ORIGINAL ARTICLE

Identification of Lead Exposure Through Saliva and the Occurrence of Gingival Pigmentation at Fuel Station Indonesian Officers

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Abstract

Objective: To identify the exposure of lead through saliva and the presence of gingival pigmentation in the risk group. **Material and Methods:** The type of this research is analytic observational with cross-sectional study design. The sample consisted of 40 subjects, who were divided into two groups of 20 each: G1: Gasoline Fuel Station and G2: Officer in Dental Hospital. To determine the level of leads in saliva is measured by atomic absorption spectrophotometry and assessment of the severity of lead-gingival lead line in this study based on the area of the gingival surface based on the number of dental areas. The data were analyzed with the t-test and Chi-square test. The level of significance was set at 5%. **Results:** The average lead content in saliva group exposed was 6.66 μg / ml and control group 4.72 μg / ml (p≤0.05). There is a correlation between gingival lead line incidence with exposure to lead (OR = 3.33; p = 0.001). **Conclusion:** The identification of exposure to lead poisoning can be determined by examination in saliva and ginggiva state, it is proven that the risk of gingival lead (Ginggival Lead Line) in the worker group at the gas station station is 3.3 times more risk than the control group. Occupational safety factors are important for workers at high risk of exposure to the element of lead for additional education on the importance of using masks to prevent the severity of the occurrence of lead effects on overall health.

Keywords: Diagnosis, Oral; Saliva; Gingiva; Pigmentation; Lead.

Introduction

The teeth and gingiva affect a person's appearance; the color of the gingiva is affected by the thickness of the gingival epithelium, the degree of keratinization and pigment in the gingival. Main pigments such as Melanin, carotene, hemoglobin, and oxy-hemoglobin contribute to the normal color of the oral mucosa [1]. The color change in the gingiva can be caused by various lesions and conditions associated with some endogenous and exogenous factors. Eg Heavy metals: lead, bismuth, mercury, silver, arsenic, and gold [2,3]. Lead is known to interfere with the health of oral tissue through several cellular and molecular pathways. Lead can alter humoral and cell-mediated immunity, and recent evidence suggests that lead exposure may affect the regulation of inflammatory cytokines in high-risk worker groups, such as workers at gas stations [4].

High-risk jobs with lead exposure from gasoline and airborne dust are gas station workers. With an 8 hour work time each day, lead exposure times tend to be higher when compared to workers who are not working gas station stations. Moreover, if workers do not use personal protective equipment such as masks that have small pore size and gloves.

Generally, the examination of leads to blood plasma examination, but some studies that serve as an objective indicator of detection, risk assessment, diagnosis, prognosis, and disease monitoring. Such biomarkers may be hormones (cortisol), toxins, enzymes (amylases), immunoglobulins (IgA), DNA, and heavy metals (leads))[5-8].

The level of lead in the blood plasma is divided into two components; namely, un-diffused base bus protein leads and unbound diffusible leads that are directly excreted in the saliva. The lead level in the saliva comprises 15-50% of the total blood count and there is an unbound form in the saliva [9,10].

Prolonged exposure to lead in the oral cavity can be identified by the presence of gingiva lines or Burtonian lines. Gingival Lead Line (GLL) is a condition characterized by gingival pigmentation in the form of a grayish blue line, with a width of 1 inch in the gingiva. Pigmentation occurs due to the reaction between bacteria depositing sulfide in the gingival and tooth border. The lead that is also carried on the gingival blood vessels will increase capillary permeability due to constant inflammation. Sometimes it looks like a bluish black line on the gingival border and teeth, especially in the maxillary anterior region [11,12].

Lead can be sourced from nature as well as the result of human activities. The presence of leads in the environment increases with human activity. The sources of lead pollution can come from industrial sectors such as paint industry, battery, battery, printing (ink), the cable industry, gilding, pesticides, household appliances, and fuel. Fuel is the source of lead pollutants in the environment because the use of Pb as an octane booster often emits lead to the environment, which can further be absorbed in the body through inhalation or penetration through the skin [4].

Excess lead content may be toxic to the human body as it interferes with hemopoietic activity, the nervous system, the urinary system, the gastrointestinal system, the cardiovascular system, the reproductive system, the endocrine system, as well as disrupting the musculoskeletal

system such as bones and teeth. In addition, lead can also alter humoral immunity and mediated cells, and recent evidence suggests that exposure may influence inflammatory cytokine regulation in workers exposed at work. Furthermore, lead can induce oxidative stress in a number of tissues and organs of the body, including salivary glands. These lead effects are associated with dental caries and periodontitis, which are further attributed to tooth loss [13].

