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## THE EXPOSURE OF Pb TO HAIR AND NAILS IN CHILDREN AROUND "X" COAL MINES USING ATOMIC ABSORPTION SPECTROSCOPY (AAS) METHOD

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### Abstract

Introduction: The exposure of Pb from coal mining activity could be accumulated primarily in the hair and nails. It was a presence as a bioindicator of Pb contamination in the community. Children who lived around coal mining were also likely to experience contamination of Pb. The children could absorb up to 50% of Pb metal. Methods: This study used a cross-sectional design. The sample was taken using cluster sampling and classified based on the age group, namely 1-3 years, 4-6 years, and 7-9 years. Exposed respondents lived around 1-10 kilometers from the mining area, and the non-exposed respondents lived outside. Some hair and nails would be taken from all respondents, which were sent to a laboratory directly. Atomic Absorption Spectroscopy was used to measure the levels of Pb. Results and Discussion: This study showed that Pb levels in the hair of exposed respondents in all age categories were above the WHO threshold limits value (≤12µg/g). In addition, in nails, Pb levels exceed the threshold limits value in the exposed sample groups 4-6 years and 7-9 years. Differences in Pb levels in the hair and nails between exposed dan non-exposed respondents showed significantly by the statistical test. Conclusion: Communities living in coal mining areas are at greater risk of exposure to heavy metals than others. Pb is one type of heavy metal that accumulates in the body, specifically in hair and nails, because it did not release by metabolism. Hence, it becomes a bioindicator to ensure our entire body exposure to hazardous materials.

### INTRODUCTION

Coal is the main resource needed in various industries, such as steam power plants, cement, fertilizer, paper, briquettes, and textiles (1). The demands have greatly influenced the development of coal mining production in Indonesia. Data from the Directorate General of Mineral and Coal, overall coal production has been increased an average by 11% over the last 13 years. Because of the high demand for coal, the mining was carried out neither by large companies nor small companies in the upstream rivers. Furthermore, it will cause environmental pollution by the side products (2). The study in Pangkalan Kuansing Village around the coal mine of PT. TBS shows a heavy metal content of lead (Pb) in the community water wells, approximately 2.93 mg/L (3). It is in line with the study in the Bandung Regency that there is a contamination of Pb in the ambient air from coal user industrial locations up to 5.87  $\mu g/m^{3}(4)$ .

Pb is a type of neurotoxin that causes various health problems such as disordering the nervous system and intelligence in children, increasing the risk of miscarriage, damaging the organ system, increasing blood pressure, and poisoning (5). Children exposed to Pb in a short period can impact the prefrontal cerebral cortex, hippocampus, and cerebellum. There are three basic ways Pb enters the body, namely, inhalation, skin contact, and ingestion. Most of the Pb that enters the body will be absorbed in the blood, a small portion accumulates in the body tissues (kidneys, hair, and nails), and the rest will be excreted through urine and feces (6). The presence of Pb in people's hair and nails can be used as an indicator that pollution occurred in the community (7).

The study conducted in Bengkulu shows that the exposure of Pb in coal waste collectors communities is present that the levels of Pb in the hair are between 0 -206.3 µg/g since they have been working in 3 - 8 years (6). Moreover, the study in PT Bukit Asam Kertapati Jetty Unit shows a linear correlation between the hair levels of Pb with the increase of blood pressure in workers (8). These results indicate that the community around the coal mining has a risk of being exposed to Pb, especially in children. Children might have a potential health risk greater than adults because they absorb up to 50%, while adults in 10-15%, from the exposure of Pb to the body (9). A study in Italy shows the Pb finds in children's hair approximately 13.27  $\pm$  1.7  $\mu$ g/g that live close to mining sites (10). Similar results reports from the largest coking plant in China show that children who live close to it are more susceptible to poisoning due to Pb exposure (11). These results indicate that children are more likely to be exposed to Pb.

Furthermore, the Pb exposure for a long time will decrease intelligence, behavior, memory, and brain volume in adulthood (12). So, it can conclude that study on Pb exposure in children is very important to do. Our study will measure and analyse the Pb exposure levels on the hair and nails from children who lived around the coal mining in the "X" mine using the atomic absorption spectroscopy (AAS) method.

### **METHODS**

This study used a cross-sectional design with an exposed and non-exposed group. The respondent in this study was taken using cluster sampling, which was classified by age group according to the previous study, namely 1-3 years, 4-6 years, and 7-9 years (13). The criteria for the exposed group whom children who lived around 1-10 kilometers from the "X" coal mine. Then, the non-exposed group was lived outside the mine for comparison. The total number of the respondent was 24 children who agreed with confirming the informed consent. Some parts of hair and nails will be taken from each respondent then analyse using the atomic absorption spectroscopy (AAS) method in Surabaya Industrial Research and Consulting Center.

