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Bioaccumulation and Mapping Heavy Metals of Lead (Pb) in *Turbinaria sp* in Teluk Kodek, Pemenang, North Lombok

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Abstract

Pollution of the aquatic environment can come from domestic, agricultural and industrial activities. The waters of the Teluk Kodek were suspected of being polluted by the activities of waste fuel dumped by ship moorings. This study aims to analyze lead (Pb) contamination in the waters of the Teluk Kodek and to map the distribution of Pb in *Turbinaria Sp*. This research method is the collection of water samples, substrate and *Turbinaria Sp* in the waters of the Teluk Kodek with three replications. The heavy metal content of Pb was tested using the Atomic Absorption Spectrophotometer (AAS), and then the data was processed by analysis of variance (ANOVA). At the same time, they are mapping the distribution of Pb in *Turbinaria Sp* using Scanning Electron Microscopy (SEM). The results showed that the waters of the Teluk Kodek had experienced accumulation of the heavy metal Pb in the substrate samples and *Turbinaria Sp*. While the results of water samples are still below the threshold. Mapping the distribution of Pb in the *Turbinaria Sp* sample shows the distribution results in each part of the sample. It is necessary to carry out early pollution prevention activities in the waters of the Teluk Kodek so that the aquatic environment remains sustainable.

Keywords: Enviromental, Heavy Metals, Lead, Pollutan, Teluk Kodek.

1. Introduction

Pollution of the aquatic environment is a serious and growing problem (Irhamni et al., 2017). Including water pollution in the Teluk Kodek area, Pemenang, West Lombok. Usually caused by an increase in domestic, agricultural and industrial activities. Meanwhile, Teluk Kodek is a mooring area for ships departing from Lombok Island to the Three Gilis, namely Gili Trawangan, Gili Meno and Gili Air. One source of pollution is the waste fuel from the ship in the form of heavy metals (Johnston, 1976). However, heavy metals are considered as contaminants of all environmental pollutants, due to their bioaccumulation and tendency towards toxicity. Other properties of heavy metals are that they settle easily, are absorbed in sediment particles, suspended in water. The metal is then absorbed through the gills and skin and/or ingested through food so that it can accumulate and become toxic to fish (Adam et al., 2021). Bioaccumulation intensity and toxicity are affected by temperature, oxygen concentration, pH and water hardness. One of the heavy metals that is dangerous and easily absorbed/accumulated is lead (Pb).

Lead (Pb) is a natural component and is usually found in small amounts in soil, plants and water. Pb is found in aquatic environments with increasing levels due to anthropogenic activities including the manufacture of batteries, paints and cement (Adam et al., 2019). The results of observations made by the team (Handayani, 2020). in several lakes and streams found an increase in Pb levels in waters, substrates and aquatic biota such as fish. The increase in Pb-based waste in waters causes an increase in Pb pollution. Specifically forms flexible bonds with oxygen and sulfur atoms in proteins (Johnston, 1976), (Adam et al., 2019). The ability to form stable complexes with these elements increases the finability of Pb in proteins. Therefore, exposure to Pb can be fatal for aquatic biota even at low concentrations due to bioaccumulation (Hidayah et al., 2014).

Heavy metals are divided into two types: essential heavy metals and non-essential heavy metals. Essential heavy metals are metals in certain amounts that are needed by organisms, but these metals can cause toxic effects if in excessive amounts, for example: Zn, Cu, Fe, Mn and others, while non-essential metals are metals that are present in the body's benefits are still unknown and even toxic, for example, namely: Hg, Cd, Pb, Cr and others (Hutagaol, 2012). Heavy metal lead (Pb) in the Kodek Bay is mostly produced from ship/boat traffic, industrial waste, and domestic waste (Hynes, 1978). Ships/boats have a role in contributing the heavy metal lead (Pb), this is because ships/boats use fuel oil, the fuel oil is added with tetraethyl which contains lead (Pb) to improve the quality of the fuel ([IACD/CEDA] International Association of Draging, 1999).

