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IDENTIFICATION OF LEAD CONTAMINATION IN THE FOOD CHAIN AND ENVIRONMENT SURROUNDING BREASTFEEDING MOTHERS IN HIGHLAND AGRICULTURAL AREAS

Dina Rahayuning Pangestuti[1]*, Apoina Kartini[1], Suhartono Suhartono[2], Naintina Lisnawati[1], Thomas Triadi Putranto[3], Novie Susanto[4]

[1]Department of Public Health Nutrition Faculty of Public Health,Universitas Diponegoro, Semarang 50275, Indonesia
[2]Department of Health Environment Faculty of Public Health, Universitas Diponegoro, Semarang 50275, Indonesia
[3]Department of Geological Engineering Faculty of Engineering, Universitas Diponegoro, Semarang 50275, Indonesia
[4]Department of Industrial Engineering Faculty of Engineering, Universitas Diponegoro, Semarang 50275, Indonesia

Corresponding Author:
*) dinapangestuti@lecturer.undip.ac.id

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Abstract

Introduction: *Lead in the environment can be more easily absorbed by individuals with nutritional deficiencies, particularly breastfeeding mothers residing in agricultural areas near lead sources. Contaminated breast milk can disrupt infants' growth and development. This study aims to identify lead contaminants in the environment and assess the nutritional status of breastfeeding mothers to provide preventive measures.* ***Methods:*** *This cross-sectional study was conducted in the highland agricultural area of Semarang Regency in October 2021. Environmental samples, including air from agricultural land, settlements, groundwater, and raw food were carried out as environmental samples from 31 breastfeeding mothers. Dietary intake, haemoglobin, MCV, MCH, MCHC, and lead levels in breast milk was carried out. Lead content was analysed using ICP-OES. Geographic information system (GIS) was used to compare spatial distribution lead status levels with identified exposure factors.* ***Results and Discussion:*** *Median age of breastfeeding mothers was 24 years, with 72% being housewives, having a senior high school education. Median age of the infants was 2.5 months, 32% being boys, and 48% were exclusively breastfed. Mean hemoglobin level of the mothers was 13 ± 1.4 g/dL (13.8% was anemic) and median lead level in breast milk was 0.019 ppm. None of the mothers met their recommended macro- and micronutrient intake. Lead content in foods was 0.02-0.180 ppm, groundwater 0.017-0.034 ppm, and air 0-1.56 µg/Nm3 over a three-hours.* ***Conclusion:*** *The environment surrounding breastfeeding mothers contains lead, particularly in the air, and these mother experiences nutritional deficiencies, thereby increasing the risk of lead absorption.*

INTRODUCTION

The presence of heavy metals, such as lead, is generally associated with emissions from residual fuel and industrial pollution. Although lead concentrations in fuel have decreased since 2006, a study found that lead was present in the air in samples from India, Hong Kong, and Indonesia (specifically in Bandung), especially in densely populated or urban areas (1). Despite this, the presence of lead in agricultural areas has received little attention, particularly in Indonesia. Research conducted in the shallot growing sector in Brebes Regency, Central Java, Indonesia, revealed that lead is found in pesticides commonly used by farmers (2). Another study has found that fertilizers used by farmers also contain lead (3).

Lead (Pb) is a heavy metal that is abundant in nature and cannot be degraded. Lead comes in a variety of forms, including elemental, inorganic, and organic, and all of them are poisonous. Inorganic lead is a type of lead found in old paint, dirt, dust, and other materials. Leaded gasoline contains organic lead, often known as tetraethyl lead. Organic types of lead are more hazardous because they are absorbed through the skin and are more poisonous to the brain and nervous system than inorganic lead (4). Lead exposure can have various health effects and symptoms depending on age, exposure level, and other health conditions. Although lead can

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be harmful to individuals of all ages, fetus, newborns, and young children are most at risk. It is commonly known that blood lead concentrations higher than 10 micrograms per decilitre can be harmful to a number of physiological systems. Symptoms include abnormally low haemoglobin, poor renal function, an increased risk of respiratory illnesses, and reduced cognitive abilities have been described in both adults and children (5–8). Lead exposure has also been shown to cause reductions in IQ and neurological disorders in vulnerable groups such as infants and children (9–10). Lead stored in the mother's bones can be transferred to the fetus during pregnancy. Moreover, breastfeeding mothers can contribute to lead exposure in infants through breast milk, particularly during the first six months of life (11). Growth retardation has been observed in infants with high lead levels after birth at the age of two years (12–14).

