

A CLINICAL STUDY TO COMPARE VARIOUS BLOOD CELL PARAMETERS FOR ASSESSMENT OF ANAEMIA OF CHRONIC DISEASE IN HEALTHY SUBJECTS AND CHRONIC PERIODONTITIS PATIENTS

ABSTRACT

Periodontitis is an inflammatory disease of the supporting tissues of the tooth. An immune response to bacteria and their products induces a major vascular response, offering explanatory mechanisms for the interactions between periodontal infection and a variety of systemic disorders. Therefore, periodontitis results in low-grade systemic inflammation, may cause decreased erythropoiesis and lower haemoglobin concentration.

Aims: This study is aimed at the assessment of various blood cell parameters in healthy subjects and those with chronic periodontitis

Methods and Material: 100 patients with severe chronic periodontitis (test group) and 100 periodontally healthy subjects (control group) in the age group of 20-60 years participated in the study. The blood sample obtained by venopuncture from antecubital fossa, was taken in anticoagulant coated vacuum tubes. Measurement of all blood cell parameters was done.

Statistical analysis used: Mann Whitney U test and Pearson chi square test were used.

Key-words: anaemia, chronic periodontitis, systemic inflammation

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INTRODUCTION

Periodontitis is an inflammatory disease of the supporting tissues of the tooth caused by specific microorganisms in a susceptible host.^[1] Studies have associated periodontitis with various systemic conditions, indicating that periodontitis leads to low-grade systemic inflammation.^[3]

Anaemia of chronic disease (ACD), is the anaemia occurring in chronic infections that is not due to dysfunction of bone marrow cells or other diseases, and occurring despite the presence of adequate iron stores and vitamins.^[4] It is due to the increased production of cytokines that mediate the immune or inflammatory response, which are also released

by periodontal tissues in response to bacterial infection, suggesting that periodontitis may cause ACD. However, conflicting results have been reported regarding the association between the two.^[5,6]

The present clinical study is undertaken to compare the various blood cell parameters in healthy subjects and those with chronic periodontitis.

MATERIALS AND METHODS:

A total of 200 systemically healthy patients visiting the out patient department of Sri Guru Ram Das Institute of Dental Sciences and Research, Amritsar participated in the study. The following selection criteria were applied to the

Table 1. Demographic and characteristics of two study groups

	Males	Females	p value
n(200)	68	132	0.073
Test (100)	28	72	
Control (100)	40	60	

Table 2. Mean Gingival Index and Mean Plaque Index values of two study groups

	Test (100)	Control (100)	z value	p value
Mean GI	2.37±0.48	1.21±0.49	1.16	<0.05
Mean PI	2.25±0.43	1.35±0.49	1.5	<0.05

Table 3. Reference ranges, mean values, p values of all the blood parameters in both the study groups.

	Range	Test	Control	p value
Hb (mg/dl)	M: 13-18 F: 11.5-16.5	12.02±1.01	13.4±1.5	<0.05
RBC (million/mm ³)	M: 4.74-5.49 F: 4.14-4.79	4.5±0.19	4.6±0.41	<0.05
MCV (n)	80-100	83.4±1.5	85.4±3.8	0.43
MCH (mg/dl)	32-38	30.4±1.2	33.1±1.6	0.06
MCHC (%)	37-49	35.4±1.6	36.2±1.5	<0.05
TLC (million/mm ³)	4000-11000	9582.09±648.6	8950.3±859.4	<0.05
N (%)	50-70	70.14±1.3	69.91±2.8	<0.05
E (%)	1-6	2.73±0.6	2.37±0.5	0.43
L (%)	30-40	38.15±2.7	36.67±2.4	<0.05
M (%)	2-10	1.84±0.7	1.81±0.67	0.2
BT (min)	1-5	2.15±0.5	2.25±0.38	0.81
CT (min)	2-7	4.35±0.4	4.31±0.4	0.57

patients:

Inclusion Criteria

1. Patients aged 20-60 years of age.
2. Patients diagnosed with chronic periodontitis i.e. probing depth 5mm.

Exclusion Criteria

1. Those with acute or chronic medical conditions like bacterial, viral or fungal infections.
2. History of smoking
3. Any history of recent trauma or tooth extraction.
4. History of periodontal surgery or antibiotic uptake in last 3 months.
5. Pregnant/ lactating females.

The study population was divided into two groups:

Group 1 (Control): Periodontally healthy subjects

Group 2 (Test): Those with moderate to severe periodontitis.

