

## ORIGINAL RESEARCH

### Evaluation of serum mineral micronutrients (Zn, Cu, Fe, Mg) and their correlation with clinical parameters (gingival index, probing pocket depth, clinical attachment loss) in chronic periodontitis patients

Dr. Sunil Kumar<sup>1</sup>, Dr. Kumari Menka<sup>2</sup>, Dr. Sarbil Kumari<sup>3</sup>, Dr. Prakash Chandra Jha<sup>4</sup>

<sup>1</sup>Tutor, Department of Physiology, Bhagwan Mahavir Institute of Medical Sciences, Pawapuri, Nalanda, Bihar, India.

<sup>2</sup>Senior Resident, Department of Pedodontics and Preventive dentistry, Nalanda Medical College & Hospital, Patna, Bihar

<sup>3</sup>Professor and Head of Department, Department of Physiology, Bhagwan Mahavir Institute of Medical Sciences, Pawapuri, Nalanda, Bihar, India

<sup>4</sup>Assistant Professor and Head of Department, Department of Oral Medicine and Radiology, Nalanda Medical College & Hospital, Patna, Bihar

**Corresponding Author:** Dr. Kumari Menka

Senior Resident, Department of Pedodontics and Preventive dentistry, Nalanda Medical College & Hospital, Patna, Bihar

Email: [menka.kumari959@gmail.com](mailto:menka.kumari959@gmail.com)

Received Date: 10 January 2024

Acceptance Date: 15 February 2024

#### ABSTRACT

**Background:** Periodontitis, as a common chronic infectious disease in dentistry, is mediated by the immune-inflammatory response to the accumulated periodontal pathogens in the periodontal tissues, which involves both innate and acquired immunity.

**Aims and objectives:** To evaluate serum mineral micronutrients (Zn, Cu, Fe, and Mg) and their correlation with clinical parameters (gingival index, probing pocket depth, clinical attachment loss) in chronic periodontitis subjects.

**Materials and methods:** A total of 100 subjects were enrolled. 50 were in the case group and 50 in the control group. The age of the subjects was between 30 and 60 years. The data was collected and analysed using SPSS software. An independent t-test was done. The chi-square test was done. P <0.05 was considered significant.

**Results:** Mean Cu and Fe levels were significantly higher in the chronic periodontitis group in comparison to the control group, while mean Zn and Mg levels were significantly lower in the chronic periodontitis group in comparison to the control group.

**Conclusion:** Serum zinc and magnesium levels showed a negative correlation with periodontal clinical parameters (gingival index, PPD, and clinical attachment loss).

**Keywords:** periodontitis, gingival index, micronutrients.

#### Introduction

Periodontitis as a common chronic infectious disease in dentistry is mediated by the immune inflammatory response to the accumulated periodontal pathogens in the periodontal tissues, which involves both innate and acquired immunity.<sup>1</sup> It decreases the supporting bone level and eventually leads to tooth loss. It is widely known that periodontal diseases (PDs) not only induce local inflammation but also lead to higher systematic inflammation such as vascular dysfunction and cardiovascular events.<sup>2</sup> Periodontitis is also an inflammatory disease with host immune responses against bacterial infections. Globally, the prevalence of severe periodontitis is approximately 10%.<sup>3</sup> Untreated periodontitis is the main cause of tooth loss and is considered a significant threat to systemic health.<sup>4,5</sup> In periodontitis, pathogenic microorganisms and their products evoke immune-inflammatory responses in host tissues, resulting in increased white blood cell (WBC) counts as well as serum levels of C-reactive protein (CRP) and various inflammatory cytokines, including interleukin (IL), interferon, and members of the tumour necrosis factor superfamily.<sup>6</sup> Magnesium is an essential nutrient that is required for a variety of physiologic functions in the body. The consequences of magnesium deficiency go beyond oral lesions and may have an impact on the course of the COVID-19 pandemic.<sup>7</sup> Most of the magnesium absorbed by the body comes from foods such as nuts, seeds, whole grains, and green leafy vegetables, while magnesium is necessary for the maintenance and formation of calcified tissues such as bone.<sup>8</sup> Studies have shown a complex internal relationship between periodontitis and diabetes.<sup>9</sup> In obese men, the risk of periodontitis is significantly higher in those on a high-fat diet than in those on a healthy diet.<sup>9</sup>

