neoplastic disorders that is not caused by marrow deficiencies or other diseases, and occurring despite the presence of adequate iron stores and vitamins[8] It is a type of microcytic anemia.[9,10]H The present study was undertaken to investigate any association between periodontitis and anemia in healthy, gingivitis and periodontitis patients.

II. Materials And Methods

The study was conducted in the department of periodontics, M .R, Ambedkar dental college and hospital, Bangalore, Karnataka, India. A total of sixty subjects were taken from both sexes (age group 35–65 years; mean age 46 years) and were included into three groups : group I – Healthy, group II- Gingivitis, group III- Periodontitis ; 20 subjects in each group. The inclusion criteria included good general health without any systemic diseases or acute or chronic medical conditions including diabetes, viral, fungal or bacterial infections; and no history of periodontal therapy prior to 6 months of examination, use of vitamin or iron supplementation within the previous 3 months, any special dietary requirements (e.g. coeliac disease), or current usage of antibiotics or steroids.The exclusion criteria included pregnant and lactating mothers, smokers, patients with recent trauma or tooth extractions, any heamatological disorders and patients with the lack of desire to participate in the study. None of the patients were alcohol consumers.For the group I – Healthy subjects showing no clinical signs of gingivitis or periodontitis were taken ;group II- gingivitis included subjects showing clinical signs of gingivitis and group III –Periodontitis included subjects with clinical attachement loss that were assessed both clinically and radiographically . The group subjects were matched for age and socioeconomic status. All the procedures were fully explained to the patients before the study, and written informed consent was obtained from all the patients. The study protocol and consent form were approved by the Ethical Committee, Rajiv Gandhi University of Health Sciences, Karnataka, India, and the study was conducted on the basis of the principles outlined in the Declaration of Helsinki of 1975, as revised in 2008, on experimentation involving human subjects.

Examination of the patients was preceded by a thorough medical history of all the groups. A complete periodontal examination was undertaken using a mouth mirror and a UNC-15 graduated periodontal probe (Hu-Friedy, Chicago, IL, USA) by a single examiner (SP). Periodontal status was assessed by using clinical parameters such as Oral hygiene index,gingival index (GI), probing depth (PD) and clinical attachment level (CAL). Six sites per tooth (buccal, mesio-buccal, disto-buccal, lingual, mesio-lingual, and disto-lingual) were evaluated. PD was measured from the free gingival margin (GM) to the base of the pocket. CAL was determined by measuring the distance between the cemento-enamel junction (CEJ) and the GM, adding the PD at the same site. An orthopantamo-graph was taken for all the groups to assess the bone loss.

Hematological assessment

Blood samples (5 mL) were collected between 9.30 and 11.30 AM by venepuncture of the cubital vein in the antecubital fossa by using a 5 mL disposable syringe and a 23 gauge needle. A part of the blood sample was then transferred to sterile vacuum tubes containing an anticoagulant ethylene diamine tetraacetic acid (EDTA), for whole blood analysis. The remaining blood was collected in sterile vacuum tubes with no added anticoagulant and was kept at room temperature for 2 h, where it was allowed to clot, as this was designated for serum separation. The tubes were then transported to a laboratory (pathology department of ambedkar dental college and hospital, Karnataka, India) for processing within 4 h after venepuncture and were analyzed by using hematological investigations consisting of the RBC count, estimation of Hb, MCV, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and ESR, .

The hematological parameters like RBC count, Hb, MCV, MCH and MCHC were estimated in an automated blood counting machine (SYSMEX XE-2100; Sysmex Corporation, Kobe, Japan). ESR was estimated by using the Westergren method manually.

STATISTICAL ANALYSES

Statistical analysis was done using Pearsons correlation test to correlate the heamatological values with the gingivitis and periodontitis group and paired sample test was used to compare the heamatological values between the three groups. A p-value of .05 or less was considered statistically significant.

