Original Research Article

THE CHARACTERISTICS OF HEAVY METAL CORRELATION Pb and Cu WIDENG CRAB TISSUE IN INDUSTRIAL WASTEWATER IN GONJOL RIVER, DEMAK

ABSTRACT

Demak's brackish water pond farmers rely heavily on the Gonjol river. The moment, this river is barely polluted by factory wastes such as paint, furniture, ceramics, and vehicle factories. The goal of this study was to determine the levels of heavy metals Pb and Cu in Wideng crab, water, and sediment in the upper course and estuary of the Gonjol river. This study was conducted from January to March at the river's upper course and estuary and six replications. The Atomic absorption spectroscopy was used in the heavy metal content analysis. The data were analyzed using the linear regression method. The observations revealed that water quality based on Pb and Cu in water, sediment, and wideng crab tissue was still of standard quality, with the exception of Cu in upper course and estuary sediment. In the upper course and estuary, heavy metal Pb and Cu in both sediment and tissue did not show a significant correlation. However, there is a correlation between water and tissue in the upper course and estuary. In the upper course and estuary, there was no correlation between sediment and water. According to the water analysis results that correspond to Decree of the Minister of the Environment of Indonesia No.115, 2003 about standard level water quality with STORET method or infection index method, the water has C categories, which is the middle pollution level for heavy metal Pb and Cu. While the levels in the tissue and sediment were below the tolerance limit.

Key Words: correlation, Pb and Cu, gonjol river, wideng crab tissue, characteristics

INTRODUCTION

The Gonjol River is located in the Pandansari area, a Demak coastal area with high ecological and economic value. Because there are still many mangrove plants present, it serves as a source of nutrients, spawning grounds, nurturing areas, and foraging for various marine biota. While one of them is the economic value as a source of irrigation for pond farmers in the surrounding area. The river is located near the Jatengland Industrial Park Sayung industrial area. (Dinulislam, Lumbanbatu and Affandi, 2021) Increased waste disposal from industrial activities in the area will degrade environmental quality; in relation to the use of coastal and coastal areas as industrial areas, some industries may pose a threat to coastal and coastal sustainability (Danish and Wang, 2018). At this time, the Gonjol river receives significant pollutant overflow from ceramic factories, motor vehicle manufacturers, dyeing and paint factories, and furniture factories, and is estimated to contain a high concentration of heavy metals such as lead (Pb) and copper (Cu) that are discharged into the canal in the flow the river Gonjol (Afandi, Widowati and Ambariyanto, 2019) Disposal of waste into the Gonjol river system will cause an increase in some of the waste entering the aquaculture waterways. The quality of the waters in the area surrounding the estuaries of the Demak, East Flood, and Mangkang rivers have declined due to pollution from the textile, plastic, pharmaceutical, paint, ceramic, fish auction place, car painting industry, and lubricant industry. (Budiyanto, 2018), This results in a significant amount of heavy metal contamination of Pb and Cu (Kuchhal and Sharma, 2019) (Zhang *et al.*, 2019). Heavy metals such as Pb and Cu can build up and accumulate in crustacean tissues.(Razak and Abdullah, 2021)

Wideng crab is a type of crab that plays an important ecological role as a nutrient converter, mineralizer, oxygen distributor, and organic matter decomposer. The presence of heavy metals such as Pb and Cu will have an impact on the lives of Wideng crabs that live in aquaculture areas. (Nur *et al.*, 2021) The Wideng crab is abundant in the study area and is regarded as a pest by the local community. Heavy metals Pb and Cu can have an impact on the life of Wideng crab in water bodies to a certain extent because they are non-selective filter feeders, live permanently (sessil), can live in polluted areas, and can accumulate heavy metals with a concentration factor of 105, allowing heavy metals to accumulate in the body. (Cochard, 2017). Pb metal can penetrate cells and accumulate in the tissues of Wideng crab., and it tends to form complex compounds with organic substances found in the body of Wideng crab whereas copper (Cu) is an essential metal that is required by organisms as a coenzyme in the body's metabolic processes in low levels, and its toxic nature only appoints it as a coenzyme in the body's metabolic processes..

The goal of the study was to look at water quality, heavy metal content in water and sediments, and the characteristics of heavy metal correlations, particularly Pb and Cu, between sediment, water, and the Wideng crab in the Gonjol river.

METHOD OF RESEARCH

A quantitative approach was used in this exploratory descriptive research (Baarda, 2019). Heavy metal content data in water, sediment, and wideng crab were quantitatively analyzed in the lab, while water quality data such as temperature, pH, salinity, dissolved oxygen content, and brightness were collected during the survey**.**(Hussain *et al.*, 2017)

The scope of this study is as follows: The purpose of this study is to investigate the levels of heavy metals Pb and Cu in the Gonjol River, Demak in wideng crab, and sediment as their habitat.**.**

Locations of Sampling: The Gonjol river was sampled at two locations: upstream and estuary. For homogeneous sampling of sediment, water, and Wideng crab at each location, 5 points were taken far apart. Environmental physical parameters are measured only under the same environmental conditions as when samples are collected.