The optimal level of zinc is important for the growth and development of human health.<sup>10</sup> In the human body, zinc is found in muscles (60%), bones (30%), and skin (5%).<sup>11</sup> It has an array of functions, including being involved in the activation of various enzymes and proteins, and zinc contributes to the absorption of vitamin A, E, and folate.<sup>10</sup> Low levels of zinc can be associated with an increased chance of developing infections and degenerative pathologies.<sup>10</sup> Zinc also plays an important role in the psychosocial functioning of human behaviour. In the oral cavity, zinc is found in saliva, dental plaque, and the hydroxyapatite of the dental enamel.<sup>12</sup> It contributes to healthy tooth formation and is used in mouth rinses and toothpaste due to its important role in the prevention of plaque and dental calculus formation.<sup>11</sup> Zinc also contributes to the reduction of halitosis in the mouth.<sup>11</sup> It has been implicated in the composition of dental biomaterials and orthodontic materials due to its properties for enhancing immunity as well as its effects on cell division and skeletal development.<sup>13</sup> Clinical trials have demonstrated that zinc ions decrease the rate of enamel demineralization.<sup>11</sup> The concentration of zinc on the enamel surface ranges from 430 to 2100 parts per million (ppm), and it is deposited mostly before tooth eruption.<sup>14</sup> Zinc is important for maintaining periodontal health because of its local and immunological effect on oral soft tissues. As with other micronutrients that fall into the category of minerals that are needed in quantities of <100 mg/day, the recommended daily allowance for zinc ranges between 2 and 13 mg/day, depending on the stage of life and sex of the individual, with the upper limit for zinc being set at 40 mg/day.<sup>10</sup>

**Aims and objectives:** The present study was conducted to evaluate serum mineral micronutrients (Zn, Cu, Fe, and Mg) and their correlation with clinical parameters (gingival index, probing pocket depth, and clinical attachment loss) in chronic periodontitis subjects.

**Materials and methods**

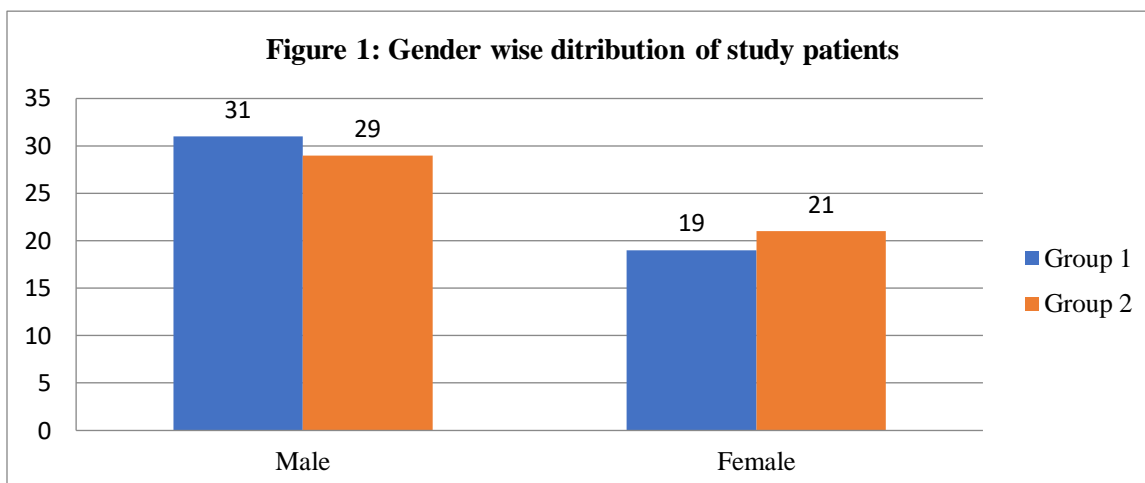
A prospective cross-sectional, randomised, double-blinded study was undertaken on 100 subjects attending the OPD of General Medicine in collaboration with the Department of Physiology at Bhagwan Mahavir Institute of Medical Sciences, Pawapuri, Nalanda, Bihar, India, and the Department of Pedodontics and Preventive Dentistry, Nalanda Medical College & Hospital, Patna, Bihar. The Institutional Ethics Committee granted ethical approval beforehand. The patients provided their informed consent. The study was carried out over a six-month period, from July 2023 to December 2023. Data such as name, age, etc. was recorded. 50 were in the case group and 50 in the control group. The age of the subjects was between 30 and 60 years. According to the 2018 categorization of periodontal disease, those with mild periodontitis had generalised (i) PPD ≤5 mm and (ii) CAL 3–4 mm. (iv) No tooth loss; (iii) Gingival index score < 2. The subjects were divided into two groups: group 1 were the subjects with chronic periodontitis, and group 2 were the healthy controls. Laboratory investigations were done. Blood samples were taken and centrifuged. Zinc, Cu, Fe, and Mg levels were measured. The data was collected and analysed using SPSS software version 22.0. An independent t-test was done. The chi-square test was done. P <0.05 was considered significant.