III. Results

Comparison of hematocrit values using paired sample test showed that in Periodontitis and Healthy group ;Hematocrit values (Hb%,MCHC%) was lower in periodontitis compared to healthy (p= 0.039, 0.015) shown in table 1 & 3 ;ESR and MCV was more in periodontitis (p= 0.008,0.0001)table 4&5 compared to healthy .Comparing the Periodontitis and Gingivitis group results showed Hematocrit values (MCHC%) was

significantly lower (p=0.001) in periodontitis compared to gingivitis as seen in table 3. MCV and ESR was higher in periodontitis group (p=0.012,0.047).Gingivitis and Healthy group values showed Hematocrit values between gingivitis and healthy did not show any statistical significant difference except for ESR (p=0.010) values that were higher in gingivitis group . MCH and RBC values showed no statistical difference among the three groups. Means of the heamatocrit values are shown in table 8.Comparing the Pearsons correlation values showed that in the Gingivitis group :Negative corelation between GI vs Hb %(graph 1);Negative correlation between GI vs RBC (graph 2)and Positive correlation between GI vs MCHC% (graph 3)was seen. In the Periodontitis Group :Negative correlation between CAL vs Hb% (graph 4) ;Negative correlation between CAL vs MCHC% (graph 5)and Positive correlation between CAL vs RBC (graph 6)was observed.(table 7)

TABLE No. 1
MEAN VALUES OF THE HAEMOGLOBIN LEVELS IN THREE GROUPS

PAIR	GROUPS	Mean±SD	Mean diff	P value
PAIR 1	PERIO-GING	12.10±1.94 12.80±1.24	0.70	0.180
PAIR 2	GINGI- HEALTH	12.80 ±1.24 13.20 ±0.83	0.40	0.214
PAIR 3	PERIO- HEALTH	12.10 ±1.94 13.20 ±0.83	1.4	0.039

TABLE 2 :MEAN VALUES OF THE RBC LEVELS IN THREE GROUPS

Pair	Group	Mean±SD	Mean Diff	P value
Pair 1	Perio Gingi	3.89± 0.98 4.310± 0.567	0.45	0.069
Pair 2	Gingi Health	4.310± 0.567 4.380± 0.449	0.07	0.701
Pair 3	Perio Health	3.89± 0.98 4.380± 0.449	0.49	0.056

TABLE 3 : MEAN VALUES OF MCHC% LEVELS IN THREE GROUPS

Pair	Group	mean± SD	Mean diff	P value
Pair 1	Perio Gingi	32.32± 1.57 34.70± 2.60	2.39	0.001
Pair 2	Perio Health	32.32± 1.57 34.75± 3.40	2.44	0.008
Pair 3	Gingi Health	34.70± 2.60 34.75± 3.43	0.05	0.963

TABLE NO 4.
MEAN VALUES OF ESR LEVELS IN THREE GROUPS

Pair	Group	Mean± SD	Mean Diff	P value
Pair 1	Perio Gingi	30.35± 23.51 16.60± 12.15	13.75	0.047
Pair2	Perio Health	30.35± 23.51 7.75± 3.65	22.60	0.001
Pair 3	Gingi Health	16.60± 12.15 7.75± 3.65	8.85	0.01

TABLE NO 5
MEAN VALUE OF MCV LEVELS IN THREE GROUPS

Pair	Group	mean ± SD	MePan diff	P value
Pair 1	Perio Gingi	92.80± 9.31 85.80± 6.28	6.28	0.012
Pair 2	Gingi Health	85.80± 6.28 85.35± 5.30	0.45	0.816
Pair 3	Perio Health	92.80± 9.31 85.35± 5.05	6.73	0.015