Lead absorption in the human is influenced by both internal and external factors. Internal factors include nutritional status, particularly calcium and iron levels (7–8,15). Micro and macronutrient deficiencies in the body can facilitate lead absorption, follow blood circulation, and interfere with body metabolism. External factors include our surroundings, specifically the air, water, and soil (dust) (16). Other external factors that contribute include sources of contaminants in food, such as drinking water, food or vegetables that absorb lead from pesticides, and the use of traditional herbs (17–20). However, no similar research on food commodities has been conducted in the agricultural area of Semarang Regency. This study aims to build on previous research, which found that breastfeeding mothers were exposed to lead and experienced micronutrient deficiencies (21–22). Therefore, this study aims to identify lead contamination in food and agricultural environments, as well as the home environment of breastfeeding mothers. We can limit efforts to improve the nutritional status of breastfeeding mothers exposed to lead through subsequent nutritional intake by investigating potential environmental lead contaminants.

METHODS

This research was conducted in October 2021, using an observational study cross-sectional design in the agricultural area of Sumowono District, Sumowono District and Banyukuning village, Bandungan Regency, Semarang Regency, Central Java Province, Indonesia. The study site is a highland region situated at an altitude of 650 to 1800 meters above sea level and known for its vegetable production. A total of 31 breastfeeding mothers participated in this study, meeting the criteria of having an infant aged between zero and six months, currently breastfeeding, and residing at the study site since their last pregnancy.

Interviews were conducted using a questionnaire to gather information on general characteristics, nutritional intake (24-hour recall), and potential risk factors for lead exposure in the surrounding environment. Maternal nutritional status was assessed by inputting the 24-hour dietary recall data into the NutriSurvey software to determine the percentage of nutrient adequacy based on Indonesia's Recommended Dietary Allowances (RDA) (23). Data were also collected on the coordinates of the mother's place of residence and the location where food was purchased. Blood tests were carried out including hemoglobin levels, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC). As much as 5 cc of venous blood was taken by a competent health analyst, placed in an EDTA tube, and then analyzed using a hematology analyzer in CITO laboratory, Semarang. Anemia status was defined by a hemoglobin level of less than 12 μg/dL, with microcytic anemia characterized by low levels of MCV (less than 80 fl), MCH (less than 27 pg), MCHC (less than 32 g/dL) (24).

Air samples were taken around residential areas of breastfeeding mothers based on villages and on nearby agricultural lands. Groundwater samples were collected from deep water wells around the nursing mother's house. Food samples include vegetables, fruit and herbs that are often consumed by breastfeeding mothers based on preliminary research. The location for sampling vegetables and fruit came from a market frequently visited by breastfeeding mothers.

The process for determining the lead content in food and water samples is based on the process outlined in the Indonesian National Standard for determining the upper limit for heavy metal contamination in food (SNI 7387:2009) (25–26). At least 100 grams of food were taken, while at least 100 ml of water and 50 ml of breast milk were taken. Food samples were placed into plastic sealed bag, while water samples were in sterile glass bottles, which had been washed clean and free of heavy metals by rinsing first with nitric acid: distilled water (1:1) and rinsing with distilled water. Breast milk sampling is carried out at least 50 ml of breast milk with a sterile manual breast pump after breastfeeding to take advantage of the let-down reflex from the mother (27). In this study, trained enumerators were involved in sampling. Breast milk sampling was carried out procedurally with aseptic principles to avoid the risk of contamination. The lead content in breast milk is then categorized as safe if it is less than 0.035 ppm (28–29).