Table 1 The locations of the sampling points.

Station	Location	Distance from pollutant source	Information

1	Upstream Gonjol River	10 m	The location is close to the source of the pollutant. dense industrial area, route Semarang-Demak outside the city
2	Gonjol River Estuary	6 km	Many mangrove plants are found at the confluence of rivers and sea water, the most common of which are *Avicennia* and *Rhizopora*.

The condition of the waters surrounding the source of pollution, including topography and slopes, water base (substrate) characteristics, and the direction and pattern of water currents, must all be considered when selecting a sampling location. Research location as follow : (Fig 1)

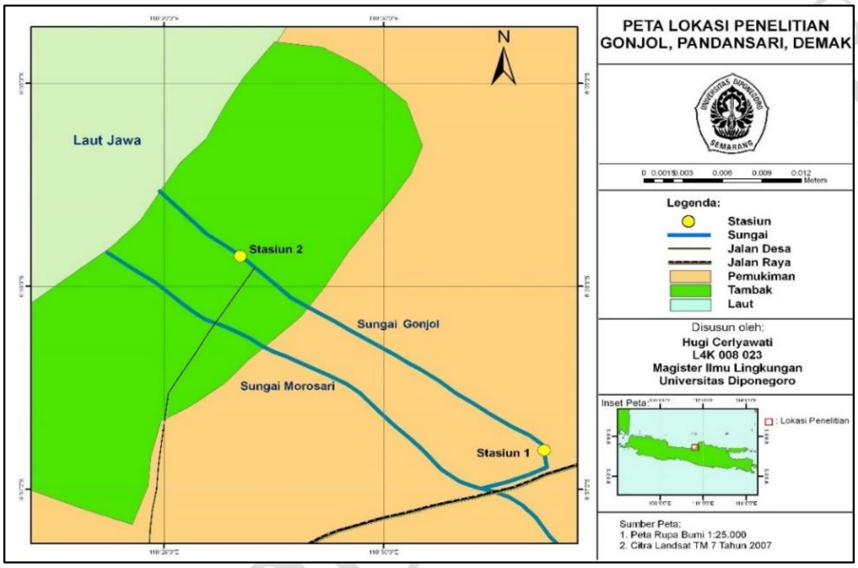

Figure 1. Research location in Gonjol river, Demak

The location was widened by dividing it into four zones. The first and second zones are separated by 3 meters, while the third and fourth zones are separated by 5 meters from the second zone. In each zone, approximately 15 widengs crab were taken, which were then measured for length, width, height, weight, and sex from the wideng crab. Water was sampled at the upstream and estuary locations by dividing each into five zones. In each zone, 1 liter of water is collected and analyzed in the laboratory, and sediment samples are treated similarly by collecting 10 grams of sample. The levels of heavy metals in Wideng tissue and sediment samples were determined using the nitric acid digestion method. The digestion treatment's goal is to reduce disturbances caused by organic matter and convert metals associated with particulates into free metal ions that can be determined using AAS (Alpha Acetylene Slame) (APHA, 1992), whereas heavy metal analysis in water uses the APDC extraction method.

Water quality parameters were measured three times in each zone for each observation. Three observations were made. The average of the measurements is then obtained.. Data on heavy metal content in water, sediment, and soft tissue of crabs, as well as data on physical and chemical parameters of the waters, were analyzed descriptively, and the STORET (Storage and Retrieval of Water Quality Data System) method was designed to determine good and bad water quality in a reservoir or other water body for a specific water designation. Furthermore, this method can determine which parameters exceeded or did not meet the quality standard requirements (Canter, 1977). To determine the close relationship between the levels of heavy metals Pb and Cu in water, sediment, and wideng, regression and correlation analysis will be made (Manttjik and Sumertajaya, 2002). The correlation coefficient is calculated based on the following formula:

$$r = \frac{S_{xy}}{\sqrt{(S_x)^2 (S_y)^2}}$$

$$S_{xy} = \frac{\sum (xi - x)(Yi - y)}{n - 1}$$

$$S_x{}^2 = \frac{\sum (Xi - x)^2}{n - 1}$$

$$S_y{}^2 = \frac{\sum (Yi - y)^2}{n - 1}$$

Information: r = Correlation coefficient, Sxy = Distribution of observation values x and y and Sx2 = Diversity of x , Sy2 = Diversity of y values