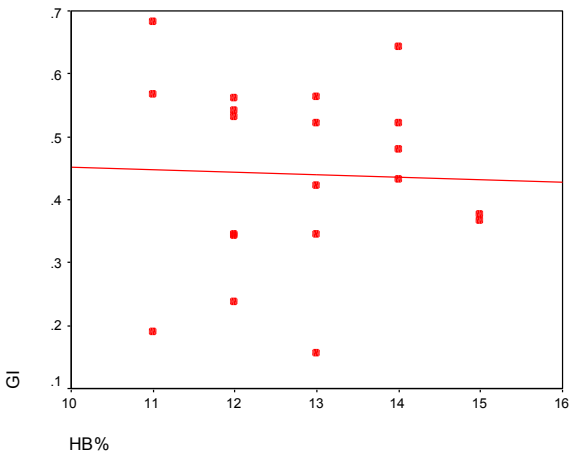
TABLE NO 6
MEAN VALUES OF THE MCH LEVELS IN THE THREE GROUPS

Pair	Group	mean± SD	Mean diff	P value
Pair 1	Perio Gingi	31.33 ±5.15 30.30 ±2.00	1.02	0.321
Pair 2	perio Health	31.33 ±5.15 29.95 ±1.36	1.38	0.289
Pair 3	Gingi Health	30.30 ±2 29.95 ±1.36	0.35	0.520

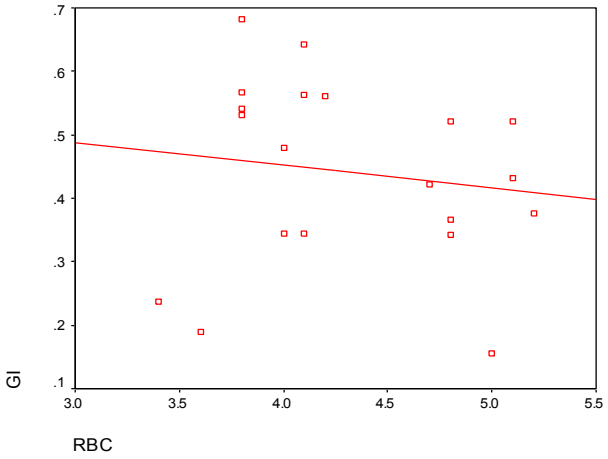
TABLE NO 7
CORRELATION VALUES BETWEEN PERIODONTAL PARAMETERS AND HEMATOCRIT LEVELS

Interactions	R value
GI vs Hb	- 0.033
GI vs RBC	- 0.138
GI vs MCHC	0.335
CAL vs RBC	0.221
CAL vs MCHC	- 0.241
CAL vs Hb	-0.025

