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2 Accumulation of Lead (Pb) in the Lichen Thallus of Mahogany Trees in Medan City Road

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Abstract Rapid growth of vehicles in Medan, Indonesia, is one of the causes in the increasing of air pollution, in which approximately 85% is contributed merely by vehicles. On the other hand, the use of lead-based fuel in motor vehicle increases the air contamination in Medan. This study aimed to obtain an accumulation of lead (Pb) in the thallus of lichens in mahogany trees in four different locations in Medan, Sumatera Utara, Indonesia, in which the lichens act as a bioindicator of air contamination as well as measuring the lichen's lead correlation and traffic densities. Purposive sampling location was determined based on the traffic density level with different air pollutions; the location which was far from traffic circulation was used as the control. The analysis of Pb was conducted using atomic absorption spectrophotometry (AAS). The data were analyzed descriptively to discover and compare Pb accumulation between each location with different traffic density

levels. The result showed that there were 11 species of 7 genera and 7 families with two types of the thallus (foliose and crustose) in mahogany trees. The traffic density level influenced the diversity of lichens as the traffic density was quite significant with the number of lichen types. The levels of Pb and traffic density correlated very significantly at the level of $\alpha = 0.01$ for *Parmelia saxatilis*, *Lepraria incana*, and *Pertusaria amara* type, while *Opegrapha atra* had a significant correlation. The accumulation of Pb in the thallus of *Pertusaria amara* ranged from 5.23 to 15.07 $\mu\text{g/g}$, whereas medium in *Lepraria incana* ranged from 1.19 to 4.88 $\mu\text{g/g}$. Thus, *Pertusaria amara* which had greater Pb level than *Lepraria incana* had the potential as a resistant bioindicator. The correlation analysis of Pb levels and traffic density showed that *Pertusaria amara* had a significantly high correlation compared with *Parmelia plumbea*, *Parmelia glabratula*, and *Graphis scripta*. Furthermore, *Lecanora conizaeoides* was a tolerant bioindicator of air pollution whereas *Parmelia saxatilis* had the potential to be a tolerant bioindicator.

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1 Introduction

Environmental problems become increasingly uncontrolled, especially in Medan, Indonesia. This is due to the rapid increase in the number of public and private vehicles, followed by the growth of

development. The growth of the industrial and transportation sectors has an impact on environmental degradation. Motorcycles are one of the main sources of air pollution because they contain various harmful contaminants for humans. Imperfect combustion from motor engines and industrial manufacturing has been producing contaminated materials including black smoke, carbon monoxide, nitro oxide, sulfide oxide, hydrocarbon, phosphorus constituents, and lead (Sastrawijaya 2009).

One of the most dangerous sources of pollutants to living things is lead (Pb). Lead is a hazardous material which is harmful both for human and environmental features. These metals enter the human body through respiratory and digestive systems or directly through the skin surface. The largest contributor to Pb pollution in the air is the transport sector. Fergusson (1990), Dahlan (1992), and Asmiwyati (2012) state that motor vehicles are the main source of Pb that pollutes the air in urban areas. Lead is also considered the main metal pollutants within the air (Jamhari 2009). It is estimated that about 60 to 70% of Pb particles in urban air come from motor vehicles, and about 75% of Pb added to fuel oil will be emitted back into the atmosphere (O'Neill 1993). Furthermore, Fergusson (1990) states that Pb released from motor vehicles averages 0.02–0.05 μm . When the particle size becomes smaller, the duration of attachment will also be longer. The terms "Premium" and "Pertalite," which is one of the Indonesian fuel products, have been investigated to contain amounts of lead, which probably is emerged during the processing stages (Mairizon 2019). Although the bans in using the Tetraethyllead (TEL) for the processing of "Premium" has been issued, the lead contents only decrease from 0.3 to 0.0013 g/l. Subsequently, this lead content is not considered a safety amount, even though the amount is reduced to be 0.001 g/l, unless it is 0.000 g/l. By considering the presence of lead in fuels which accounted around 70–80%, high amounts of lead could have been airborne in a form of particulate matter (Francis 1994).