Environmental analysis was carried out in the

Sumowono agricultural area using a Manual Haz Dust Environmental EPAM 5000 apparatus and a membrane filter (mixed cellulose ester) for samples of lead and other heavy metals (Whatman 0.45 m diameter 47 mm). Wind speed is measured using an anemometer and air humidity with a thermo-hygrometer. Air sampling was carried out in vegetable plantations and settlements around breastfeeding mothers and close to agricultural land. Sampling was carried out for three hours at each point due to weather constraints with moderate to high rainfall. Sample preparation prior to analysis was carried out in the laboratory at Centre for Industrial Pollution Prevention Technology Semarang City according to the characteristics of each sample. The analysis of all samples was carried out using the ICP OES tool with limit detection of 0.02 ppm.

Sampling of 12 ordinate points for geographic information systems GIS analysis was carried out at locations representing agricultural land (6 locations), settlements (5 locations) around breastfeeding mothers and market where mothers used to buy foods (1 location). The data needed in this analysis was the coordinates of the sample location in the form of latitude (X) and longitude (Y) coordinates. Map creation was assisted using arcGIS software. The analysis results are entered into the software to determine the point. Meanwhile, the air lead content obtained from the results of sample analysis in the laboratory is put into three levels for 3-hour measurements, and two levels for 24-hour interpolation results (based on the Regulation of the Indonesian Government Number 41 of 1999, which sets the national ambient air quality standard for lead metal at 2 g/Nm^3 for 24 hours).

The chi-square test was used to identify the association between the status of locations positive for lead in the air and the status of lead in breast milk with a significance level of 0.05. Enumerators have attended training related to the use of tools and recording coordinates. This research has obtained permission from the Ethics Committee of the Faculty of Public Health, Universitas Diponegoro with Decree No. 401/EA/KEPK-FKM/2021. All participant information was kept confidential and collected only after obtaining informed consent.

RESULTS

Most of mothers are housewives of reproductive age with upper secondary education, married to self-employed workers with incomes below the minimum wage (Table 1). None of mother fulfil the requirement for energy, protein, carbohydrate, fat, iron, calcium, vitamin A, folic acid, vitamin B1, 2, 6, magnesium, zinc. Only 16% of mothers met the recommended intake for vitamin C and P. Although in general the average maternal Hb level is normal (13±1.4 μg/dL), 13.8% of mothers experienced anemia and had a tendency to be classified as microcytic based on low levels of MCV, MCH, MCHC (Figure 1).

Table 1. General Characteristic of Breastfeeding Mother

Variables	n (%)	Median	Min.	Max.
Age (years old)		24	16	42
Housewives	18 (72)			
Low education	12 (48)			
Self-employed husband	20 (80)			
Low educational husband	16 (64)			
Family income (IDR)		2,000,000	1,000,000	6,000,000
Infant's age (months old)		2.5	0	6
Boy infant	8 (32)			
Parity (children)		1	1	4
Exclusively breastfed	12 (48)			

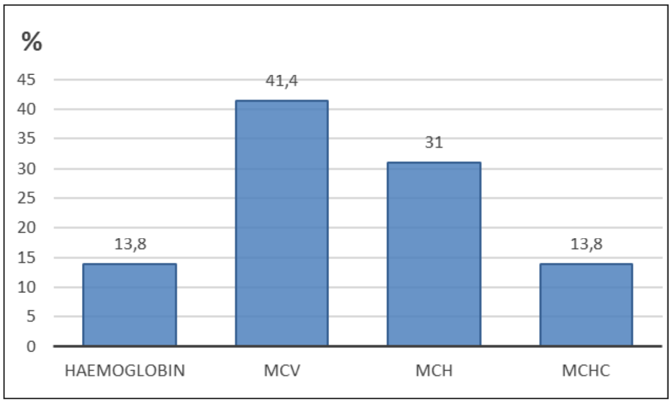


Figure 1. Percentage of Abnormal Blood Profiles Among Breastfeeding Mothers

Food samples harvested from agricultural lands around Semarang Regency and sold in the nearby market were used to assess the food system. Based on previous research, the food ingredients studied in this study were foodstuffs commonly consumed by breastfeeding mothers (unpublished data). Breastfeeding mothers in the study site primarily purchase food from the nearby traditional market (*Sumowono* market). The lead content of foods commonly consumed by breastfeeding mothers ranges from 0.02 to 0.180 ppm, with spinach having the highest concentration (Table 2).

