Biological Trace Element Research (2018) 186:68–73
https://doi.org/10.1007/s12011-018-1296-z

Association of Periodontitis with the Concentration Levels of Germanium and Tin in Hair

Hye-Sung Kim[1] · Hyun-Jae Cho[2,3] · Soo-Myoung Bae[4] · Young-Youn Kim[1] · Sung-In Baek[1] · Kwang-Hak Bae[1]

Received: 10 January 2018 / Accepted: 1 March 2018 / Published online: 10 March 2018

Abstract

The purpose of this study was to investigate whether or not the concentration levels of certain kinds of trace elements in hair are associated with periodontitis. We studied a total of 109 participants, which are composed of 25 participants with periodontitis and 84 participants without periodontitis. Periodontal conditions were assessed by measuring the periodontal clinical attachment loss and pocket depth, which were determined at six sites of all teeth. Periodontitis was defined according to the criteria of periodontitis proposed by CDC-AAP. The hair samples were washed with acetone, water, and extran (1%*v*/*v*), and then aliquots of hair samples were wet-ashed. This sample solution was analyzed by Perkin-Elmer Mass Spectrometer. The odds ratios and 95% confidence intervals of the concentration levels of trace elements for periodontitis were calculated by multivariate logistic regression analysis. After adjusting all confounders, it was found that the higher concentration level of germanium in hair was significantly and positively associated with periodontitis (odds ratio [OR] 7.12; 95% confidential interval [CI] 2.03–25.00). The higher concentration level of tin in hair was significantly and negatively associated with periodontitis (OR 0.27; 95% CI 0.08–0.94). It was concluded that there was a significant relationship between periodontitis and the concentration level of germanium and tin in hair.

Introduction

Periodontitis is a chronic inflammatory disease that leads to destruction of the connective tissue and alveolar bone around teeth [1]. Due to its high prevalence in adults, it is a major cause of teeth extraction in adults and represents one of important public health problems increasing the burden of chronic diseases in many countries [2, 3]. In Korea, the prevalence of periodontitis is about 30% among adults in recent years [4].

Hye-Sung Kim and Hyun-Jae Cho contributed equally to this work.

✉ Kwang-Hak Bae
sadent@naver.com

1 Oral Health Science Research Center, Apple tree Dental Hospital, Jungang-ro 1573, Goyang-si, Gyounggi-do 10381, South Korea

2 Department of Preventive and Public Health Dentistry, School of Dentistry, Seoul National University, Seoul, South Korea

3 Dental Research Institute, School of Dentistry, Seoul National University, Seoul, South Korea

4 Department of Dental Hygiene, College of Dentistry & Research Institute of Oral Science, Gangneung-Wonju National University, Gangneung-si, South Korea

Since it may increase a chronic burden of endotoxin and inflammatory cytokines at local as well as distant sites, it has been considered as a risk factor for systemic illnesses including cardiovascular disease, atherosclerosis, and cerebrovascular ischemia [5–8]. To explain the link between periodontitis and chronic systemic disorders, various hypothetical mechanisms have been proposed. Some studies reported that periodontitis was associated with chronic systemic disorders via oxidative stress [9–13]. Recently, the nitrate-nitrite-nitric oxide pathway has also been noted as the link between dysbiotic oral microbial communities and systemic diseases [14].

Since minerals usually act as cofactors in enzymes and their presence in balanced concentrations in different body compartments is needed for multiple physiological processes (heart rhythm, muscle contraction, nerve conduction, and acid-base balance homeostasis), it has been suggested that the risk of periodontitis may be associated with minerals [15]. It has been widely reported that nutritional minerals such as calcium, magnesium, and phosphorus are associated with periodontitis [16–18]. Various observational studies and animal experiments showed the effects of nutritional minerals on the prevalence of periodontitis or the conditions of periodontium [17, 19]. Some studies reported that toxic minerals such as lead, cadmium, and mercury are also associated with periodontitis because they can cause serious adverse health effects including kidney disease and poor bone remodeling [20, 21]. Lately, there are a few studies reporting the cytotoxicity or the usefulness of disease biomarkers of trace elements, which are not clearly understood whether they are nutritional or toxic to human body [22, 23]. However, there is no study reporting the associations between periodontitis and trace elements.