Pb contained in the air can accumulate in the body tissues of living things, especially in thallus of lichens. Lichens have the potential to absorb contaminants in the atmosphere (Kar et al. 2014; Naila et al. 2017). Based on several reports, it is known that the lichen thallus can accumulate Pb from exhaust emissions of motor vehicles. Studies by Bargagli et al. (1987), Majumder et al. (2013),

and Kuldeep and Prodyut (2015) found that lichens are a good indicator of air pollution. Furthermore, Sequiera and Kumar (2008) state that lichens act as indicators of air quality, climate change, and biodiversity components. Some previous studies have measured the ability of lichens to accumulate Pb. In the Tuscany region, Italy, the concentration of Pb in the lichen thallus was 13.2 $\mu\text{g g}^{-1}$ dry weight (Bargagli et al. 1987). Most Pb concentrations were found around vehicle parking areas and near the highway. The accumulation of Pb in *Parmelia physodes* decreased proportionally over the distance from the highways (Kovács 1992). Derucclé (1981) also showed that Pb accumulation was found at 15 m from the highway with 1002 $\mu\text{g g}^{-1}$ dry weight, while the Pb accumulation was only 65 $\mu\text{g g}^{-1}$ dry weight at 600 m from the highway.

In determining the level of urban air pollution, it is necessary to conduct research by using bioindicators so that the quality of air can be obtained. Bioindicators are organisms whose existence can be used to detect, identify, and qualify environmental pollutions (Conti and Cecchetti 2001). Current research on the use of bioindicators in monitoring the presence of air pollution is still limited; thus, in-depth studies should be done about the ability of lichens to be used as a bioindicator of air pollution. This research, therefore, aimed to investigate the accumulation of Pb in the thallus of lichens in mahogany trees at four different sites based on the traffic density and air pollution levels in which the site located far from the traffic circulation was used as the control. Atomic absorption spectrophotometry (AAS) was used to analyze Pb. Data collected were analyzed descriptively to compare Pb accumulation among the sites, and the correlation analysis was performed using Pearson correlation.

2 Experimental Section

The research was conducted in Medan, Indonesia, in four different locations in which the data collection was performed for 6 months. The locations were selected by purposive sampling based on different traffic density and air pollution levels. There were four different locations as mentioned in Table 1. Pancur Batu which was located far from the traffic circulation was used as the control.

The traffic density was measured on Mondays, Wednesdays, and Fridays for a full month from 7:00 to 8:00 am, 11:00 am to 12:00 pm, and 15:00 to 16:00 pm. The traffic density was measured by

calculating the total number of motorcycles passing through the counting station using a hand tally counter. It was then followed by sampling the lichens on the bark surface of 20 mahogany trees in every location in which the lichen locations are 2 m above the ground. As many as 20 trees in four locations were determined, so total sampling of trees were 80 trees. The mahogany trees stances were determined via purposive samplings, in which the lichens were sampled from the mahogany due to the use of this tree as the street shelters. The samples of lichens were identified, and Pb analysis was conducted using atomic absorption spectrophotometry (AAS). Lichen calculations were performed within individual thallus in every site. Matching method via literature studies by utilizing the books *Key to the Lichen Genera of Bogor, Cibodas and Singapore* (Sipman 2003), *Lichen Identification* (Saipunkaew 2009), and *Grasses, Ferns, Mosses & Lichen* (Phillips 1990) was performed, while unidentified lichens were treated with spot test reagent KOH 10% to obtain cation K and C. The analysis of lichens as bioindicators was performed by obtaining the highest concentration of lead per 1 cm², and its areas of thallus for every lichen samples. Subsequently, statistical analysis by using correlation analysis of Pearson correlation was performed to determine and compare the Pb accumulation among the locations. In this study, focus on lead correlation and types of lichens as well as traffic densities on the location is performed, while the lichen morphology is observed via an optical microscope.

3 Results and Discussion

3.1 Lichens with Ecological Characteristics

The traffic density level and measured environmental factors in the research locations are shown in Table 2.

Table 1 Locations of the research

No	Location	Level of traffic density
1	Pancur Batu, Medan	Far from traffic circulation (control)
2	Yos Sudarso Street, Medan	High traffic circulation
3	Sudirman Street, Medan	Medium traffic circulation
4	Cik Ditiro Street, Medan	Low traffic circulation

The traffic density level and measured environmental factors at the four research sites varied from one another. The high level of traffic density was due to its location. The road position is a factor which causes different traffic density levels in each location. The location with high traffic circulation is a protocol road located from various roadways, and it is crowded by motor vehicles every day. Therefore, this location has the highest traffic density compared with other locations. Location with the medium traffic circulation was located at the city center directly adjacent to the location with low traffic circulation with a certain path in which this location is on the area of schools and offices which have low traffic access.