One of the biological communities or organisms that directly feel the effects of these contaminants is macroalgae. Macroalgae are organisms that live at the bottom of the waters (Nurcahya & Nugraha, 2014). The nature of macroalgae which tend to be sedentary and unable to avoid contaminants also contributes to the possibility of accumulation of heavy metals in macroalgae organisms (Nurhayati & Putri, 2019). This makes macroalgae a group of organisms that are often used as indicators of environmental pollution and indicators of heavy metal pollution (Adam et al., 2019). One of the macroalgae found in the waters of the Teluk Kodek is *Turbinaria Sp*. No research has been found that specifically addresses the effects of heavy metal Pb contamination on *Turbinaria Sp*. seaweed. Therefore, this study aimed specifically to determine the extent of the bioaccumulation of the heavy metal lead (Pb) and how to map this accumulation in *Turbinaria Sp* in the waters of the Teluk Kodek

2. Methods

2.1. Sample Collections

Determining station locations and sampling in this study were not sequential between station locations and subsequent stations, so random sampling was used. Water samples taken from the surface of the waters are carried out directly using a water sample container that is immersed in the water. The number of water samples taken was 250 ml, followed by providing the station sampling code, and the water samples were put into the cool box. Sediment sampling was carried out vertically using a modified Ekman grab. Depth of 1-1.5 m depth interval for subsequent uptake vertically as well as sediment thickness taken ± 10 cm from the surface of the Ekman grab. After the sediment is taken, the preservation method is put into a plastic clip and then put into the coolbox. Meanwhile, samples of *Turbinaria Sp* seaweed were taken directly at each existing station. Then, put it in a plastic clip and store it in a coolbox. Furthermore, the samples were stored in the freezer until the next analysis was carried out..

2.2. Heavy Metal Analysis

Heavy metal measurements were carried out on water samples and *Turbinaria Sp* The tools used are Atomic Absorption Spectrophotometer (AAS), pH meter, Analytical Balance, Heater, Shaker, Erlenmeyer, Separatory funnel, Burette, and other glassware. While the materials needed are nitric acid 65%, 4 M and 0.15 M, methyl isobutyl ketone (MIBK), ammonium pyrrolidine dithiocarbamate (APDC), sodium hydroxide (NaOH), potassium bichromate (K2Cr2O7) and copper (Cu). The preserved sample was then taken with a 200 mL pipette. The sample was put into a 250 mL beaker containing 2 mL of 1% APDC, set the pH to 4 and heat to boiling. After cooling down to room temperature, place in Erlenmeyer and add 7 mL of MIBK then shake with a shaker for 20 minutes. The solution was put into a separatory funnel and left for 20 minutes. Take the organic layer (top) and place it in the erlenmeyer. For re-extraction, 5 mL of 4 N HNO3 was pipetted and added to the organic layer that was separated earlier, stirred for 20 minutes. Then the mixture was put into a separatory funnel until the boundaries were found (± 20 minutes). The bottom layer (acid layer) was taken and analyzed with AAS (Sudunagunta, 2012). The same thing was done for each standard solution of the metal being analyzed. The purpose of preconcentration of the sample by the solvent extraction method is to separate the metal ions which are determined in the measurements with the Atomic Absorption Spectrophotometer (García and Báez, 2012).

2.3. Bioaccumulation Data Analysis

The formula used to view chemical bioaccumulation is usually expressed in terms of bioaccumulation factor (BAF) which is the ratio of chemical concentrations in organisms (Co) and water (CW) while (BSAF) is for the ratio of bio/organism (Co) and sediment (CS) concentrations (Saleh et al., 2021). $BAF = CB / CWBSAF = CB / CS$ Note: BAF: Bioaccumulation Factor BSAF: Biota Sediment Accumulation Factor Co : Concentration in organism CW : Concentration in water CS : Concentration in sediment. The results of the analysis of the calculation of heavy metal bioaccumulation and SEM observations were analyzed descriptively. Heavy metal bioaccumulation data was processed by analysis of variance (ANOVA). Then proceed to the BNT test to find out the difference between each treatment based on a 5% confidence interval.