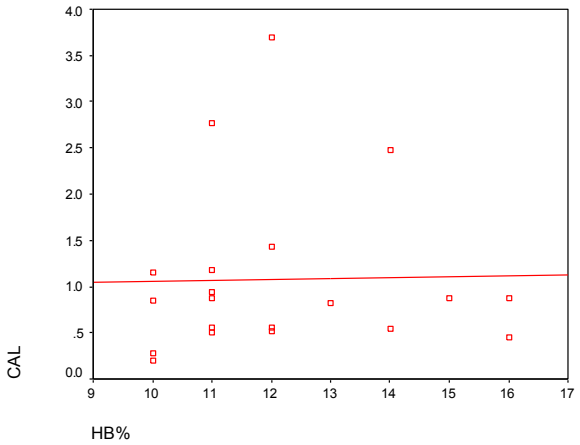
GRAPHS
GRAPH .1 CORRELATION BETWEEN GI VS Hb%



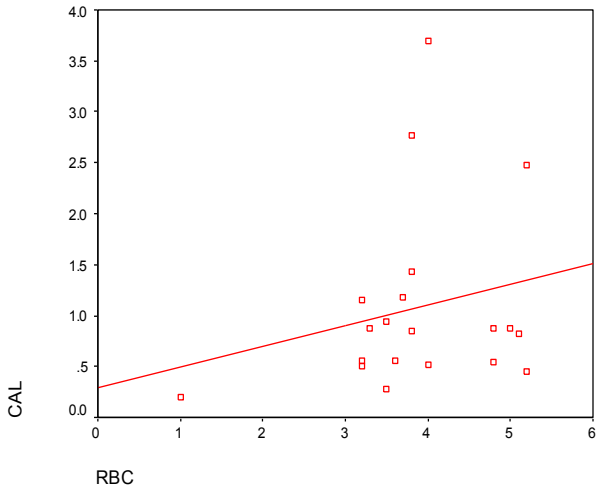
GRAPH 2. CORRELATION BETWEEN GI VS RBC



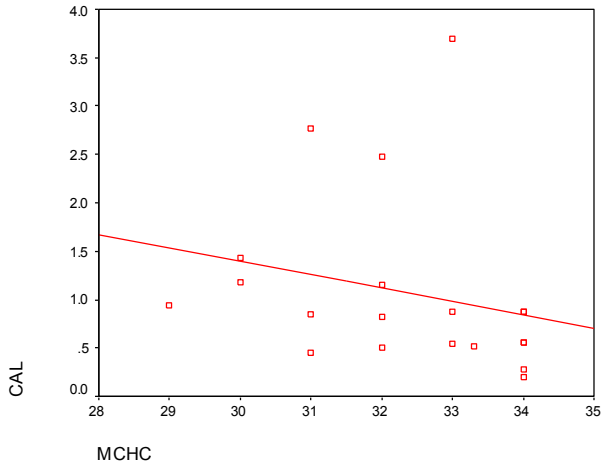
GRAPH 3.CORRELATION BETWEEN CALVS Hb%



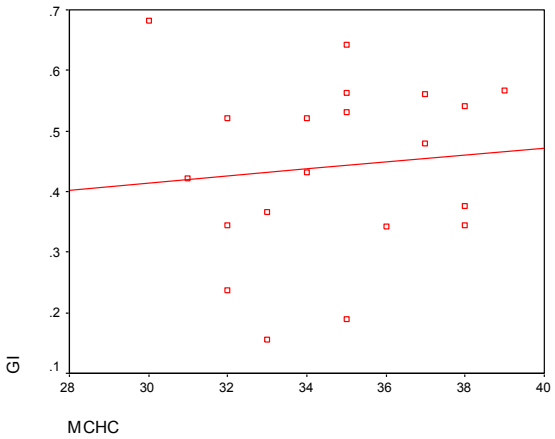
GRAPH 4. CORRELATION BETWEEN CAL VS RBC



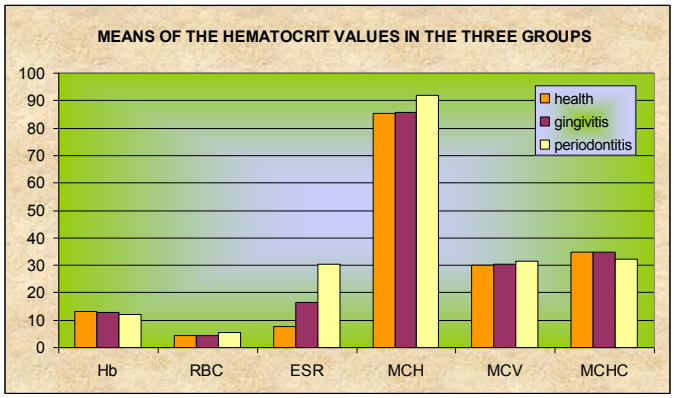
GRAPH 5.CORRELATION BETWEEN CAL VS MCHC%



GRAPH 6.CORRELATION BETWEEN GI VS MCHC



GRAPH 6. CORRELATION BETWEEN GI VS MCHC



GRAPH 7.MEANS OF THE HEMATOCRIT VALUES IN THE THREE GROUPS

IV. Discussion

Periodontal medicine has thrown light on the increasing association between periodontal disease and systemic conditions.[1,2,3,4,5]Periodontitis being a infectious as well as inflammatory disease is known to have systemic effects via bacteremia and is also known to show signs of systemic inflammation.[1,5] Previous studies have shown that periodontitis patients have elevated levels of WBCs and CRP[2,4 11,12]. These and many other pro inflammatory cytokines that are released as a host response to the predominantly gram negative bacteria in periodontitis that evoke a low grade systemic inflammation. Anemia has been reported in cases of rheumatoid arthritis.[13] Rheumatoid arthritis demonstrates a pattern of hard and soft tissue destruction caused by an

inflammatory process.[14] This process is similar to the inflammatory process seen in chronic periodontitis, although the etiologic factors of both diseases are different. Thus, long-standing chronic inflammation can lead to anemia. Similarly, chronic diseases such as bacterial, fungal, and parasitic infections have also been reported to show signs of anemia.

Current study states that periodontitis is also a chronic disease that can result in lower hemoglobin percent and is in accordance with Hutter et al study[7].Although lower hemaglobin levels in periodontitis patients was observed , the MCV and MCH values which is Hb per erythrocyte is comparable between three groups .It is important to note lower levels of Hb% in periodontitis is not due to iron or vitamin deficiencies since the MCV values are not altered [15.] Mild to moderate anemia has been reported as a manifestation of several chronic diseases, infections and neoplasms .Hence the term anemia of chronic disease (ACD) came into being where ACD is defined as the anemia occurring in chronic infections, inflammatory conditions or neoplastic disorders that is not caused by marrow deficiencies or other diseases, and occurring despite the presence of adequate iron stores and vitamins.