The traffic density was considered to determine the environmental quality that can be affected by exhaust gases of vehicles passing through the research sites. According to Nursal et al. (2005), high traffic density is one source of Pb pollution in the air. Furthermore, Wardoyo and Hidayat (1998) stated that the higher the number of motor vehicles, the higher the Pb pollutant emitted to the surrounding environment. When associated with the measured Pb accumulation in the lichen thallus obtained from each location, the accumulation of Pb in the lichen thallus had a correlation with the traffic density level and other measurable environmental factors. The accumulation power of Pb in each type of lichens was not the same; even certain types have a high accumulation power. *Pertusaria amara* has a high correlation rate compared with the other types. Environmental factors, such as air humidity, air temperature, light intensity, and wind speed, have a greater influence on Pb accumulation in lichen thallus than the other measured factors. This relates to the nature of lichens' life and the growth of thallus which is more suitable for humid air conditions (Zedda and Rambold 2009). Then, Sundberg et al. (1997) claim that lichens can grow and photosynthesize in highly humid habitat conditions (85%). Humidity and temperature in the three research sites were not much different compared with the light intensity and wind speed. Nursal et al. (2005) argue that lichens can live better and more fertile in more humid environmental conditions, so the absorption of water, minerals, and the accumulation of contaminants becomes more effective and numerous. The light intensity at the research sites still supported the life of lichens. According to Ray Showman (1972), the lowest light intensity value needed by lichens to do photosynthesis effectively is 1025 lux meter.

Table 2 Measurement data of environmental parameters in the research locations

Parameter	Research location				Total	Average
	Far from traffic circulation (control)	High traffic circulation	Medium traffic circulation	Low traffic circulation		
Traffic density (vehicle/h)	0	15.909	8.893	7.765	32.567	10.855
Humidity (%)	76	77	79	81	237	79
Air pollution (°C)	25	27.6	27.8	27.9	83.3	27.8
Light intensity (lux)	1100	1500	1371	1241	5212	1303
Wind velocity (m/s)	2.0	2.9	1.8	1.6	6.3	2.1

3.2 Lichen and Pb Accumulation

Based on the exploration results of lichens at four research sites, there were 11 species of 7 genera and 7 families. According to Galloway and Moberg (2005) and de Silva and Senayake (2015), the number of lichens can be decreased due to the disruption of human activities and extreme environmental conditions. Lichens found in the study were grouped into two types of the thallus, namely foliose and crustose type. The average of Pb accumulation in the lichen thallus is shown in Table 3.

Based on the results, varied lead contents in the lichen samples were found. The highest lead amount which is high traffic circulation had lichen species of *Parmelia saxatilis*, and in medium traffic circulation, the

Parmelia plumbea were commonly found, whereas the *Parmelia glabulata* was found. This genus is classified into the family of Parmeliaceae with thallus type of foliose, freely attached and loosed on the substrate. The distinguishing characteristics in Parmeliaceae are the presence of *pseudocyphellae* which covers the porous epicortexs, white-colored medulla, and commonly contained salazinic acid. Characterized thallus Parmeliaceae with reagents test KOH 10% demonstrated kation K with yellow-to-red colors. Rhizines protect the entire surface of the body. The lichen group that has the highest foliose forms specific and recognizable thallus. In this species, the differences in lead contents are caused by the different environmental conditions of sample sites. Environmental features contribute higher in lead accumulation within the thallus of lichens of

Table 3 The average Pb accumulation in the lichen thallus in the research locations

Species	Type of thallus	Total level of Pb (µg/g)			
		Far from traffic circulation (control)	High traffic circulation	Medium traffic circulation	Low traffic circulation
<i>Parmelia plumbea</i>	Foliose	0	0	44.99	19.99
<i>Parmelia caperata</i>	Foliose	0	0	0	0
<i>Parmelia glabratula</i>	Foliose	0	0	29.82	38.04
<i>Parmelia saxatilis</i>	Foliose	0	21.37	33.82	0
<i>Graphis elegans</i>	Crustose	0	0	0	0
<i>Tryphethelium virens</i>	Crustose	0	0	0	0
<i>Lecanora conizaeoides</i>	Crustose	0	5.23	0	0
<i>Lepraria incana</i>	Crustose	0	4.88	4.88	1.19
<i>Graphis scripta</i>	Crustose	0	0	20.47	0.95
<i>Opegrapha atra</i>	Crustose	0	0	0.47	0.20
<i>Pertusaria amara</i>	Crustose	0	14.43	15.07	5.23

Table 4 Correlation of Pb in lichens and the number of lichen thallus

Correlations (<i>r</i>)			
Component		Lichens	Lead (Pb)
Lichens	⁴⁸ Pearson correlation	1	-.345
	Sig. (2-tailed)		.363
	N	9	9
Lead (Pb)	⁴⁵ Pearson correlation	-.345	1
	Sig. (2-tailed)	.363	
	N	9	9

Parmelia saxatilis than the other factors (Cameron and Richardson 2006).