Therefore, the aim of this study was to identify the exposure of lead through saliva and the presence of gingival pigmentation in these individuals.

Material and Methods

Study Design

The type of this research is analytic observational with cross-sectional study design. A research on saliva lead and lead-gingival lead line analysis has been conducted at the petrol station in Makassar. This research was conducted from November to December 2017 and got 40 research subjects consisting of 20 people as test group (G1: Gasoline Fuel Station) and 20 people as a control group (G2: officer in Dental Hospital Faculty of Dentistry Hasanuddin University).

Assessment Criteria

Assessment of lead levels in saliva is done with the help of Atomic Absorption Spectrophotometry. Assessing of the severity of lead-gingival lead line in this study based on the area of the gingival surface based on the number of dental areas as follows: Score 0 = There is no gingival lead line (normal category); Score 1 = There are gingival lead lines on 1-2 teeth (light category); Score 2 = There are gingival lead lines on 3-4 teeth (medium category); Score 3 = There are gingival lead lines on> 4 teeth (weight category).

Data Collection

Researchers come to the Gasoline Fuel Station and Dental Hospital to provide an explanation of the procedures, intentions and objectives of the research to be undertaken and then they fill out a inform consent letter to be the subject of the study. Interviews were conducted on the relevant subject matter, gender, length of service, and use of personal protective equipment (masks and gloves).

Saliva Sampling Procedure

Prior to salivary sampling, subjects were instructed not to consume the food for the previous hour. After that, the subjects were instructed to rinse with distilled water for 30 seconds. Saliva sampling technique without stimulation was done in the morning at 09.00-11.00 with passive drooling method ie the subject of passive salivary flow as much as 6 ml into the plastic tube. Assessment of lead levels in saliva is done with Atomic Absorption Spectrophotometry 4100 MP-

AES (Agilent Technologies Inc., Santa Clara, CA, USA) with a wavelength of 217 nm in Soil Laboratory, South Sulawesi Agricultural Technology Assessment Institute.

Procedure of Examination of Gingival Lead Line

Each anterior gingiva is cleaned first with a cotton swab to remove attached saliva. The maxillary and mandibular labial mucosa are retracted with a mirror, then visually observed when there is a white/blue line on the gingival and tooth mucosal boundaries in accordance with the assessment criteria.

Data Analysis

Data were analyzed using IBM SPSS Statistics for Windows Software, version 22 (IBM Corp., Armonk, NY, USA). Descriptive statistics were used to calculate the absolute and relative frequencies, mean and standard deviation. To know the difference of lead level between test group and control group use t-test and to know the relationship and risk of gingival lead line with Chi-square. The level of significance was set at 5%.

Ethical Aspects

This research has a recommendation of ethical approval from Dental Hospital of Faculty Dentistry Hasanuddin University with number 0009 / PL.09 / KEPK FKG-RSGM UNHAS / 2018.

Results

Table 1 shows the distribution of demographic and clinical characteristics. Male subjects were more prevalent (68%) and the age group 21-30 years was more frequent (67%). With regard to levels of lead in saliva, the ≤ 5 µg / ml category was found in the test group (20%). Based on the criteria of gingival lead line score, in the test group, the most weight category was found (55%).

Table 1. Distribution of demographic and clinical characteristics.

Variables	Test Group		Control Group		Total		Mean (SD)
	N	%	N	%	N	%	
Gender							
Male	19	95.0	8	40.0	27	67.0	
Female	1	5.0	12	60.0	13	33.0	
Age							27.7 ± 6.75
21-30 Years	10	50.0	17	85.0	27	67.0	
31-40 Years	8	40.0	3	15.0	11	27.0	
> 41 Years	2	10.0	0	0.0	2	6.0	
Salivary Lead Levels							6.16 ± 1.25
≤ 5 µg/ml	4	20.0	9	45.0	13	32.5	
> 5 µg/ml	16	80.0	11	55.0	27	67.5	
Work Experience							5.62 ± 2.7
< 5 Years	10	50.0	-	-	10	50.0	
> 5 Years	10	50.0	-	-	10	50.0	