Therefore, we aimed to determine whether the concentration levels of certain kinds of trace elements in hair are associated with periodontitis.

Materials and Methods

Study Design and Subject Selection

The study was conducted in compliance with the principles of the Helsinki Declaration. Ethical clearance of the study was approved by the Institutional Review Board of Appletree Dental Hospital (Institutional Review Board no. 0841-2017-001).

This study was designed as a hospital-based cross-sectional study, which was performed from February to October 2017. This study's participants consisted of outpatients in the Appletree Dental Hospital in Ilsan, South Korea. Criteria for inclusion in the study participants were (1) 18 years and over, (2) those with at least 20 teeth, (3) and those who received no periodontal treatment in the past 3 months. Criteria for exclusion were (1) pregnant women, (2) those who had taken antibiotics in the past 3 months, and (3) those who were diagnosed with diabetes.

Clinical Variables

Periodontitis

One trained dentist performed periodontal examination on all participants. The dentist conducted periodontal probing with a periodontal probe (Hu-Friedy, Chicago, IL). The participants lied on a dental unit chair (Summit Chairs, Wiltshire, UK) during examination. Periodontal conditions were assessed by measuring the periodontal clinical attachment loss, pocket depth, and bleeding on probing which were determined at six sites (mesio-buccal, mid-buccal, disto-buccal, disto-lingual, mid-lingual, and mesio-lingual surfaces) of all teeth except for the third molars and distal sites of the second molars. Periodontitis was defined according to the criteria of periodontitis proposed by CDC-AAP [24]. The participants diagnosed as "no" or "mild" periodontitis were categorized into "no periodontitis" group, and those diagnosed as "moderate" or "severe" periodontitis were categorized into "periodontitis" group.

The Concentration of Trace Elements in Hair

The hair should be free of all gels, oils, and hair creams on sample collection day. Hair samples were analyzed by US TEI (Dallas, TX, US). Each hair sample was washed with acetone, water, and extran (1%*v*/*v*), and aliquots of hair samples were wet-ashed according to the following procedure: a 250-mg sample was wet digested overnight with 2.5 ml of HNO_3 in a closed, graduated polypropylene tube (50 ml) at room temperature, and then for 1 h at 60–70 °C in a drying oven. After cooling to room temperature, the sample was diluted to a final volume of 25 ml with Milli Q water. This solution was analyzed by Perkin-Elmer Mass Spectrometer (SciexElan 6100, Perkin-Elmer corporation, Foster, CA, US). More details of the analysis methods are described in Miekeley et al.'s study [25].

Covariates

Socio-demographic variables comprised gender, age, and household income. Household income was the self-estimated levels of the family income. Oral health behaviors included daily frequency of tooth brushing, usage of dental floss or an interdental brush, and regular dental visit. General health behaviors included smoking and drinking status. Participants were divided into three groups depending on the current smoking status: non-smokers, current smokers, and past smokers (those who had smoked in the past but were not currently smoking). They were also divided into three groups depending on the current drinking status: non-drinkers, current drinkers, and past drinkers (those who had drunken in the past but were not currently drinking).

Statistical Analysis

Statistical significance was determined at $p < 0.05$. The outcome variable was whether a participant had periodontitis or not, and the explanatory variables were whether the concentrations of trace elements in a participant's hair were over median or not. Covariates consisted of age, gender, household income, present smoking, present drinking, daily frequency of tooth brushing, usage of floss or interdental brush, and regular dental visit. Comparison between the no periodontitis and periodontitis groups for the explanatory variables and covariates was performed by chi-square test for categorical variables and by independent samples t test for normally distributed continuous variables. Comparison between the two groups for the concentrations of trace elements in hair was performed by Mann-Whitney U test because the data was not normally distributed. The odds ratios and 95% confidence intervals of explanatory variables for periodontitis were calculated by multivariate logistic regression analysis. All of the mentioned covariates were considered as confounders in the multivariable logistic regression model. Statistical analyses were performed using SPSS statistical software version 19.0 (SPSS, Chicago, IL).