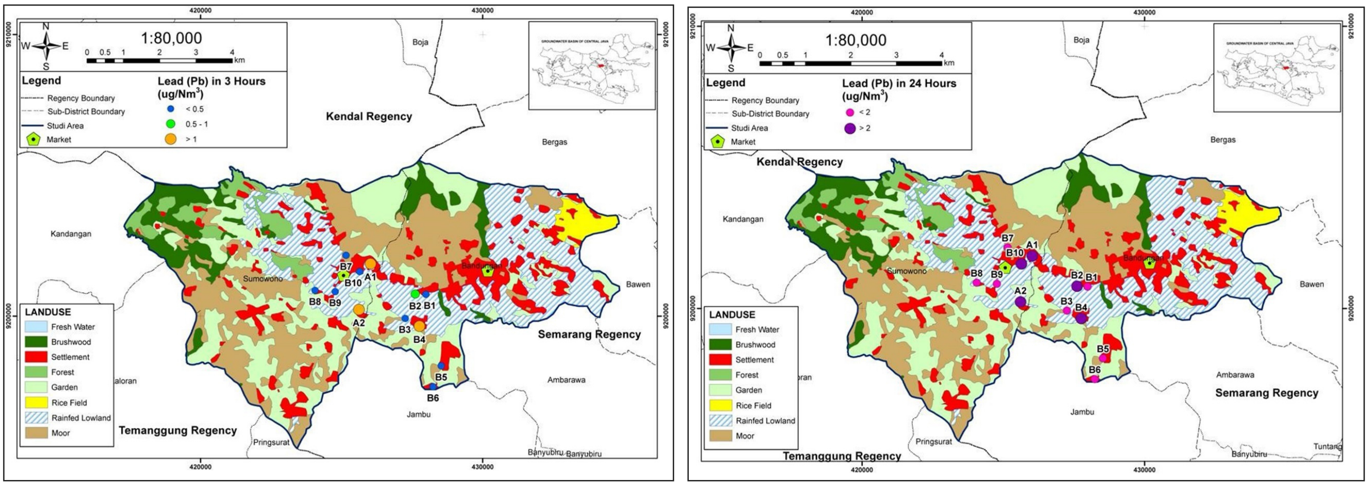
Table 2. Food Lead Levels Consumed by Breastfeeding Mothers at the Research Site

Food Item	Lead Content (ppm)
Tomato	<0.020
Spinach	0.180
Kale	<0.020
Chinese cabbage	<0.020
Pakcoy	0.08
Broccoli	<0.020
Cauliflower	<0.020
Tofu	0.02
Tempeh	<0.020
Banana	<0.020
Traditional herbal medicine	<0.020

Based on the Indonesian National Standard (SNI 7387: 2009) concerning the maximum limit of heavy metal contamination in food, it is stated that the lead content in foodstuffs in the category of fruits and vegetables as well as seeds and their derivatives the maximum limit of lead contained is 0.5 ppm. Meanwhile, based on the Codex Alimentarius Commission (CAC), the limit is 0.3 ppm. The lead level at the level of consumption in the form of drinking water also did not show the lead content exceeding the maximum limit (0.01 mg/L). Drinking water that is usually consumed is well water or spring water.

According to these findings, the groundwater in the Sumowono and Bandungan sub-districts had lead concentrations of 0.034 ppm and 0.017 ppm, respectively. These findings show that the content is considered safe because it is less than the 0.05 ppm upper limit specified in the Republic of Indonesia's Minister of Health's Regulation No. 416/MEN.KES/PER/IX/1990 on requirements and supervision of water quality. In light of the fact that locals rely on this groundwater source for clean drinking water (44%) of the respondence, its safety is crucial to investigate. Additionally, irrigation is needed for agricultural operations in the region, which is related to the potential for absorption into the vegetables and other plant products that are grown in the research area.