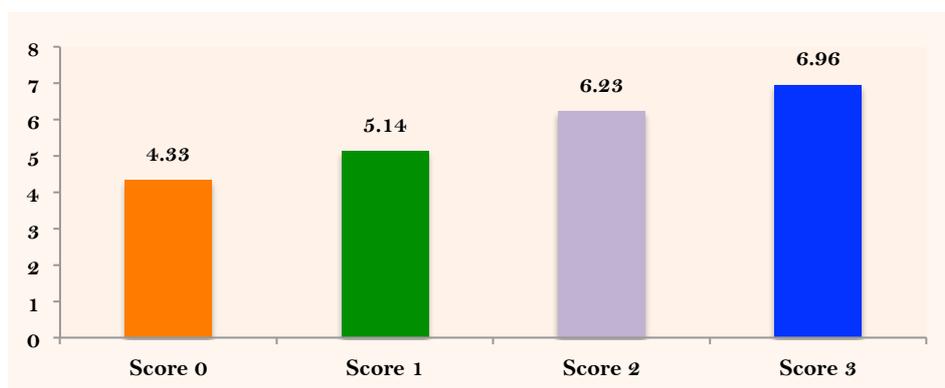| Gingival Lead Line Score						
0 (Normal)	0	0.0	14	70.0	14	35.0
1 (Light)	7	35.0	5	25.0	12	30.0
2 (Medium)	2	10.0	1	5.0	3	7.5
3 (Weight)	11	55.0	0	0.0	11	27.5

Table 2 shows the difference in mean saliva lead values based on test and control group variables, duration of work and lead line gingival scores. In the test group, the average value of salivary lead level was 6.16 µg / ml, with statistically significant difference (p<0.05). Based on the length of work, the average value of the worker with the working duration is 5 years of 5.3 µg / ml, with statistically significant difference (p<0.05). There was a significant difference in the lead rates in subjects based on gingival lead line findings (p<0.05).

Table 2. Differences in mean saliva lead values, work experience and lead line gingival.

Variables	Saliva Lead Level (µg/ml)		p-value[*]
	Mean	SD	
Group			
Test	6.16	1.25	0.030
Control	4.72	1.26	
Work Experience			0.045
≤ 5 Years	5.3	0.9	
> 5 Years	6.5	1.2	
Gingival Lead Line			0.001
Yes	6.03	1.2	
No	4.33	1.0	

[*]t-test.

Figure 1 shows the mean saliva lead level diagram based on the severity of lead-gingival severity. The highest salivary lead was found in subjects with a lead-gingival lead-3 (weight) score with a mean grade of 6.9 µg / ml.

Figure 1. The mean saliva lead level in saliva-based on the gingival lead-line severity score.

Table 3 shows the difference between lead-gingival scores based on the length of the work in the test group. In the test group that worked ≤ 5 years, the most frequent gingival lead line score was experienced in the light category was 4 (20%). However, for the test group that has been

working for> 5 years, the highest gingival lead line score in the medium and heavy category was 55% (p<0.05). The OR score was 7.33 (95% CI = 0.877-61.3320) indicating that the subjects who worked > 5 years were 7.33 times more likely to have moderate and large leads gingival leads when compared with subjects who worked ≤5 years.

Table 3. Differences in gingival lead line scores based on length of work in the test group.

Work Experience	Score Gingival Lead Line		Total	p-value*	OR	95%CI
	Light	Medium & Weight				
	N (%)	N (%)	N (%)			
≤ 5 Years	4 (20.0)	2 (10.0)	6 (30.0)	0.052	7.33	0.877-61.332
> 5 Years	3 (15.0)	11 (55.0)	14 (70.0)			
Total	7 (35.0)	13 (65.0)	20 (100.0)			

*Chi-square test.

Table 4 shows the differences in gingival lead line findings. In the control group most found subjects who did not experience gingival lead line as many as 14 people (35%). While in the test group most found the subjects who experience gingival lead line that is as many as 20 people (50%). In addition, there is also a p-value <0.05 which leads findings in the control group and test group. The OR score was 3.33 (95% CI = 1.707-6.511) indicating that the test group was 3.33 times more likely to have gingival lead line compared with the control group.

Table 4. Differences in gingival lead line findings by test and control group.

Group	Gingival Lead Line		p-value*	OR	CI (95%)
	Yes	No			
	N (%)	N (%)			
Exposure	20 (50.0)	0 (0.0)	0.001	3.33	1.707-6.511
Control	6 (30.0)	14 (70.0)			

*Chi-square test.

Discussion

The lead material contained in gasoline may be inhaled directly through the vapor or from the accumulation of motor vehicle combustion results in the form of dust/particulates in the air. The increasing number of vehicles and traffic congestion will eventually lead to exhaust emissions from motor vehicles. High-risk jobs with exposure to lead from gasoline vapors and particulate dust in the air are gas station attendants. With a working time of 8 hours every day, the lead-time for exposure to lead tends to be higher when compared with workers who do not work at gas stations. Moreover, if the worker does not use personal protective equipment such as a mask with small pore size and gloves.