**Results**

A total of 50 chronic periodontitis patients and 50 controls were enrolled. The mean age of the patients in the chronic periodontitis group and the control group was 49.2 years and 51.7 years, respectively. Mean Cu and Fe levels were significantly higher in the chronic periodontitis group in comparison to the control group, while mean Zn and Mg levels were significantly lower in the chronic periodontitis group in comparison to the control group. While correlating clinical parameters with Cu and Fe levels, a significant positive correlation was seen. However, Zn and Mg levels showed a significant negative correlation while correlating with clinical parameters.

**Table 1: Demographic characteristics of patients**

Variable	Category	Chronic periodontitis group (Group 1)		Control group (Group 2)	
		Number	Percentage	Number	Percentage
Gender	Males	31	62	29	58
	Females	19	38	21	42
Age group (years)	Less than 40	17	34	20	40
	More than 40	33	66	30	60
Mean age (years)		49.2		51.7	



**Table 2: Comparison of Micronutrients**

Micronutrients	Group 1		Group 2		p-value
	Mean	SD	Mean	SD	
Cu (mg/dL)	0.251	0.95	0.113	0.56	0.002*
Fe (mg/dL)	0.310	1.02	0.173	0.63	0.000*
Zn (mg/dL)	0.079	0.13	0.118	0.58	0.001*
Mg (md/dL)	1.49	0.88	1.79	0.97	0.000*

**Table 3: Correlation of micronutrients with clinical parameters among chronic periodontitis patients**

Micronutrients	Clinical parameters					
	Gingival index		PPD		Clinical attachment loss	
	r-value (Pearson's correlation)	p-value	r-value (Pearson's correlation)	p-value	r-value (Pearson's correlation)	p-value
Cu (mg/dL)	1.253	0.000*	0.945	0.003*	1.325	0.000*
Fe (mg/dL)	0.845	0.009*	1.135	0.000*	0.948	0.004*
Zn (mg/dL)	-1.225	0.001*	-1.625	0.007*	-1.278	0.002*
Mg (md/dL)	-1.315	0.000*	-1.187	0.000*	-1.339	0.001*

\*Significant

### Discussion

Periodontitis is an inflammatory disease of the supporting tissues of the tooth caused by specific microorganisms in a susceptible host. 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.<sup>15</sup> Chronic periodontitis is the most common form of periodontal disease, which progresses relatively slowly and is more common in adults.<sup>16</sup> Hence, this study was conducted to evaluate serum mineral micronutrients (Zn, Cu, Fe, and Mg) and their correlation with clinical parameters (gingival index, probing pocket depth, and clinical attachment loss) in chronic periodontitis subjects. In the present study, a total of 50 chronic periodontitis patients and 50 controls were enrolled. The mean age of the patients in the chronic periodontitis group and the control group was 49.2 years and 51.7 years, respectively. Mean Cu and Fe levels were significantly higher in the chronic periodontitis group in comparison to the control group, while mean Zn and Mg levels were significantly lower in the chronic periodontitis group in comparison to the control group. A study by Anusree Raju et al.<sup>17</sup> estimated the serum levels of Zn, Cu, Fe, and Mg of chronic periodontitis patients and normal healthy controls to measure the clinical parameters (gingival index, probing pocket depth [PPD], and clinical attachment loss) in chronic periodontitis patients and normal healthy controls., to compare the levels of serum Zn, Cu, Fe, and Mg levels of chronic periodontitis patients and healthy controls, and to correlate the levels of serum micronutrients with clinical parameters (gingival index, PPD, and