Table 1 Univariate comparisons of characteristics in participants with and without periodontitis ($n = 109$)

Characteristics	No periodontitis		Periodontitis		p^a
	Number	%	Number	%	
Age	41.38 (13.35)[b]		51.96 (9.29)[b]		< 0.001
Gender					0.184
Male	25	29.8	11	44.0	
Female	59	70.2	14	56.0	
Household income					1.000
High	10	11.9	3	12.0	
Middle	47	56.0	14	56.0	
Low	27	32.1	8	32.0	
Present smoking					0.228
Non-smoker	64	76.2	15	60.0	
Past smoker	14	16.7	8	32.0	
current smoker	6	7.1	2	8.0	
Present drinking					0.384
Non-drinker	25	29.8	10	40.0	
Past drinker	6	7.1	3	12.0	
Current drinker	53	63.1	12	48.0	
Daily frequency of tooth brushing					0.263
Once or less	49	58.3	10	40.0	
Twice	32	38.1	14	56.0	
Three times or more	3	3.6	1	4.0	
Usage of floss or interdental brush					0.066
No	20	23.8	2	8.0	
Occasionally	33	39.3	16	64.0	
Everyday	31	36.9	7	28.0	
Regular dental visit					0.112
No	32	38.1	14	56.0	
Yes	52	61.9	11	44.0	
Bleeding on probing	0.92 (0.55)[b]		1.14 (0.45)[b]		0.069
Pocket depth (mm)	1.88 (0.43)[b]		2.35 (0.34)[b]		< 0.001
Attachment loss (mm)	1.93 (0.44)[b]		2.69 (0.57)[b]		< 0.001

[a] Determined by chi-square test for categorical variables and by *t* test for continuous variables

[b] Mean (standard deviation)

Results

Table 1 compares socio-demographic characteristics and health behaviors between no periodontitis and periodontitis groups. The mean age was significantly different between the two groups ($p < 0.001$). The proportion of users of dental floss or interdental brush was marginally higher in the no periodontitis group than in the periodontitis group ($p = 0.066$).

Table 2 compares the concentration levels of trace elements of hair between the no periodontitis and periodontitis groups. The median of the concentration of germanium was 0.050 in the no periodontitis group and 0.070 in the periodontitis group, which represents a significant difference between the two groups ($p = 0.003$). The concentration of tin was significantly lower in the periodontitis group than in the no periodontitis group ($p = 0.029$).

Table 3 shows the association between the concentration levels of trace elements in hair and periodontitis in the multivariate logistic regression model. The concentration level of germanium in hair was significantly associated with periodontitis (odds ratio [OR] 7.12; 95% confidential interval [CI] 2.03–25.00). The concentration level of tin in hair was also significantly associated with periodontitis (OR 0.27; CI 0.08–0.94).

Discussion

This study investigated whether or not the concentration levels of trace elements in hair were associated with periodontitis among 109 participants after adjusting for socio-demographic variables and oral and general health behaviors.