The AAS method procedures were used in our study modified from previous research (14), including preparation of mains standard solution, preparation of standard serial solutions, wash hair and nails, wet digestion, and preparation of sample solutions. Preparation of the Mains Standard Solution (100 ppm) was carried out by dissolving 1.5985 grams of Pb (NO $_3$ ) $_2$  with 10 mL of HNO $_3$ , then diluted with 1L distilled water. The next step was to make standard serial solutions such as 0.1 ppm, 0.2 ppm, 0.3 ppm, 0.4 ppm, 0.5 ppm that were carried out by taking Mains liquor 100 ppm then diluted with 1% HNO $_3$  to 100 mL. Each solution was then measured its absorbance with AAS.

Sample preparation was carried out by washing the hair and nails. They were weighed 0.5 grams each and put into a glass beaker, then soaked in 10 mL of acetone for 15 minutes. The results were rinsed with distilled water and dried in an oven at 105°C for 2 hours, then calculated. The hair and nail samples were then subjected to wet destruction. Wet digestion was carried out by adding 15 mL of 65% HNO $_3$  to each sample were washed and dried before and then heated until the redbrown acid disappears. The result was added 5 mL of 30%  $\rm H_2O_2$  while still carrying out the destruction process until a clear solution was obtained. This solution was then cooled and diluted with distilled water to 50 mL.

In the final step, all finished samples were measured by AAS to obtain the absorbance of Pb.

The concentration of each sample is calculated by entering the absorbance value of the sample in the standard series linear curve equation. The Pb levels in hair and nails were then compared with the threshold limits value of Pb in hair and nails by WHO in ≤ 12µg/g.

In this study, data analysis was carried out by statistical tests using the IBM SPSS Statistics 25 application. The statistical tests began with the normality and homogeneity tests. A one-way ANOVA and a Tukey follow-up test were performed to compare all pairs of the Pb ratio in each group.

### RESULTS

The results of Pb measurement on children's hair and nails showed that the exposed group had higher levels than the non-exposed group, detailed in Table 1. In addition, the older child also had higher Pb levels than others. Figure 1 showed that the Pb levels in hair in all age categories and nails for age 4-6 years and 7-9 years exceeded WHO's threshold limit values.

The results of the analysis of normality, homogeneity, and one-way ANOVA levels of Pb in the hair and nails of children around the "X" coal mines are shown in Table 2. The-way ANOVA test showed a significant difference in Pb levels in the hair and nails of the exposed group.

Table 1. Pb Levels in Hair and Nails of Children Around The "X" Coal Mines

Group	<mark>Pb</mark> Levels in Hair (μg/g)	Pb Levels in Nail (μg/g)
2	Mean ± SD	Mean ± SD
0 – 3 Years	$14.8856 \pm 1.0036$	$11.2998 \pm 0.9377$
4-6 Years	$19.8396 \pm 0.9378$	$14.1543 \pm 0.9554$
7-9 Years	$24.2864 \pm 1.4749$	$16.9851 \pm 0.9554$
Control	$10.6275 \pm 0.5988$	$9.1177 \pm 0.2838$

Table 2. Analysis of Normality, Homogeneity, and One Way ANOVA Levels of Lead in Children's Hair and Nails Around Coal Mine "X"

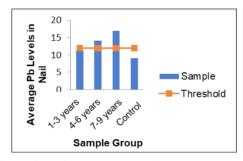
Test -	Sig. (2-tailed)	
	Hair Samples	Nail Samples
Shapiro-wilk	0.799	0.578
Homogeneity	0.149	0.149
One way ANOVA	0.000	0.000

Table 3. Tukey Test Results on Children's Hair Samples Around Coal Mine "X"

(I) Hair	(J) Hair	Sig.
1-3 Years	4-6 Years	0.002
	7-9 Years	0.000
	Coppol	0.009
	1-3 Years	0.002
4-6 Years	7-9 Years	0.003
	Control	0.000
7-9 Years	1-3 Years	0.000
	4-6 Years	0.003
	Control	0.000
Control	1-3 Years	0.009
	4-6 Years	0.000
	7-9 Years	0.000

Table 4. Tukey Test Results on Children's Nail Samples Around Coal Mine "X"

(I) Nail	(J) Nail	Sig.
2 1-3 Years	4-6 Years	0.006
	7-9 Years	0.000
	Corprol	0.050
	1-3 Years	0.006
4-6 Years	7-9 Years	0.012
	Control	0.000
	1-3 Years	0.000
7-9 Years Control	4-6 Years	0.012
	Control	0.000
	1-3 Years	0.050
	4-6 Years	0.000
	7-9 Years	0.000



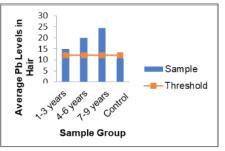


Figure 1. Comparison of Average Pb Levels Among Respondents Group

### DISCUSSION

Pb exposure in children and adulthood is very important to analyze because it will affect their intelligence, behavior, memory, and brain volume in the longterm exposure (12). While, a short period of Pb exposure might be impacted to the prefrontal cerebral cortex, hippocampus, and cerebellum. A study in Egypt also reported that children who have high Pb levels in their hair could increase the risk of ADHD (Attention-Deficit Hyperactivity Disorder) (13).