clinical attachment loss) in chronic periodontitis patients and healthy controls. A total of 110 subjects—55 subjects with chronic periodontitis and 55 healthy control subjects in the age group 35–65 years—were selected for the study. The serum micronutrient levels of Cu, Fe, Zn, and Mg and the clinical parameters were measured. Serum concentrations of Cu and Fe showed a statistically significant increase, and serum Zn and Mg showed a significant decrease in periodontitis patients as compared to normal healthy controls. Copper and Fe showed a significant positive correlation, and Zn and Mg showed a significant negative correlation with clinical parameters (gingival index, PPD, and clinical attachment loss). The assessment of serum Mineral micronutrients can serve as possible biomarkers or indicators for an inflammatory condition like chronic periodontitis. In the present study, while correlating clinical parameters with Cu and Fe levels, a significant positive correlation was seen. However, Zn and Mg levels showed a significant negative correlation while correlating with clinical parameters. Another study by Li XY et al.<sup>18</sup> examined the association between dietary magnesium and periodontitis using a multivariable logistic regression model. Based on odds ratios (OR) and 95% confidence intervals (CIs), a strong association was detected. Multivariable logistic regression analysis showed that the OR for periodontitis comparing the highest to the lowest quintile of dietary magnesium intake was 0.69 (95% CIs = 0.52~0.92). The restricted cubic spline (RCS) analysis showed that the non-linear association between dietary magnesium and periodontitis was statistically significant and that dietary magnesium supplementation reduced the prevalence of periodontitis. Dietary magnesium intake is associated with the prevalence of periodontitis. Dietary magnesium deficiency increases the prevalence of periodontitis. According to Sundaram G et al.<sup>19</sup> one hundred and twenty patients were included in this study, which was further divided into three groups. Group 1 consisted of forty patients with CP; Group 2 consisted of forty patients with CP with controlled diabetes; and Group 3 consisted of forty patients with CP with uncontrolled diabetes. Periodontal parameters such as plaque index, gingival index, bleeding on probing, pocket depth, and clinical attachment levels (CALs) were evaluated. Blood samples were collected to assess the levels of fasting blood sugar, glycosylated hemoglobin, Zn, Mg, and Cu. All parameters were evaluated at baseline and 3 months after nonsurgical periodontal therapy. The results showed a statistically significant reduction in all the clinical parameters within the groups except for the CAL in group 1 patients ( $P = 0.05$ ). The glycemic status also showed a statistically significant reduction after treatment ( $P < 0.001$ ). The intragroup comparison was taken between the values of micronutrients and showed a substantial increase in the levels of both Zn and Mg and a decrease in the level of Cu after nonsurgical periodontal treatment ( $P < 0.001$ ). Patients with diabetes and periodontitis had altered metabolisms of Zn, Mg, and Cu, contributing to the progression and complications of diabetes mellitus and periodontitis. Nonsurgical periodontal treatment improved the variation and concentration of plasma micronutrients, as well as the periodontal status and glycemic level. Zinc supplementation is effective in preventing gingival disease; it can fight against *Fusobacterium nucleatum* and *Prevotella intermedia*, which cause gingivitis. Zinc also inhibits proteases produced by *Porphyromonas gingivalis*.<sup>20</sup> zinc oxide nanoparticles were found to disrupt the cell membrane and oxidative stress in *Campylobacter* species as well as in other gram-positive and gram-negative bacteria found in gingivitis.<sup>20</sup> A six-month randomized clinical study comparing two dentifrices, one containing fluoride and zinc and the other containing fluoride alone, revealed that the zinc-based dentifrice provided a meaningful clinical reduction in gingivitis and dental plaque.<sup>21</sup> A study performed by Hong and colleagues demonstrated that A20, an amino-acid protein with zinc finger C-terminal domains, may be a potential element in preventing inflammatory bone loss diseases such as periodontitis due to its anti-inflammatory and anti-osteoclastogenic effects.<sup>22</sup> Conversely, researchers have highlighted an association between zinc deficiency and gingivitis.<sup>20</sup> Understanding the role and implication of different nutrients should allow identifying nutritional risk factors and modulators of periodontal inflammation for targeted prevention and treatment approaches in patients with specific nutritional depletion.<sup>23–24</sup> The absorption and metabolism of dietary nutrients determined by genetic factors are key to the underlying reasons for inflammatory processes. Nutrition has significant effects on inflammatory processes as well as on the cellular and humoral immune mechanisms. The generation of factors responsible for the progression of periodontal disease may be due to the interaction between nutritional status and the immune response.<sup>25,26</sup>