Table 2 Univariate comparisons of the concentration levels of trace elements of hair in participants with and without periodontitis ($n = 109$)

Elements	No periodontitis		Periodontitis		p^a
	Mean (SD)	Median (Min, LQ, UQ, Max)	Mean (SD)	Median (Min, LQ, UQ, Max)	
Germanium	0.049 (0.033)	0.050 (0.010, 0.020, 0.060, 0.270)	0.061 (0.016)	0.070 (0.020, 0.055, 0.070, 0.090)	0.003
Barium	2.732 (4.018)	1.800 (0.200, 0.800, 3.700, 34.50)	1.972 (1.811)	1.100 (0.100, 0.450, 3.250, 6.600)	0.215
Rubidium	0.090 (0.113)	0.051 (0.009, 0.025, 0.100, 0.708)	0.103 (0.092)	0.056 (0.014, 0.027, 0.175, 0.301)	0.325
Lithium	0.013 (0.007)	0.010 (0.010, 0.010, 0.010, 0.040)	0.012 (0.004)	0.010 (0.010, 0.010, 0.010, 0.020)	0.877
Nickel	0.423 (0.592)	0.200 (0.100, 0.125, 0.400, 4.000)	0.360 (0.380)	0.300 (0.100, 0.100, 0.450, 2.000)	0.860
Vanadium	0.034 (0.018)	0.030 (0.010, 0.020, 0.040, 0.090)	0.041 (0.029)	0.030 (0.010, 0.020, 0.040, 0.140)	0.331
Strontium	6.442 (5.244)	4.600 (0.300, 2.325, 10.275, 29.800)	5.440 (4.855)	4.000 (0.200, 1.000, 8.500, 18.600)	0.275
Tin	0.665 (1.627)	0.100 (0.100, 0.100, 0.400, 12.100)	0.180 (0.263)	0.100 (0.100, 0.100, 0.150, 1.400)	0.029
Titanium	1.261 (0.874)	1.000 (0.500, 0.800, 1.400, 5.900)	1.356 (0.874)	1.200 (0.600, 0.700, 1.550, 4.200)	0.659

The unit of the concentration: part per million (ppm)

SD standard deviation, *LQ* lower quartile, *UQ* upper quartile, *Min* minimum, *Max* maximum

[a] Determined by Mann-Whitney *U* test

In the analyses, the levels of trace elements in hair were dichotomized according to the median value of the concentration in hair. The trace elements analyzed in this study do not have any recommended values for the concentration in hair, and their data were not normally distributed. For that reason, it was the most appropriate choice to dichotomize the levels of them into two categories according to the median values.

Table 3 Adjusted odds ratios and 95% confidence intervals of the concentration levels of trace elements in hair for periodontitis

Elements	Adjusted odds ratio[a]	95% confidence interval	p^b
Germanium	7.115	2.025–24.997	0.002
Barium	0.943	0.288–3.091	0.923
Rubidium	0.836	0.283–2.465	0.745
Lithium	2.050	0.495–8.496	0.322
Nickel	2.094	0.662–6.624	0.209
Vanadium	1.180	0.395–3.527	0.767
Strontium	1.244	0.388–3.990	0.713
Tin	0.272	0.079–0.935	0.039
Titanium	2.206	0.700–6.955	0.177

[a] Adjusted for age, gender, household income, present smoking, present drinking, daily frequency of tooth brushing, use of floss or interdental brush, regular dental visit

[b] Determined by multivariate logistic regression

The higher concentration level of germanium in hair was significantly associated with the higher prevalence of periodontitis. There were no previous studies reporting that higher concentration of germanium in body can be a risk factor for periodontitis. Kopp et al. [22] investigated the genotoxic and cytotoxic properties of germanium using high-throughput screening (γH2AX assay) in two human cell lines (HepG2 and LS-174T) representative of target organs (liver and colon), but they could not find any toxicity of it. The toxic elements such as lead, cadmium, and mercury in body that can be considered as risk factors for periodontitis are likely to have the influence on the direct or indirect mechanism of periodontium destruction [20, 21, 26]. Hence, it is suggested that germanium may also influence the break-down of bone and connective tissues around the teeth.