[8,13] It is a type of microcytic anemia.[8] The Cause of ACD is multifactorial. Cartwright[16] postulated that at least three pathologic processes are involved in ACD: 1) shortened erythrocyte survival, 2) failure of the bone marrow to increase RBC production to compensate for this increased demand, and 3) impaired release of iron from the reticuloendothelial system. The proinflammatory cytokines are thought to act as mediators in suppressing erythropoiesis from the bone marrow leading to anemia. The cytokines interleukin (IL)-1a, IL-6, and tumor necrosis factor (TNF)-a have been related to suppression of erythropoiesis.TNF-a administration to animals by intermittent injections or implantation of TNF-a-producing cells resulted in development of anemia.[17]Johnson et al.[18]exposed mice to a single intravenous dose of TNF-a,which resulted in suppression of spleen and marrow erythroid colony-forming units. Johnson et al.[18]treated mice with single or repeated intraperitoneal injections of recombinant human IL-1a, which resulted in suppression of mature erythroid progenitors within 6 hours.Maury et al.[19] reported that recombinant human IL-1 (a and b) inhibited in vitro colony formation by erythroid burst-forming units and erythroid colony forming units from normal human marrow. In vitro colony formation by marrow granulocyte-macrophage progenitors was not inhibited by IL-1a. Erythropoietin is the hormone responsible for regulation of erythropoiesis. Faquin et al.[20]reported that IL-1 (a or b), TNF-a, and TGF-b inhibited production of erythropoietin from the hepatoma cell line Hep3B.The distinctive feature of ACD is low serum iron in the presence of adequate reticuloendothelial iron stores. Alvarez-Hernandez et al.[21] injected rats and mice with recombinant TNF-a and induced anemia and hypoferremia. The hypoferremia was associated with abnormalities of iron release from the reticuloendothelial system and incorporation into RBCs.

Periodontitis is also a chronic inflammatory disease that leads to elevation of the above mentioned cytokines. The severity of anemia may depend on the elevation of proinflammatory cytokines. Currently it is thought proinflammotry cytokines from a given chronic disease such as rheumatoid arithritis ,may down regulate the erythropoiesis in bonemarrow[13].Interleukin –1(IL –1) ,IL-6 and TNF-a have been implicated as cytokines suppersing erythropoiesis [13].All the above cytokines are known to be released as a result of periodontitis.