2.4. Scanning Electron Mikroskopis (SEM) Observation

The sample preparation stage before being observed with SEM is very important to be able to get quality observation results and to be able to provide the right analysis. All treatments given during sample preparation must not change the original structure of the sample so that the SEM observation results represent the original structure of the sample. The main stages of preparing biological samples before observing using SEM are cutting the sample with the desired orientation, fixation, dehydration, drying, and coating with a conductive layer with a modification of the method (Devos et al., 1998). The fixation stage is carried out to maintain the original structure of the sample so that it does not collapse or break easily. Fixation which is generally carried out has two stages, namely the first stage using glutaraldehyde plus cacodylate buffer, then the second stage is using osmium tetroxide in the buffer. Dehydration aims to remove the water content from the sample. Dehydration is carried out by immersion in alcohol with a concentration level that increases gradually until it reaches 100%. Drying is usually carried out using critical point drying (CPD) (Humphreys and Henk, 1979) or applying certain chemicals such as hexamethyldisilazane which aims to remove the liquid content from the sample without collapsing the sample. Coating with conductive materials can be done using a sputtering machine with conductive materials that are generally used are C, Au, Pt. Observation of several types of samples, namely conductive samples, biological samples without and with preparation, as well as biological samples were observed with a special observation mode, namely variable pressure (VP)-SEM. Observations were made using the Hitachi SU3500 SEM at the Biology Research Center through the Science E-Services Application (ELSA) of the National Research and Innovation Agency (BRIN).

3. Results and Discussion

Bioaccumulation is the entry of chemicals into the environment and living things. According to (Abalaka, 2015). bioaccumulation includes the absorption of chemicals by organisms through food consumption or sediment intake. Bioaccumulation factors are divided into 2, namely BAF and BSAF. The BAF value can be obtained by comparing the ability of organisms (shellfish) to absorb metals from water. While BSAF is to obtain heavy metal accumulation found in organisms with sediment. Therefore there are two BAF values, namely aquatic organisms BAF(o-w) and BSAF sediment organisms BAF(o-s). The results of Pb metal bioconcentration in *Turbinaria Sp* can be seen in Table 1.

Table 1. Results of Bioaccumulation of Pb Heavy Metals in Teluk Kodek

Parameter / Station	Lead (Pb) Water (ppm)	Lead (Pb) Sediment (ppm)	Lead (Pb) Turbinaria (ppm)	Factor Bio-accumulation Water	Factor Bio-accumulation Sediment
S1	0.0025	0.0176	0.0106	4.2400	0.6023
S2	0.0039	0.0198	0.0165	4.2308	0.8333
S3	0.0048	0.0217	0.0182	3.7917	0.8387

Pb metal is a non-essential metal whose use is not yet known in living bodies so that the presence of this element in excess of normal can cause poisoning. The presence of Pb metal in the body often replaces essential metals in enzyme work activities and inhibits enzyme work (Adam et al., 2019). Based on Table 1, the BCF value of Pb metal in tilapia is low accumulative. The BCF value of Pb metal ranged from 3.7 to 4.2. The accumulation of Pb metal in *Turbinaria Sp* in Teluk Kodek comes from the disposal of fuel containing Pb metal from motor boats used for tourism activities and transportation by fishermen. In addition, it is suspected that it also came from agricultural waste containing pesticide residues. This agricultural waste comes from hilly areas, namely in Pemenang District which is a plantation and agriculture which of course uses a lot of pesticides as plant pesticides. Based on research from (Usman et al., 2020) on soil quality for food, vegetable and fruit crops, data obtained that pesticide residues used by farmers to eradicate pests contained heavy metals Pb, Cu and Hg. The research data also showed that the highest BCF value of Pb metal in *Turbinaria Sp* was at station 1 of the mooring zone which is the area where water enters from several tributaries. As an estuary area, of course, it accommodates all waste from various streams with a higher concentration of waste compared to other stations.

SEM Observation

The results of observations from SEM are shown in Figure 1 below.

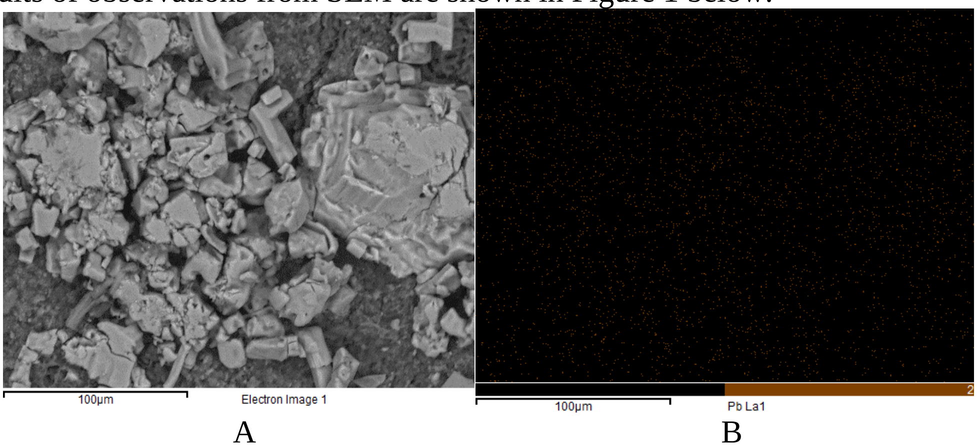

Figure 1. (A) Observation of Turbinaria Structure with SEM observation of 100 µm magnification (B) Mapping of Pb distribution in Turbinaria Structure with SEM observation of 100 µm magnification