Based on the description of air lead levels performed on four agricultural lands, it demonstrates the presence of lead levels in the air. This is also influenced by agricultural activities that occur during the sampling period. The highest air lead content in one agricultural land, among others, is most likely caused by pesticide spraying activities near the location. However, it has been discovered that lead levels in the air have also been detected in settlement areas. The results are above the air lead content quality standard when interpolated into a 24-hour measurement, both on agricultural land and in residential areas (Figure 2) (Table 3).



(a) *(b)*

Figure 2. Air Lead Levels Measured For (a) Three Hours and (b) Assumed for Twenty-Four Hours in Agricultural and Settlement Areas

Table 3. Air Lead Levels in Agricultural and Settlement Areas at the Research Site

Type of Area	Farming Activities Nearby	Humidity (%)	Wind Speed (m/s)	Lead in 3 Hours (μg/Nm³)	Assumed Lead in 24 Hours (μg/Nm³)
Agricultural area					
A-1	Planting time, spraying activity	52.3	0.34	1.28	10.26
A-2	Post-harvest, no spraying	52.5	2.15	0.21	1.69
B-3	Planting time, no spraying	51.0	5.70	1.07	8.57
B-7	Planting time, no spraying	58.9	5.70	0	0
Settlement area*					
B-2	None	65.3	1.36	0.92	7.36
B-4	None	64.5	1.39	1.56	12.48
B-10	None	66.5	1.49	0.49	3.99

** Lead was not found in B-1, B-5, B-6, B-8, and B-9*

Lead was detected in breast milk samples, with a median of 0.019 ppm (0.008 to 0.791 ppm). Approximately 26.7% (n=8) of these samples were categorized as unsafe because as they exceeded the safety limit of 0.035 ppm. However, there is no association between the status of locations positive for lead in the air and the status of lead in breast milk (p=0.191). Nevertheless, there was a tendency for mothers living in areas with higher air lead levels to have breast milk lead concentrations that exceeded the safe limit (87.5% versus 54.5%) (Figure 3).

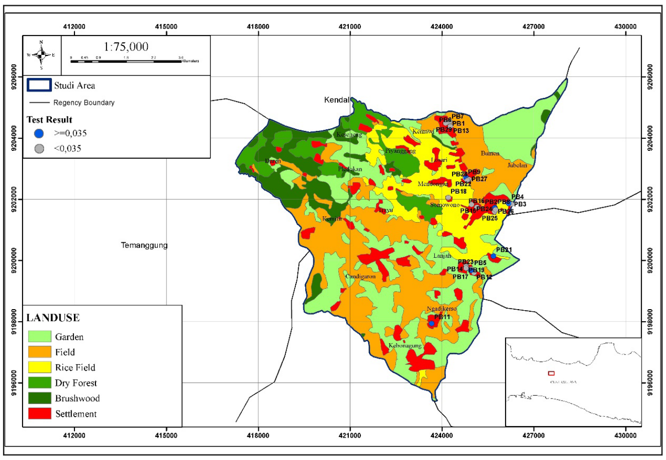


Figure 3. Distribution of Lead Status in Breast Milk Among Breastfeeding Mothers

The smoking habit of family members in almost all breastfeeding mothers, and more than half of them smoke at home, is the dominant factor that can affect lead exposure in breastfeeding mothers around the study site (Figure 4). This behavior was common, partly due to the cold climate in the region, which has contributed to the prevalence of smoking among men, including the husbands of the breastfeeding mothers.