Some types of lichens are sensitive to air pollutants, so they are rarely found in polluted areas ((Mokni et al. 2015). More tolerant species can accumulate a certain amount of pollutant to a tolerable concentration range. Tolerant species can be used as an indicator of accumulation to detect the pollutant levels, especially in the air (Nursal et al. 2005). Metals absorbed by lichens accumulate in the tissue of the thallus. The structure of lichen thallus is one of the factors that affect the efficiency of metal absorption. According to Kinalioglu et al. (2010), the efficiency level of pollutant accumulation in the thallus is foliose > crustose > fruticose in sequence. Moreover, Scerbo et al. (2002) mentioned that the surface area of the thallus causes foliose lichens to have greater contact with the pollutants, so the accumulation of pollutants is more efficient than the other thallus types. Foliose lichens or known as leafy lichens have a wide structure of thallus and can be easily removed from its substrate. The foliose type in this study was found in several species of *Parmelia* sp. Furthermore, the

correlation between the number of lichen thallus and Pb is presented in Table 4.

The correlation between the number of lichen thallus and Pb was found to be negative ($r = -0.345$); thus, it was not significant and directly proportional. Negative correlations showed that the lower the number of lichen thallus, the higher accumulation of Pb in the lichens. Therefore, Pb can inhibit the growth of lichens.

3.3 Lichens as a Bioindicator of Air Pollution

In this study, *Lecanora conizaeoides* was found to be a bioindicator which was tolerant of air pollution. *Parmelia saxatilis* can be a tolerant bioindicator which was found only in the locations with high ⁵³ medium traffic density. Moreover, the result also showed that there was a very significant correlation between *Parmelia saxatilis* and traffic density ($r = 0.842$) as shown in Table 5. *Parmelia saxatilis* can accumulate a certain amount of pollutant ⁴⁶ a tolerable concentration.

According to the graph in Fig. 1, it can be seen that the comparison of lead content in high traffic circulation (with *Parmelia saxatilis*) is smaller compared with that in medium traffic circulation, in which this comparison is significantly correlated. The type of motor vehicles and imperfect combustion of "Premium" caused the unequal amounts of lead. This non-perfect combustion from motor vehicles contaminated the air via lead (Sastrawijaya 2009), while the other environmental factors including the air moisture and temperature, light intensity, and wind speeds have greater impact to the lead accumulation.

Tolerant species can be used ⁵⁰ as an indicator of accumulation to detect the level of pollutants, especially in the air (Nursal et al. 2005). *Lecanora conizaeoides* widely found in industrial areas can be used as a

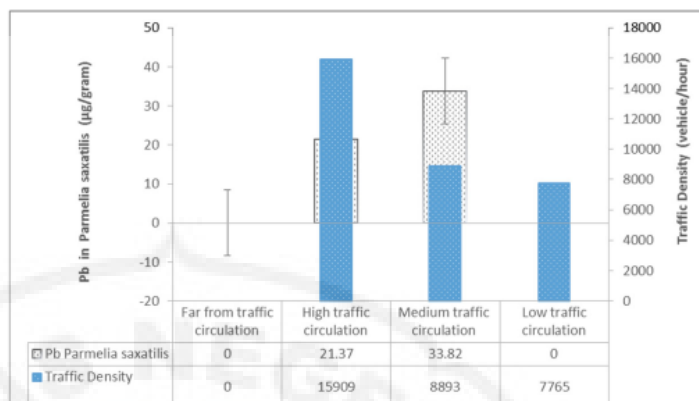
Table 5 The level of Pb in the thallus of *Lecanora conizaeoides* and *Parmelia saxatilis* lichens and its correlation with traffic density

Location	<i>Lecanora conizaeoides</i>		<i>Parmelia saxatilis</i>	
	Level of Pb (µg/g)	Correlation (<i>r</i>)	Level of Pb (µg/g)	Correlation (<i>r</i>)
Far from traffic circulation (control)	0	0	0	0
High traffic circulation	5.23	0.710*	21.37	0.842**
Medium traffic circulation	0	0	33.82	0.624
Low traffic circulation	0	0	0	0

*Significant

**Very significant

Fig. 1 Correlation of Pb in *Parmelia saxatilis* with the number of motor vehicles in each location



bioindicator of air pollution. *Lecanora conizaeoides* can be used as an air pollution bioindicator, resistant to air pollution, and can grow at high traffic density. This lichen had a yellowish green crustose type on the upper surface. The morphology of *Lecanora conizaeoides* (crustose) and *Parmelia saxatilis* (foliose) with different colors of the thallus is shown in Fig. 2.