Prior to the study, the design and purpose of the study was explained to the patients, a detailed proforma was prepared, and informed consent was obtained from all subjects. A full mouth periodontal examination of all the subjects was done using the following clinical parameters:

Gingival Index (Loe and Silness, 1963)⁷

Plaque Index (Turesky Gilmore modification of Quigley Hein Plaque Index)⁸

Pocket probing depth (PPD) using calibrated William's probe)

Sample Collection

After recording the clinical parameters, 5 ml of venous blood was drawn under aseptic conditions, from the ante-cubital fossa. It was transferred immediately to EDTA (ethylenediaminetetraacetic acid) containing vacutainers to be transported to the Medical Laboratory. The estimation of the following haematological parameters was done:

Haemoglobin (Hb), RBC count, Total Leukocyte Count (TLC), Differential Leukocyte Count (DLC), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC), Bleeding Time (BT), Clotting time (CT)

All the measurements were done using standardized and automated procedures using Poch 100i.

The data was collected and analysed using Mann Whitney U test. Pearson Chi square test was also applied to study the statistical significance between the blood parameters and the periodontal variables.

RESULTS

The present study showed the differences in the blood cell parameters in healthy subjects and those with chronic periodontitis. There were more females in both the groups. The study showed no significant gender predisposition between male and female patient groups suffering from anaemia of chronic disease. (Table 1)

The mean gingival and plaque index were significantly higher in patients with chronic periodontitis as compared to healthy group subjects. The mean gingival index of test group was 2.37 with a standard deviation of 0.48 i.e. 2.37 ± 0.48 as compared to control group which was 1.21 ± 0.49 , whereas the mean plaque index of test group was 2.25 ± 0.43 as compared to control group i.e. 1.35 ± 0.49 , both the values being statistically significant. (Table 2)

The present study showed decrease in haemoglobin levels and erythrocyte count in chronic periodontitis patients as compared to healthy subjects. The mean RBC count in test group is 4.5 ± 0.19 whereas it is 4.6 ± 0.41 in control group (p value < 0.05), which is statistically significant. The mean MCHC concentration of test group is 35.4 ± 1.6 , which is less as compared to that of control group i.e. 36.2 ± 1.5 (p value < 0.05), which is statistically significant. The mean MCH and MCV values were almost similar in both the groups and were not statistically significant (p values 0.43 and 0.06 respectively).

The TLC count was increased in test group (9582.09 ± 648.6) as compared to control group (8950.3 ± 859.4), with increase in counts of neutrophils, lymphocytes and eosinophils. The increase in neutrophil count was statistically significant (70.14 ± 1.3 in test group and 69.91 ± 2.8 in control group) and increase in lymphocyte count was also significant statistically (38.15 ± 2.7 in test group and 36.67 ± 2.4 in control group). The increase in eosinophil count was not statistically significant (p value 0.2).

(Table 3)

The present study showed decrease in levels of HB, RBC count, MCHC and increase in TLC levels in periodontitis patients, as compared to healthy control group subjects

DISCUSSION

Various epidemiological studies showed that periodontitis is associated with an increased risk of systemic disorders like cardiovascular diseases, cerebrovascular ischemia, and atherosclerosis, preterm low birth weight.^[9] The present study was aimed at showing an association between chronic periodontitis and anaemia of chronic disease.

In India, anaemia is more prevalent in females because of poor nutrition, increased menstrual losses, high incidence of

tropical and intestinal infections, and other miscellaneous factors. Females are also prone to hormonal imbalance during puberty, during the reproductive phase, and toward menopausal age. The microbial flora and host immune response are altered leading to exaggerated response of the periodontal tissues to local factors^[10]. The present study did not show any significant association between any gender predilection and anaemia of chronic disease in male and female patients (p 0.073). In a study by Viridi et al (2013)^[11] it was observed that haematological values in females were less as compared to males. The possible reason for this could be hormonal changes encountered in females during pregnancy, lactation, menstrual cycle, reproduction etc. as reported by Bentley ME and Griffiths PL in 2003.^[14]

The present study showed that there is decrease in values of haemoglobin, RBC count and MCHC in patients with chronic periodontitis as compared to periodontally healthy subjects. These results are in accordance with the study of Viridi H (2013)^[11], which showed that the mean values of haemoglobin and red blood cell indices were significantly lower and the values of erythrocyte sedimentation rate (ESR) were significantly higher in test group as compared to control group, suggesting a mild anaemia.

Studies by Hutter JW et al (2001)^[2], Seigel EH (1945)^[13], Gokhale SR (2010)^[12] also indicated that periodontitis patients have a lower hematocrit, lower numbers of erythrocytes, lower haemoglobin levels and higher erythrocyte sedimentation rates, hence indicating that chronic periodontitis can lead to anaemia.

All the processes involved in the development of ACD can be attributed to the cytokines, including shortened red cell survival, blunted erythropoietin response to anaemia, impaired erythroid colony formation in response to erythropoietin, and the abnormal mobilization of reticuloendothelial iron stores.