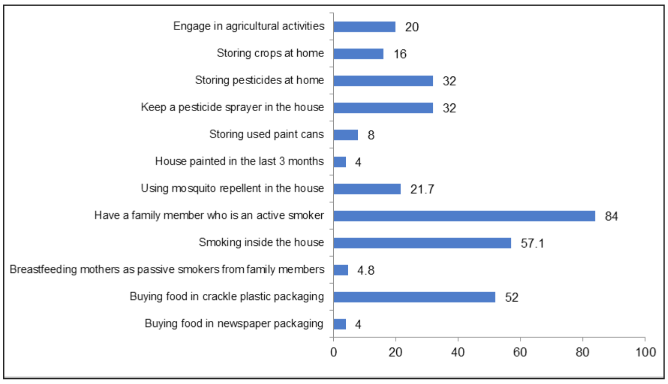


Figure 4. Percentage of Breastfeeding Mother's Behaviours Related to a History of Lead Exposure

DISCUSSIONS

Breastfeeding mother are a vulnerable population that often receives insufficient attention for their health despite playing a crucial role in the First 1000 Days of Life stunting prevention program. Breast milk serves as an infant's main nutritional supply, and in the first six months following delivery, it is especially susceptible to chemical contaminants from both internal and external (environmental) sources. In Indonesia, improper disposal of battery waste is a well-known source of health concern due to lead exposure (30). However, research on lead contamination in agricultural areas remains limited in terms of exploring its impact on public health, particularly in relation to maternal and child health.

Overseas study has revealed the lead content of breastfeeding mothers' breast milk and its effect on children's development (29). Even though the United States Centers for Disease Control (CDC) has issued management recommendations for breastfeeding mothers with lead in their blood and breast milk, studies on this topic in Indonesia, particularly on the nutritional status of breastfeeding mothers and lead levels in breast milk, are limited. Previous research has discovered lead in breast milk in the agricultural areas of Brebes and Semarang Regency (21). The data demonstrate that breastfeeding mothers in both areas had lead levels in their breast milk that exceeded the US CDC's advised limit. The level of lead pollution in agricultural areas is influenced by pesticide application patterns. Consistent with previous studies, this study also found that approximately one-quarter of breast milk samples exceeded the safe limit of lead contamination. It is essential to identify lead considering that livelihoods in this environment rely on agriculture.

As previously stated, lead enters breast milk via the mother's blood, which may also be retained in the mother's bones (11). During pregnancy and lactation, lead is released from bone deposits into the blood, where it is afterward secreted into the breast milk at an extent determined by the mother's blood lead levels. Lead levels in the mother's blood correlate with the amount of lead transferred into breast milk. Studies have indicated that elevated levels of lead in the blood of mothers correspond to elevated levels of lead in breast milk (11,31–33). The circulatory system is also affected by the body's micronutrient levels, such as calcium and iron levels in the blood. Considering the nutritional outcomes of breastfeeding mothers at the research site, particularly in relation to adequate intake of calcium and iron, can provide insight into how easily lead compounds are absorbed by the body. Moreover, the detection of breastfeeding mothers who were anemic and based on the MCV, MCH and MCHC profiles was suspected to be of the microcytic anemia type as a marker of disturbances in the formation of red blood cells due to lead exposure (6).

One possible route for lead to enter the body is through ingestion. Lead can enter the body through contaminated food and beverages, for example, through lead dust that contaminates food surfaces and then is eaten. Children who play on contaminated dirt or consume lead from paint chips are examples of cases that are frequently investigated. The structure and functionality of the digestive system can be damaged or disrupted by lead entry, which can also have an impact on the function of the body's distal organs. Lead exposure affects intestinal mechanisms by reducing the expression of intestinal tight junction proteins, disrupting the intestinal microbiota's metabolic profile, causing dysbiosis, and activating the oxidative stress response in the intestinal immune barrier, which in turn alterations the inflammatory response and eventually the immunomodulatory genes in the intestine (7). Additionally, studies reveal that plants are capable of absorbing lead from groundwater and soil. This may potentially be a point of entry for lead to enter the digestive system of humans.