Our study also has also shown that there is negative correlation between GI vs Hb% as well as GI vs RBC. This is in accordance with the Hunter et al study and can be interpreted as follows that in the presence of gingivitis the Hb% and the RBC count reduces and this maybe due to the downregulation of erythropoesis by the proinflammatory cytokines[22]. The positive correlation seen in GI vs MCHC% can be because this value is more dependent on the iron stores in the body. In the periodontitis group negative correlation between the CAL vs Hb% and CAL vs MCHC% is seen and this can be interpreted as the downregulation of erythropoesis by the pro inflammatory cytokines released in periodontitis.[22](means et al). Positive correlation seen between CAL vs RBC count is not in accordance to Hunter et al study and this can can be due to the following reason that chronic inflammatory disorders are associated with an increased risk of patients developing anaemia. Also as previously discussed various studies have shown that cytokines released during cell-mediated immune responses are capable of inhibiting bone marrow haematopoiesis. . The mode of action of these cytokines is probably associated with their antiproliferative capacity. Decrease of serum iron and increase of storage iron in patients appears to be a consequence of the defense strategy of macrophages during long-lasting inflammatory disorders. Decreased serum iron correlates to decreased haemoglobin concentrations. In view of this, the development of anaemia seems likely to result from the altered iron metabolism induced by stimulated macrophages. Low haemoglobin levels and associated hypoxia up-regulate the release of erythropoietin, which can explain why increased circulating erythropoietin is usually found in patients with anaemia[23].(Fucsh et al). This inturn can lead to increase in RBC count which can be a possible explanation for the positive correlation seen in CAL vs RBC in our study.

The change in hemoglobin and RBC values in the present study is statistically significant but the difference is not as striking as observed in anemia caused by other inflammatory conditions, such as rheumatoid arthritis, neoplastic conditions, and fungal or parasitic infections. This may be explained by the fact that chronic

periodontitis is a milder inflammatory condition compared to other systemic infections or conditions. Limitaions of the present study are that we should have taken a more homogenous sample of probably only males since in India the females suffering from anemia due to various factors is very high. Second limitation is that we should have done an interventional trial instead of an observational study , by performing scaling on the samples and checking for their blood sample post therapy. This would have given a better picture on the relationship of anemia and periodontal disease.

Further studies need to be conducted to correlate the amount of periodontal inflammation and its effects on the systemic health of an individual. [11]However the RBC count remains unchanged between all the three groups . Also ESR provides a multifactorial measure of a systemic response to a disease .And in our study ESR was seen higher in periodontits group compared to ginigivitis or healthy group.However this parameter of inflamation seems to be of limited use as a diagonistic tool in periodontitis to measure systemic involvement.

V. Conclusion

The present study provides evidence that periodontits like other chronic infections , may tend towards anemia. However whether anemia is a cause or a consequence of periodontits is still not determined but there is an association.For a more coherent picture on this topic further investigations with larger sample sizes should be conducted.