Based on Figure 1, it appears that in the results of the Pb mapping in the *Turbinaria Sp*, it can be seen that the distribution of Pb in the samples is present in each area. This shows that Turbinaria is able to absorb the particles around it. Likewise with the distribution on the spectrum, we can see in Figure 2 and Table 3. It shows that there is a spectrum with Pb results at values that exceed the threshold.

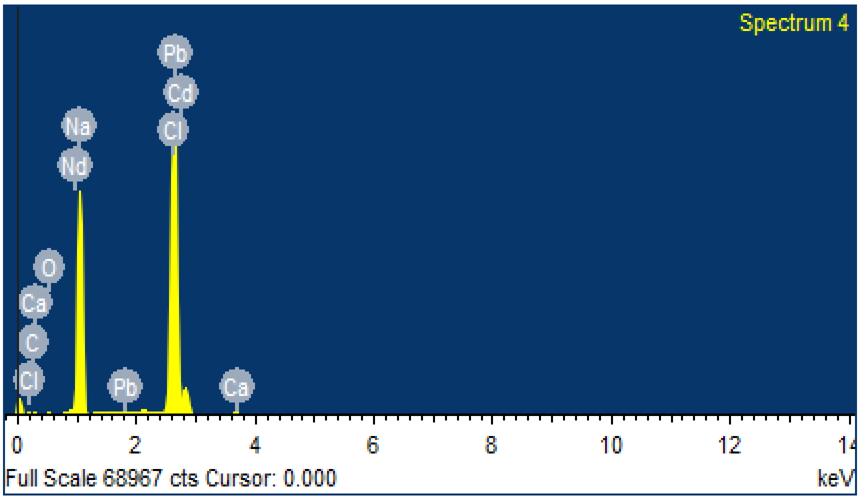

Figure 2. Distribution of Pb in the Turbinaria Structure with SEM observations at 100 µm magnification

Table 3. SEM observation data spectrum on Pb uptake in *Turbinaria Sp*

Element	Weight%	Atomic%
C K	14.20	28.84
O K	1.92	4.09
Na K	5.81	8.62
Mg K	0.02	0.03
Al K	0.10	0.13
Si K	0.11	0.13
Cl K	90.14	86.63
Ar K	0.07	0.06
Cd L	0.11	0.03
Hg M	0.18	0.03
Pb M	1.53	0.25
Totals	100.00	

Heavy metals are a toxic pollutant that can cause death (lethal) or non-death (sublethal) such as disrupting growth, behavior and morphological characteristics of various aquatic organisms (Adam et al., 2019). The impact caused by the presence of heavy metals in waters depends on the presence of metals in water and sediment, their toxicity and their concentration in the environment (Usman et al., 2020). If heavy metals enter the body of living things, they will experience bioconcentration, bioaccumulation and biomagnification. Bioconcentration is the entry of contaminants directly from the water by living things through tissues such as gills or skin (Handayani L, 2020),(Olaifa et al., 2010),(Odey et al., 2020). Meanwhile, bioaccumulation is the entry of pollutants by living things from an environment through a mechanism or pathway. While biomagnification is a process in which the concentration of pollutant increases with the increasing position of living things in a food chain (Abd-allah and Ismail, 2018). Bioaccumulation occurs in body tissues following absorption of metals from water or through contaminated feed. According to (J Weis and P. Weis, 1987) the bioaccumulation of heavy metals in fish depends on the type of metal and fish species. The highest accumulation of heavy metals is generally found in liver and kidney tissue. According to (Keshari et al., 2016). states that the bioaccumulation of chemicals in a waters is an important criterion in evaluating the ecology and level of pollution of an environment. To measure the level of pollution of a waters by chemicals caused by industrial activities, agriculture and household waste is to measure the bioconcentration of the biota that live in it.

4. Conclusion

From this study it can be concluded that there is bioaccumulation of the heavy metal lead with changes in the distribution of Pb accumulation in all parts of *Turbinaria Sp.*

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