Furthermore, lichens classified as the cosmopolitan type were found in all research locations, namely *Lepraria incana* and *Pertusaria amara*. This type was classified as the most resistant type which had the highest percentage of presence in both clean and polluted air. The accumulation of Pb in the lichen thallus and their correlation with traffic density are presented in Table 6. According to Panjaitan et al. (2010), *Leprariaceae* family was characterized by thallus resembling flour and spreading unevenly with margins that form small and pale green lobes to whitish yellow. Physiologically speaking, the family of *Leprariaceae* has no multilayer structures which are called leprose (Saag et al. 2009), and it could have an ability to be tolerating to any air contaminants.

The accumulation of lead particles (Pb) in *Lepraria incana* and *Pertusaria amara* was indicated by blackish brown stain, while the algae layer was seen in the upper cortex. *Lepraria incana* found in all research locations

includes the type that easily adapted to poor air quality conditions. *Lepraria incana* also has the ability to adapt in an unstable environment particularly that can alter from highly contaminated condition to low contamination (Syeda et al. 2017). The use of *Lepraria* sp. as the bioindicator of air pollution was conducted in Bandung (Taufikurahman 2010). This type of lichens can accumulate pollutants from the air, so it is used as the bioindicator. The presence of lichens was based on their sensitivity to air pollution. Lichen's sensitivity to air pollution can be seen through the changes in its diversity and the accumulation of pollutants in its thallus. Lichens are very sensitive to air pollution. Most lichen species are very sensitive to sulfur dioxide (SO₂) gas and other exhaust gases from motorized and industrial vehicles. According to Ohmura et al. (2009), the type of *Lepraria* sp. lichen is highly sensitive to sulfur dioxide (SO₂), and it can only live in areas with clean air. The morphology of the crustose type of *Lepraria incana* with green thallus and *Pertusaria Amara* with green to gray thallus can be seen in Fig. 3.

Furthermore, the research results on the classification of lichens showed that the crustose type was more commonly found with five types than the foliose type. Crustose lichens last longer than the other types. This is because the smaller the crustose, the flatter it is attached to the corticolous (bark). Crustose has been used in

Fig. 2 a *Lecanora conizaeoides* (crustose). b *Parmelia saxatilis* (foliose)

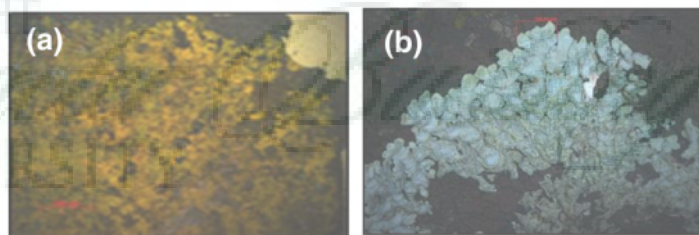


Table 6 The level of Pb in the thallus of *Lepraria incana* and *Pertusaria amara* lichens and its correlation with traffic density

	Level of Pb ($\mu\text{g/g}$)	Correlation (r)	Level of Pb ($\mu\text{g/g}$)	Correlation (r)
Far from traffic circulation (control)	0	0	0	0
High traffic circulation	4.88	0.628	14.43	0.292
Medium traffic circulation	4.88	0.863**	15.07	0.881**
Low traffic circulation	1.19	0.845**	5.23	0.710*

*Significant

**Very significant

Japan as an air pollution bioindicator. McCune et al. (2007) suggest that the crustose lichen is more tolerant of air pollution because it has a relatively simpler structure of thallus than that of other lichen thallus types. The tolerant type of lichens can survive in areas with contaminated environmental conditions whereas the sensitive type of lichens usually cannot be found in areas with poor air quality. The difference in the sensitivity of lichens to air pollution is closely related to its ability to accumulate pollutants (Conti and Cecchetti 2001).