Regarding tin, the toxicity has been reported by various studies as tin was widely exposed to human by canned food. Schäfer and Femfert [27] reviewed the studies related to the toxicity of tin and concluded that if a large amount of canned food is eaten daily over a long period, disturbances of gastric acid secretion and a reduction in iron absorption or heme metabolism cannot be excluded. Few studies on the nutritional effects of tin were reported [28, 29]. While Solomons et al. [30] reported the intestinal interaction of tin and zinc, they did not focus on the nutritional effect of tin but on the absorption of zinc. However, based on the result of this study that the concentration level of tin in hair was inversely associated with the prevalence of periodontitis, it can be suggested that tin may have the direct or indirect effect of inhibiting the initiation and progression of periodontitis.

Further studies are needed to test these hypotheses proposed by the results of this study.

There are a few limitations in this study. First of all, in this data, the concentration levels of trace elements were only measured in hair. Even though the concentration of minerals in hair has the advantage of easily measuring the amount of exposure to the minerals by diet and environment during a few months, multi-dimensional exposure measurements could be possible if we adopted various kinds of methods including food intake interview and plasma or urine analysis. Saliva can also be adopted as an appropriate source in further studies on the association between periodontitis and trace elements considering its easiness of sampling and treating.

Second limitation is that more potential confounders such as diet habits, medication, hair dyes, and dental prosthesis and restorations had not been included in this study. They need to be considered in further studies.

Another limitation is that the concentration of trace elements and peridontitis severity were not categorized into more levels due to relatively small sample size. If subdivision was possible, the dose-response relationships between trace elements and periodontitis could have been investigated. Further studies need to be performed based on the larger samples.

Finally, it is not possible to determine the direction of causal relationship between germanium and tin concentration levels in hair and periodontitis since this study is a cross-sectional study.

Nevertheless, within our knowledge, the association was firstly found between the concentration level of germanium and tin in hair and periodontitis after adjusting for various potential confounders including sociodemographic variables and health behavioral factors.

In conclusion, there was a significant relationship between periodontitis and the concentration level of germanium and tin in hair.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Williams RC (1990) Periodontal disease. N Engl J Med 323:373–382
2. Albander JM, Bunelle JA, Kingman A (1999) Destructive periodontal disease in adults 30 years of age and older in the United States, 1988-1994. J Periodontol 70:13–29
3. Petersen PE, Ogawa H (2012) The global burden of periodontal disease: towards integration with chronic disease prevention and control. Periodontol 60:15–39
4. Korea Centers for Disease Control and Prevention (2010) Korea National Health and Nutrition Examination Survey. https://knhanes.cdc.go.kr/knhanes/index.do. Accessed 10 Sep 2014
5. Seymour GJ, Ford PJ, Cullinan MP, Leishman S, Yamazaki K (2007) Relationship between periodontal infections and systemic disease. Clin Microbiol Infect 13:3–10
6. Persson GR, Persson RE (2008) Cardiovascular disease and periodontitis: an update on the associations and risk. J Clin Periodontol 35:362–379
7. Hung HC, Willett W, Merchant A, Rosner BA, Ascherio A, Joshipura KJ (2003) Oral health and peripheral arterial disease. Circulation 107:1152–1157
8. Janket SJ, Baird AE, Chuang SK, Jones JA (2003) Meta-analysis of periodontal disease and risk of coronary heart disease and stroke. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 95:559–569
9. D'Aiuto F, Nibali L, Parkar M, Patel K, Suvan J, Donos N (2010) Oxidative stress, systemic inflammation, and severe periodontitis. J Dent Res 89:1241–1246
10. Baltacıoğlu E, Yuva P, Aydın G, Alver A, Kahraman C, Karabulut E, Akalın FA (2014) Lipid peroxidation levels and total oxidant/antioxidant status in serum and saliva from patients with chronic and aggressive periodontitis. Oxidative stress index: a new biomarker for periodontal disease? J Periodontol 85:1432–1441
11. Bullon P, Morillo JM, Ramirez-Tortosa MC, Quiles JL, Newman HN, Battino M (2009) Metabolic syndrome and periodontitis: is oxidative stress a common link? J Dent Res 88:503–518
12. Allen EM, Matthews JB, O'Connor R, O'Halloran D, Chapple IL (2009) Periodontitis and type 2 diabetes: is oxidative stress the mechanistic link? Scott Med J 54:41–47
13. Trivedi S, Lal N, Mahdi AA, Mittal M, Singh B, Pandey S (2014) Evaluation of antioxidant enzymes activity and malondialdehyde levels in chronic periodontitis patients with diabetes. J Periodontol 85:713–720
14. Lundberg JO, Weitzberg E, Gladwin MT (2008) The nitrate-nitrite-nitric oxide pathway in physiology and therapeutics. Nat Rev Drug Discov 7:156–167
15. Varela-López A, Giampieri F, Bullón P, Battino M, Quiles JL (2016) A systematic review on the implication of minerals in the onset, severity and treatment of periodontal disease. Molecules 21:E1183
16. Nishida M, Grossi SG, Dunford RG, Ho AW, Trevisan M, Genco RJ (2000) Calcium and the risk for periodontal disease. J Periodontol 71(7):1057–1066
17. Meisel P, Schwahn C, Luedemann J, John U, Kroemer HK, Kocher T (2005) Magnesium deficiency is associated with periodontal disease. J Dent Res 84:937–941
18. Petti S, Cairella G, Tarsitani G (2000) Nutritional variables related to gingival health in adolescent girls. Commun Dent Oral Epidemiol 28:407–413
19. Shoji K, Ohtsuka-Isoya M, Shimauchi H, Shinoda H (2007) Effects of lactation on alveolar bone loss in experimental periodontitis. J Periodontol 78:152–156
20. Kim Y, Lee BK (2013) Association between blood lead and mercury levels and periodontitis in the Korean general population: analysis of the 2008-2009 Korean National Health and Nutrition Examination Survey data. Int Arch Occup Environ Health 86:607–613
21. Won YS, Kim JH, Kim YS, Bae KH (2013) Association of internal exposure of cadmium and lead with periodontal disease: a study of the Fourth Korean National Health and Nutrition Examination Survey. J Clin Periodontol 40:118–124