**Limitations of study:** The sample size was small and the duration of the study was short.

### **Conclusion**

Serum copper and iron showed a significant positive correlation with periodontal clinical parameters (gingival index, PPD, and clinical attachment loss). Serum zinc and magnesium levels showed a negative correlation with periodontal clinical parameters (gingival index, PPD, and clinical attachment loss).

**Acknowledgement:** The authors would like to acknowledge the entire faculty and staff members of the Department of Physiology for their valuable support and time-to-time suggestions in undertaking the present study. Dr. Sunil Kumar gave study design, data collection, and analysis, and Dr. Kumari Menka helped with manuscript drafting,

manuscript revision, data collection, and analysis. Special thanks to Dr. Sarbil Kumari, Professor and Head of Department, Department of Physiology, Bhagwan Mahavir Institute of Medical Sciences, Pawapuri, Nalanda, Bihar, India, and Dr. Prakash Chandra Jha, Assistant Professor and Head of Department, Department of Oral Medicine and Radiology, Nalanda Medical College & Hospital, Patna, Bihar.

## References

1. Cekici A., Kantarci A., Hasturk H., Van Dyke T. E. (2014). Inflammatory and immune pathways in the pathogenesis of periodontal disease. *Periodontol 2000* 64, 57–80.
2. Nibal L., Darbar U., Rakmanee T., Donos D. (2019). Anemia of inflammation associated with periodontitis: analysis of two clinical studies. *J Periodontol.* 90, 1252–1259.
3. Frencken J.E., Sharma P., Stenhouse L., Green D., Lavery D. and Dietrich T. (2017) Global epidemiology of dental caries and severe periodontitis: a comprehensive review. *J. Clin. Periodontol.* 44, S94–S105
4. Genco R.J. and Borgnakke W.S. (2013) Risk factors for periodontal disease. *Periodontol 2000* 62, 59–94
5. Tonetti M.S., Van Dyke T.E. and Working group 1 of the joint EFP/AAPw (2013) Periodontitis and atherosclerotic cardiovascular disease: consensus report of the Joint EFP/AAP Workshop on Periodontitis and Systemic Diseases. *J. Clin. Periodontol.* 40, S24–S29
6. Amano A. (2010) Host-parasite interactions in periodontitis: microbial pathogenicity and innate immunity. *Periodontol 2000* 54, 9–14 10.
7. Eskander M, Razzaque MS. Can maintaining optimal magnesium balance reduce the disease severity of COVID-19 patients? *Front Endocrinol.* (2022) 13:843152.
8. Wang K, Wei H, Zhang W, Li Z, Ding L, Yu T, et al.. Severely low serum magnesium is associated with increased risks of positive anti-thyroglobulin antibody and hypothyroidism: a cross-sectional study. *Sci Rep.* (2018) 8:9904.
9. Wright DM, McKenna G, Nugent A, Winning L, Linden GJ, Woodside JV. Association between diet and periodontitis: a cross-sectional study of 10,000 NHANES participants. *Am J Clin Nutr.* (2020) 112:1485–91.
10. Gaur S., Agnihotri R. Trace mineral micronutrients and chronic periodontitis—A review. *Biol. Trace Elem. Res.* 2017;176:225–238
11. Lynch R.J. Zinc in the mouth, its interactions with dental enamel and possible effects on caries; a review of the literature. *Int. Dent. J.* 2011;61(Suppl. 3):46–54. doi: 10.1111/j.1875-595X.2011.00049.x.