Saliva is a heterogeneous biological liquid composed of water of 99%, protein of 0.3%, and an inorganic substance of 0.2%. In addition, there are also biomarkers in the saliva that serve as an objective indicator of detection, risk assessment, diagnosis, prognosis, and disease monitoring [14,15]. Such biomarkers may be hormones (cortisol), toxins, enzymes (amylases), immunoglobulins (IgA), DNA, and heavy metals (lead) [6,8,15,16].

The level of lead in the blood plasma is divided into two components; namely, un-diffused base bus protein leads and unbound diffusible leads that are directly excreted in the saliva. The lead level in the saliva comprises 15-50% of the total blood count and there is an unbound form in the saliva [13].

In addition to such examinations, clinically chronic exposure due to lead in the oral cavity can be identified through the presence of a lead or Burtonian line gingival line. The gingival lead line is a condition characterized by pigmentation of the gingiva in the form of a grayish blue-white line, with a width of ± 1 inch in the gingiva [7]. The pigmentation occurs in the arena of reaction between lead in the circulation with sulfur ions released by the activity of bacteria depositing lead sulfide (PbS) on the gingival and dental borders. The lead that is also carried on the gingival blood vessels will increase capillary permeability due to constant inflammation. The lead line has a characteristic of a blue-purple or gray line on the gingival tissue. Sometimes it looks like a bluish black line on the gingival border and teeth, especially in the maxillary anterior region. Lead lines appear when poor oral hygiene results in subepithelial sulfide granule deposition and is released by microorganisms derived from protein deposit deposits [17].

Lead can be sourced from nature as well as the result of human activities. The presence of leads in the environment increases with human activity [18]. The sources of lead pollution can come from industrial sectors such as paint industry, battery, battery, printing (ink), the cable industry, gilding, pesticides, household appliances, and fuel. Fuel is the source of lead pollutants in the environment because the use of Pb as an octane booster often emits lead to the environment, which can further be absorbed in the body through inhalation or penetration through the skin.

Excess lead content may be toxic to the human body as it interferes with hemopoietic activity, the nervous system, the urinary system, the gastrointestinal system, the cardiovascular system, the reproductive system, the endocrine system, as well as disrupting the musculoskeletal system such as bones and teeth. In addition, lead can also alter humoral immunity and mediated cells, and recent evidence suggests that exposure may influence inflammatory cytokine regulation in workers exposed at work. Furthermore, lead can induce oxidative stress in a number of tissues and organs of the body, including salivary glands. These lead effects are associated with dental caries and periodontitis, which are further attributed to tooth loss.

Detecting as much as possible can minimize the danger of lead poisoning. However, this procedure tends to be invasive, the collection of samples requires specialist expertise and costs are quite expensive. Therefore, biomonitoring through saliva has been used to monitor environmental pollutants, since they can be distributed in salivary glands, which are reflected in saliva. The concentration of these substances in saliva depends on the nature of the material and its transport process. Many reports show the distribution of leads in salivary glands and diffusion into saliva in high concentrations [1].

This research was conducted with the aim to know the level of lead saliva in a vulnerable group that is SPBU officer in Makassar and compared with a healthy group that is RSGM FKG

UNHAS officer. In addition, this study was also carried out in terms of salivary lead levels based on the length of work and gingival lead lines.

Based on the result of data analysis it is found that there was a significant difference (p<0.05) between the mean values of the control group saliva lead level (RSGM FKG UNHAS) with the test group (Petrol Station Officer in Makassar). A previous study found that there was a significant difference in salivary lead levels between healthy populations (average 3.5 μg / ml) with the exposed population (average 5.3 μg / ml) in Iraqi Baghdad [8]. However, others researchers also found that there was a significant difference in salivary lead levels between healthy children (mean 0.90 μg / ml) with children exposed to lead (mean 2.20 μg / ml) in Malaysia [19]. There is a weak correlation between salivary lead levels and blood lead levels so the validity is still debated as an alternative biological monitor of blood substitutes [13].

In the future, assessment of saliva leads and gingival lead line as an alternative to early detection of lead poisoning as a standard in determining lead poisoning as early as possible.

Conclusion

The risk of exposure to Leaders in the larger gas stations leads to gingival lead line 3.3 times compared to the control group. Occupational safety factors are important for workers at high risk of exposure to the element of lead for additional education on the importance of using masks to prevent the severity of the occurrence of lead effects on overall health.

Conflict of Interest: The authors declare no conflicts of interest.

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