Limited amounts of food naturally contain lead, according to research. In their natural habitat, plants have the same ability to absorb heavy metals from the soil as do people. The soil's pH, the amount of heavy metals present, how well the soil interacts with other substances, the health of the soil's microflora, and the kind of plant all affect how well a soil can absorb heavy metals (33). These factors can act individually or in combination with each other and can change the soil behaviour of existing lead, as well as the rate of uptake by plants. Plants absorb it using roots and will then accumulate in several parts of their body. In plants that are in the root tuber group, they have a tendency to accumulate food reserves in the tubers (34). People frequently consume this kind of plant as food. As a result, humans may eat this meal's lead content and it could contaminate food supplies chemically. The US and China have long been the centres of research and surveillance about lead levels in food items. According to studies conducted in China, the most common foodstuffs containing lead were wheat derivative goods, green leafy vegetables, and rice. Even low levels of lead content can have an impact on children's health, even though the study's margin of exposure (MOE) technique did not reveal any emergency (35). Meanwhile, researchers in Semarang City discovered that lead levels in green leafy vegetables from traditional markets above the norm (2,462 ppm in spinach, 2,629 ppm in kale, and 2,875 ppm in mustard greens) (4). However, the results are questioned because the procedure section makes no mention of the step of washing the sample container clean and free of heavy metals by rinsing it first with nitric acid and distilled water (1:1) In this study, however, food samples and groundwater sources that mothers regularly ate were not risk factors in this study.

A further external element comes from the surroundings, specifically the air. If the particle is small enough, the lead can be inhaled all the way into the lung. The findings of lead in the air indicate that the environment in which breastfeeding mothers live should be wary of. A review identifies studies that discuss the interaction of lead exposure to stress in relation to early childhood cognitive and behaviour found that mothers who were exposed to lead during pregnancy has a detrimental effect on child cognition at 24 months of age. With the combination of stress experienced by the mother during pregnancy, this impact becomes even worse, i.e. the infant has problems in emotional development (36).

Lead contamination in agricultural air can be caused by several sources, including the use of lead-containing pesticides and fertilizers, as well as deposition from other sources (37–38). Pesticides containing lead such as lead arsenate can cause air contamination. Exposure to these pesticides can cause acute and long-term poisoning, including neurological effects in children. Monitoring lead content in soil and air is very important to reduce lead exposure. Further research on sources of lead and how to reduce them could help in reducing the risk of environmental contamination (39). The findings of lead in this study were not only found in agricultural land but also in settlement areas. This can happen because residents live adjacent to agricultural land. Even though it is not statistically significant, there is a tendency for this condition to have an impact on the presence of lead in breast milk.

Furthermore, lead can come from cigarettes, cigars, and other tobacco products when smoked (including second-hand smoke), according to research on active and passive smokers, and it can stick to clothes, furniture, curtains, walls, hair, and skin. In a US study of 6–19-year-old passive smokers, blood lead levels were considerably higher in those with moderate and high serum cotinine levels (18–29% higher) than in those with low serum cotinine levels (40). Although this study did not measure cotinine levels in mothers, the findings of lead in the blood and breast milk of breastfeeding mothers might be linked to their husbands' smoking habits.

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AUTHORS' CONTRIBUTION

DRP planned and executed sample preparation and data collection in the field, with the support from NL. Planning was directed by SS and AK, who also helped to evaluate the findings. The development of the GIS mapping and field preparation were assisted by TTP and NS. The manuscript's writing was managed by DRP. All authors contributed insightful valuable feedback and contributed to shaping the research study, analysis, and manuscript.

CONCLUSION

The micronutrient intake of breastfeeding mothers is below Indonesia's RDA. Mothers who experience anemia are suspected to be in the microcytic category, which is one of the factors suspected of being exposed to lead or iron deficiency. Lead is found in food and groundwater but within safe limits. In several agricultural and settlement areas, lead was found in the air. The majority of mothers have families who are active smokers. The lead concentration in a quarter of breast milk samples was found to be over acceptable limits. Poor internal maternal factors (macro and micronutrient deficiencies) can enhance the absorption of lead from the mother's external (environmental). It cannot be denied that efforts to limit lead exposure in agricultural areas are critical; nonetheless, enhancing the nutritional status of breastfeeding mother and lead-removal supplements are viable alternatives worth considering.

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