22. Kopp B, Zalko D, Audebert M (2017) Genotoxicity of 11 heavy metals detected as food contaminants in two human cell lines. Environ Mol Mutagen. https://doi.org/10.1002/em.22157
23. Czerny B, Krupka K, Ożarowski M, Seremak-Mrozikiewicz A (2014) Screening of trace elements in hair of the female population with different types of cancers in Wielkopolska region of Poland. ScientificWorldJournal 2014:953181
24. Page RC, Eke PI (2007) Case definitions for use in population-based surveillance of periodontitis. J Periodontol 78:1387–1399
25. Miekeley N, de Fortes Carvalho LM, Porto da Silveira CL, Lima MB (2001) Elemental anomalies in hair as indicators of endocrinologic pathologies and deficiencies in calcium and bone metabolism. J Trace Elem Med Biol 15:46–55
26. Han DH, Lee HJ, Lim S (2013) Smoking induced heavy metals and periodontitis: findings from the Korea National Health and Nutrition Examination Surveys 2008-2010. J Clin Periodontol 40: 850–858
27. Schäfer SG, Femfert U (1984) Tin—a toxic heavy metal? A review of the literature. Regul Toxicol Pharmacol 4:57–69
28. Yokoi K, Kimura M, Itokawa Y (1990) Effect of dietary tin deficiency on growth and mineral status in rats. Biol Trace Elem Res 24:223–231
29. Schwarz K, Milne DB, Vinyard E (1970) Growth effects of tin compounds in rats maintained in a trace element-controlled environment. Biochem Biophys Res Commun 40:22–29
30. Solomons NW, Marchini JS, Duarte-Favaro RM, Vannuchi H, Dutra de Oliveira JE (1983) Studies on the bioavailability of zinc in humans: intestinal interaction of tin and zinc. Am J Clin Nutr 37: 566–571