3.4 Correlation of Lead (Pb) in Lichen Thallus and Traffic Density

Lichens can absorb lead from the air. The level of lead accumulation can increase with increasing traffic density, but the level decreases if it is farther from the edge of the highway. According to Dahlan (1992), lead content in plants on the roadside can reach 50 ppm, but the number will decrease 2–3 ppm at 150 m from the highway. Furthermore, the correlation between lead (Pb) in the lichen thallus and traffic density is shown in Table 7.

In the location II which had high traffic circulation, the correlation of Pb in lichens with traffic density was low and not significant in *Pertusaria amara* ($r = 0.292$), whereas the correlation was high and very significant in *Parmelia saxatilis* ($r = 0.842^{**}$). The correlation of Pb in *Parmelia glabratula* and *Graphis scripta* and traffic density was not significant in location III than in IV. According to Sunarya et al. (1991) and Antari and Sundra

(2002), there was much more amount of Pb in the roadside plants with more densely motorized than the amount of Pb which was far from the roadside. In the location with medium traffic circulation, *Lepraria incana* and *Pertusaria amara* had a highly significant correlation, while *Opegrapha atra* had a significant correlation.

A negative correlation (–) was found in the location with low traffic circulation by *Parmelia plumbea* which indicates the opposite correlation or inversely proportional. Thus, if the volume of vehicles is high, the lichen thallus area will become smaller, and vice versa. *Pertusaria amara* had a very high and significant correlation than the other types. In contrast, in the location with high and medium traffic circulations, the correlation of Pb in lichens with traffic density was positive, significant, and directly proportional. The results of Jamhari (2009) showed that lichens were morphologically and physiologically considered relevant to metal accumulation. Lichens show tolerance to metal. Determination of metal concentrations generally uses a bio-indicative approach by utilizing lichens (Loppi et al. 2002). Some common metals measured include lead (Pb), cadmium (Cd), chromium (Cr), zinc (Zn), and copper (Cu).

4 Conclusions

The highest lead contents that are located in high traffic circulation is found in *Parmelia saxatilis*, while on medium

Fig. 3 a *Lepraria incana* (crustose). b *Pertusaria amara* (crustose)

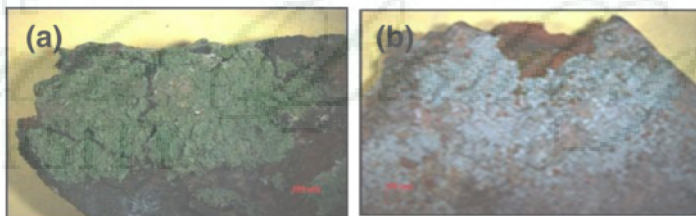


Table 7 Correlation analysis of lead (Pb) in the lichens and traffic density

Species	Type of thallus	Correlation (<i>r</i>)			
		Far from traffic circulation (control)	High traffic circulation	Medium traffic circulation	Low traffic circulation
<i>Parmelia plumbea</i>	Foliose	0	0	0.364	-0.096
<i>Parmelia caperata</i>	Foliose	0	0	0	0
<i>Parmelia glabratula</i>	Foliose	0	0	0.074	0.401
<i>Parmelia saxatilis</i>	Foliose	0	0.842**	0.624	0
<i>Graphis elegans</i>	Crustose	0	0	0	0
<i>Tryphethelium virens</i>	Crustose	0	0	0	0
<i>Lecanora conizaeoides</i>	Crustose	0	0.710*	0	0
<i>Lepraria incana</i>	Crustose	0	0.628	0.863**	0.845**
<i>Graphis scripta</i>	Crustose	0	0	0.511	0.531
<i>Opegrapha atra</i>	Crustose	0	0	0.669*	0.404
<i>Pertusaria amara</i>	Crustose	0	0.292	0.881**	0.710*

*Significant

**Very significant

traffic circulation, the highest content accumulation is found in *Parmelia plumbea*; in low traffic circulation, the highest lead composition is found in *Parmelia glabratula*. The species of *Lecanora conizaeoides* could have been used as a tolerant bioindicator, and the species of *Parmelia saxatilis*, *Lepraria incana*, and *Pertusaria amara* potentially has the ability to be utilized as tolerant bioindicators, in which the lead content within the plants was found to be correlated to traffic densities in significant numbers. *Pertusaria amara* and *Lepraria incana* are classified to be cosmopolite as well as monitoring species, while the *Parmelia plumbea* contributes to the lowest correlation among the other species.

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