References

[1]. Enhos S, Duran I, Erdem S, Buyukbas S. Relationship between iron-deficiency anemia and periodontal status in female patients. J Periodontol. 2009;80:1750–1755.
[2]. Mustapha IZ, Debrey S, Oladubu M, Ugarte R. Markers of systemic bacterial exposure in periodontal disease and cardiovascular disease risk: A systematic review and meta-analysis. J Periodontol. 2007;78:2289–2302.
[3]. Sim SJ, Kim HD, Moon JY, Zavras AI, Zdanowicz J, Jang SJ, Jin BH, Bae KH, Paik DI, Douglass CW. Periodontitis and the risk for non-fatal stroke in Korean adults. J Periodontol. 2008;79:1652–1658.
[4]. Wimmer G, Pihlstrom BL. A critical assessment of adverse pregnancy outcome and periodontal disease. J Clin Periodontol. 2008;35(Suppl. 8):380–397.
[5]. Mealey BL, Rose LF. Diabetes mellitus and inflammatory periodontal diseases. Compend Contin Educ Dent. 2008;29:402–408. 410, 412–413.
[6]. Gokhale SR, Sumanth S, Padhye AM. Evaluation of blood parameters in patients with chronic periodontitis for signs of anemia. J Periodontol. 2010;81:1202–1206.
[7]. Hutter JW, Van der Velden U, Varoufaki A, Huffels RA, Hoek FJ, Loos BG. Lower numbers of erythrocytes and lower levels of hemoglobin in periodontitis patients compared to control subjects. J Clin Periodontol. 2001;28:930–936.
[8]. Lee GR. The anemia of chronic disease. Semin Hematol. 1983;20:61–80.
[9]. De Benoist B, McLean E, Egli I, Cogswell M. Worldwide prevalence of anemia 1993–2005: WHO global database on anemia. Geneva: World Health Organization Press; 2008. p. 7.
[10]. Johnson MA. Iron: nutrition monitoring and nutrition status assessment. J Nutr. 1990;120:1486–1491.
[11]. Wakai K, Kawamura T, Umemura O, Hara Y, Machida J, Anno T, Ichihara Y, Mizuno Y, Tamakoshi A, Lin Y, Nakayama T, Ohno Y. Associations of medical status and physical fitness with periodontal disease. J Clin Periodontol. 1999;26:664–672.
[12]. Loos BG. Systemic markers of inflammation in periodontitis. J Periodontol. 2005;76:2106–2115.
[13]. Vreugdenhil G, Baltus CAM, van Eijk HG, Swaak AJ.Prediction and evaluation of the effect of iron treatment in anaemic RA patients. Clin Rheumatol 1989;8:352-362.
[14]. Mercado F, Marshall RI, Klestov AC, Bartold PM. Is there a relationship between rheumatoid arthritis and periodontal disease? J Clin Periodontol 2000;27:267-272.
[15]. Samson.D. The anemia of chronic disorders. *Post grad med J* 1983;59:543-550.
[16]. Cartwright GE. The anemia of chronic disorders.Semin Hematol 1966;3:351-375.
[17]. Johnson RA, Waddelow TA, Caro J, Oliff A, RoodmanGD. Chronic exposure to tumor necrosis factor in vivo preferentially inhibits erythropoiesis in nude mice.Blood 1989;74:130-138.
[18]. Johnson CS, Chang M-J, Furmanski P. In vivo hematopoietic effects of tumor necrosis factor-a in normal and leukemic mice: Characterization and therapeutic applications. Blood 1988;72:1875-1883.
[19]. Maury CP, Andersson LC, Teppo AM, Partanen S, Juvonen E. Mechanism of the anaemia in rheumatoid arthritis: Demonstration of raised interleukin- Iß concentrations in anaemic patients and of interleukin 1 mediated suppression of normal erythropoiesis and proliferation of human erythroleukemia (HEL) cells in vitro. Ann Rheum Dis 1988;47:972-978.
[20]. Faquin WC, Schneider T production J, Goldberg MA. Effect of inflammatory cytokines on hypoxia-induced erythropoietin. Blood 1992;79:1987-1994
[21]. Alvarez-Herna´ndez X, Lice´aga J, McKay IC, Brock JH.Induction of hypoferremia and modulation of macrophage iron metabolism by tumor necrosis factor. LabInvest 1989;61:319-322.
[22]. Means RT Jr., Krantz SB. Progress in understanding the pathogenesis of the anemia of chronic disease. Blood 1992;80:1639-1647. Review.
[23]. Fuchs D, Hausen A, Reibnegger G, et al. Immuneactivation and the anaemia associated with chronic inflammatory disorders. Eur J Haematol 1991;46:65-70.