12. Devi C.B., Nandakishore T., Sangeeta N., Basar G., Devi N.O., Jamir S., Singh M.A. Zinc in human health. *IOSR J. Dent. Med Sci. (IOSR-JDMS)* 2014;13:18–23.
13. Glenske K., Donkiewicz P., Kowitsch A., Milosevic-Oljaca N., Rider P., Rofall S., Franke J., Jung O., Smeets R., Schnettler R., et al. Applications of metals for bone regeneration. *Int. J. Mol. Sci.* 2018;19:826.
14. Rahman M.T., Hossain A., Pin C.H., Yahya N.A. Zinc and metallothionein in the development and progression of dental caries. *Biol. Trace Elem. Res.* 2019;187:51–58.
15. Pradeep AR, Anuj S. Anemia of chronic disease and chronic periodontitis: Does periodontal therapy have an effect on anemic status? *J Periodontol.* 2011;82:388–94.
16. Naik V, Acharya A, Deshmukh VL, Shetty S, Shirhatti R. Generalized, severe, chronic periodontitis is associated with anemia of chronic disease: A pilot study in urban Indian males. *J Invest Clin Dent.* 2010;1:139–43.
17. AnusreeRaju, SanjeevRavindran, Shyamala Devi MP, Shaheena KP, AnushaAnish, Hari S Pillai, & Annamala PT. (2023). Evaluation of serum mineral micronutrients (Zn, Cu, Fe, Mg) and their correlation with clinical parameters (gingival index, probing pocket depth, clinical attachment loss) in chronic periodontitis patients. *Asian Journal of Medical Sciences*, 14(6), 233–242.
18. Li XY, Wen MZ, Liu H, Shen YC, Su LX, Yang XT. Dietary magnesium intake is protective in patients with periodontitis. *Front Nutr.* 2022 Aug 25;9:976518.
19. Sundaram G, Ramakrishnan T, Parthasarathy H, Moses J, Lalitha T. Evaluation of Micronutrient (Zinc, Magnesium, and Copper) Levels in Serum and Glycemic Status after Nonsurgical Periodontal Therapy in Type 2 Diabetic Patients with Chronic Periodontitis. *Contemp Clin Dent.* 2017 Jan-Mar;8(1):26-32.
20. Fatima T., Rahim Z.B., Lin C.W., Qamar Z. Zinc: A precious trace element for oral health care? *JPMA J. Pak. Med. Assoc.* 2016;66:1019–1023.
21. Zhong Y., Li X., Hu D.Y., Mateo L.R., Morrison B.M., Jr., Delgado E., Zhang Y.P. Control of established gingivitis and dental plaque using a 1450 ppm fluoride/zinc-based dentifrice: A randomized clinical study. *J. Clin. Dent.* 2015;26:104–108.

22. Hong J.Y., Bae W.J., Yi J.K., Kim G.T., Kim E.C. Anti-inflammatory and anti-osteoclastogenic effects of zinc finger protein A20 overexpression in human periodontal ligament cells. *J. Periodontal Res.* 2016;51:529–539.
23. Ashimoto A, Chen C, Bakker I and Slots J. Polymerase chain reaction detection of putative periodontal pathogens in subgingival plaque of gingivitis and advanced periodontitis lesions. *Oral MicrobiolImmunol.* 1996;11:266-273.
24. Beck J, Garcia R, Heiss G, Vokonas PS and Offenbacher S. Periodontal disease and cardiovascular disease. *J Periodontol.* 1996;67(10 Suppl):1123-1137.
25. Hujoel PP, Drangsholt M, Spiekerman C and DeRouen TA. Periodontal disease and coronary heart disease risk. *JAMA.* 2000;284(11):1406-1410.
26. Armitage GC. Bi-directional relationship between pregnancy and periodontal disease. *Periodontol* 2000. 2013;61(1):160-176.