The bioaccumulation process of Pb through the body will deposit in the liver, hair, and nails that became the bioindicators to show any indication that Pb metal contamination has occurred in the environment (14). The study in China stated that hair's Pb levels could accurately facilitate the Pb poisoning diagnostic (15). The measurement of Pb in hair is the simplest and most effective method of screening for Pb poisoning. So, it has more advantages over blood and urine sample analysis, reflecting current and recent.

Table 1 showed that the exposed group tyho reside close to the "X" coal mine have higher levels of Pb in their hair and nails than the non-exposed group. This finding is ascertaining the indication that contamination of Pb has occurred around their environment. Our results also showed that the older the child was, the Pb levels in hair and nails would increase. The highest Pb level in the hair and nail sample was respectively 24.2864  $\pm$  1.4749  $\mu g/g$ , 16.9851  $\pm$  0.9554  $\mu g/g$ . The increasing age of children causes increased activities to do things such as holding objects and playing outside so that the children age eight years were found to have higher Pb levels than younger (16).

Our finding in the exposed group showed that only respondents in the 1-3 years category had Pb levels less than the WHO's threshold limits value with an average of 11.2998  $\pm$  0.9377  $\mu$ g/g. Because the children aged 1-3 years come down to the ground rarely, under parental supervision intensively, and apply the personal hygiene stricter than children aged above. Although this Pb level is fewer under the threshold value, it can not be stated safe. Furthermore, our results can indicate that the group of children is very susceptible to exposure Pb contamination. It is similar to the literature review In Developing and Developed Countries (Case Study in Children aged 0-18 years), which shows that children in all developing countries such as Brazil, India, China, and Indonesia are susceptible to Pb exposure then getting various diseases such as anemia and stunting (17).

Figure 1 also showed that the Pb level in the hair was higher than that in the nails. The coal mining

process produces many Pb pollutants released in the air so that most of their accumulation is in the hair. In addition, hair contains amino acids with disulfide bonds (-S-S-) and sulfhydryl groups (-SH), which can bind lead metal (Pb) strongly (6). A three-inch length of human hair sample can indicate what happened during the last 6 months, including Pb contamination that entered the body (18). Similar to a study conducted in Pekanbaru, which reported the highest Pb levels in petrol stations operator and retail gasoline sellers were found in hair (1.909 ppm) than in nail (0.275 ppm) (14). Pb metal pollutants around the coal mining might be highly adsorbed the skin, respiratory tract, water, and food as a pathway to enter the body, while the lack of personal hygiene makes it easier (8). Toxic heavy metals that enter the body will be circulated throughout the body and accumulate into the kidneys, liver, adipose tissue, and settles on the hair, nails, and teeth. The adverse effects of lead have been known for a long since metal affects practically all organs and systems of the human body. Then the metal is excreted through urine (19-20). The lead secretion process is generally slow, which is indicated by a long half-life, especially in hard tissue, which is about 25 years (21).

Aliterature review conducted in the United States showed that children have a higher risk of poisoning than adults (22). It is caused by children's behavior to bite and swallow anything that makes them easier to expose by pollutants. These results are also supported by a literature review conducted on African-American Black children from 1999 to 2010, which states that children have a higher risk of Pb poisoning in their growth periods (12). Inhaled Pb in humans will be stored in the lungs by 35-40%, and 95% will enter the bloodstream. On the other hand, this value will be higher in children, pregnant women, and people who are deficient in calcium, zinc, or iron (22-23).

The toxicokinetics mechanism of lead in the body will form bonds with sulfhydryl groups which inactivate glutathione then causes inhibition of the GSH formation. In addition, lead also deactivates the enzymes  $\delta$ -aminolevulinic acid dehydratase (ALAD), glutathione reductase (GR), glutathione peroxidase (GPX), glutathione-S-transferase, superoxide dismutase (SOD), and catalase (CAT). Furthermore, it will cause interference with antioxidant enzymes and interfere with the release of radical compounds (12).

As a precaution against lead metal exposure, especially in children, it is advisable to consume foods that contain mineral elements, flavonoids, and vitamins. These nutrients are used in preventive precautions

for body metabolism to increase toxicodynamics and toxicokinetics to excred the lead already present in the body. These nutrients also play an essential role in restoring the imbalanced prooxidant/oxidant ratio due to oxidative stress when exposed to heavy metals (15,23).

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### CONCLUSION

This study shows that the community around coal mining has the potential to be exposed to hearly metals, especially Pb. It is proven by the analysis of Pb levels in hair and nails in respondents around mining which have higher values than respondents outside mining. Such at-risk settlements must regularly close all ventilation access so that ambient air does not enter the house. In addition, it must also be very intensive to pay attention to children's activities outside the home.

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