The present study results showed increase in levels of TLC, neutrophils, lymphocytes and eosinophils, which is in correlation to the studies by Kolte R et al (2014)^[16], Agnihotram (2010)^[15], which also showed increase in TLC levels, neutrophils, lymphocytes count in chronic periodontitis patients as compared to healthy control group subjects.

It is justified by the fact that in early stages of periodontitis, the rate of blood flow is increased due to vasodilatation. But subsequently, there is a slowing and stasis of blood stream. With stasis, changes in normal axial flow of the blood in the microcirculation take place. The normal axial flow consists of central stream of cells comprising leukocytes and RBC, and peripheral cell-free layer of plasma close to the vessel wall.

Due to stasis, the central stream of cells widens and the peripheral zone becomes narrower because of plasma loss by exudation. After this margination, the neutrophils of central column come close to the vessel wall as a result of redistribution. All consequences finally cause increase in neutrophils and leukocytes.^[17]

The results of the present clinical and haematological trial indicates that chronic periodontitis may be considered to have a tendency towards the development of anaemia, as it may cause decrease in number of erythrocyte count, Hb, decreased MCHC and an increase in the levels of neutrophils, lymphocytes and total leukocyte count. Further longitudinal studies are needed to investigate the association of blood cell parameters with periodontitis.

CONCLUSION

The present study showed a positive association between the haematological parameters i.e. Hb, RBC, TLC, MCHC and severity of chronic periodontal disease. It can be concluded that like any other chronic condition, periodontitis may tend towards anaemia. Such a comparison can add to our knowledge regarding changes in blood parameters in patients with chronic periodontitis. These blood cell parameters can be utilized as predictors or risk markers of life-threatening systemic diseases and may give an early option for timely intervention and diagnosis.

REFERENCES

1. Patel M, Shakir QJ, Shetty A. Interrelationship between chronic periodontitis and anemia: a 6-month follow up study. *J Indian Soc Periodontol* 2014;18:19-25.
2. 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;28:930-6.
3. Scannapieco FA, Bush RB, Paju S. Associations between periodontal disease and risk for atherosclerosis, cardiovascular disease and stroke. A systematic review. *Ann Periodontol* 2003;8:38-53.
4. Weiss G, Goodnough LT. Anemia of chronic disease. *N Engl J Med* 2005;352:1011-23.
5. Wakai K, Kawamura T, Umemura O, Hara Y, Machida J, Anno T, et al. Associations of medical status and physical fitness with periodontal disease. *J Clin Periodontol* 1999;26:664-72.
6. Havemose-Poulsen A, Westergaard J, Stoltze K, Skjodt H, Danneskiold-Samsøe B, Locht H et al. Periodontal and hematological characteristics associated with aggressive

- periodontitis, juvenile idiopathic arthritis, and rheumatoid arthritis. *J Periodontol* 2006;77:280-8.
7. Loe H, Silness J. Periodontal diseases in pregnancy I. Prevalence and severity. *Acta Odontol Scand* 1963;21:533-51.
 8. Breuer M, Cosgrove R. The relationship between gingivitis and plaque levels. *J Periodontol* 1989;60:172-5.
 9. Scannapieco FA. Position paper of The American Academy of Periodontology: Periodontal disease as a potential risk factor for systemic diseases position paper. *J Periodontol* 1998;69:841-50.
 10. Amar S, Chung KM. Effects of hormonal variation on the periodontium in women. *Periodontol 2000* 1994;6:79-87.
 11. Kaur Viridi, H. Hematological parameters- A diagnostic mirror for periodontics. *Indian Journal of Dental Sciences* 2013;2:45-8.
 12. Gokhale SR, Sumanth S, Padhye A. Evaluation of blood parameters in patients with chronic periodontitis for signs of anemia. *J Periodontol* 2010;81:1202-6.
 13. Seigel EH. Total erythrocyte, leukocyte and differential white cell counts of blood in chronic periodontal disease. *J Dent Res* 1945;24: 270.
 14. Bentley ME, Griffiths PL. The burden of anemia among women in India. *Eur J Clin Nutrition* 2003; 57:52-60.
 15. Agnihotram G, Mahesh Singh TR, *et al.* Study of clinical parameters in chronic periodontitis. *Int J Appl Biol Pharm Technol* 2010;1:1202-7
 16. Kotle R, Kotle A, Deshpande N. Assessment and comparison of anemia of chronic disease in healthy subjects and chronic periodontitis patients: A clinical and haematological study. *J Indian Soc Periodontol* 2014;18:183-6.
 17. Noack B, Genco RJ, Trevisan M, Grossi S, Zambon JJ, De Nardin E. Periodontal infections contribute to elevated systemic C-reactive protein level. *J Periodontol* 2001;72: 1221-7.