Heavy Metal Bioaccumulation Factor Calculation : Bioaccumulation factors can be used to explain an organism's ability to accumulate heavy metals in its body from its environment. The bioaccumulation factor (KB) is the ratio of metal concentrations in the organism's body to metal concentrations in the surrounding environment (Connel and Miller, 1995). The bioaccumulation factor can be calculated using the equation below.:

$$K_B = \frac{K_1}{K_2}$$

Note: KB = Bioaccumulation factor, K_1 = Heavy metal content in biota (ppm) and K_2 = Heavy metal content in the water environment (ppm) or in the sediment (mg/kg)

RESULTS AND DISCUSSION

Wideng crabs obtained at the study site ranged in length from 10 to 21 cm, width from 4-5 cm, and weight from 35 to 72 grams, with the majority being male. The water conditions were rich in nutrients when the Wideng crab (crustácea) was sampled during the spawning (breeding) period.(Garrabou, 1997)

Heavy Metal Pb and Cu Distribution in Upstream and Estuary Sediments.

The analysis of the heavy metal content of lead (Pb) in the sediment in the upstream area, as shown in Table 2, revealed a value in the range of 16 mg/kg – 20 mg/kg, with an average value of 17.6 mg/kg. The values in the estuary range from 12 mg/kg to 17 mg/kg, with an average of 14.6 mg/kg. Table 2 shows the results of an analysis of heavy metal levels of Copper (Cu) in sediments, with upstream values ranging from 28 mg/kg to 35 mg/kg, with an average value of 30.3 mg/kg. The values in the estuary range from 24 mg/kg to 30 mg/kg, with an average of 26.1 mg/kg.

Table 2. Tin (Pb) and Copper (Cu) concentrations in Gonjol river sediments at various locations and conditions (mg/kg dry basis).

Condition	Snippets	Unit	Upstream		Estuary		Information
			Pb	Cu	Pb	Cu	
High tide	1	mg/Kg	17	31	12	27	Dry Weather
	2	mg/Kg	17	29	14	30	Rainy weather
	3	mg/Kg	16	35	17	26	Rainy weather
Low tide	1	mg/Kg	18	28	15	25	Dry Weather
	2	mg/Kg	19	32	16	24	Rainy weather
	3	mg/Kg	20	29	16	27	Rainy weather
Minimum		mg/Kg	16	28	12	24	
Average		mg/Kg	17.8	30.7	15.0	26.5	
Maximum		mg/Kg	20	35	17	30	
Standard deviation		mg/Kg	1.47	2.58	1.79	2.07	

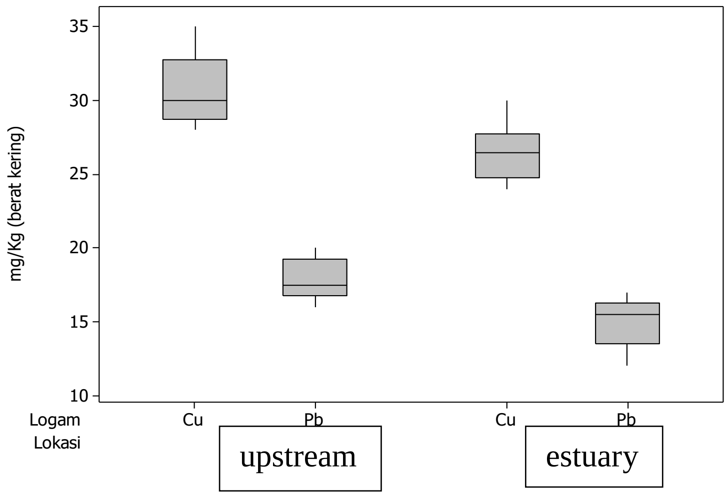

Figure 2: Heavy metal Pb and Cu box plot in sediment

Pb has the property of accumulating in dissolved organic matter in the waters, the results of Table 2 show that the average value of Pb levels in sediments in the river's estuary is lower than the levels of Pb in upstream areas. Because of the mixing of seawater at high tide and fresh water from upstream via the upwelling process, the nature of the accumulation or binding is relatively more vulnerable. This is due to the fact that Pb levels in Muara are relatively more stable than in the upstream. Meanwhile, heavy metal levels in the upstream are more variable depending on the mass of water coming from the watershed, as a result, heavy metal leaching of Pb and Cu occurs only when the water mass is present.(Žibret and Čeplak, 2021) The greater the distance from the pollutant source, the lower the concentration of heavy metals carried by rivers, some of which have been deposited during the trip to the sea, but because heavy metals are bioaccumulative, they will be quite dangerous if exposed to the environment for an extended period of time. (Ali, Khan and Ilahi, 2019) In general, low levels of both metals are common during the first sampling period. This is thought to have happened as a result of the weather conditions at the time of sampling. The season is still summer in the first observation, so the water discharge in the waters is relatively small. Meanwhile, the second and third samplings were conducted during rainy weather conditions, which meant that the water discharge was greater than usual, reducing the possibility of heavy metal accumulation in the sediment.

Heavy metal levels in sediments are higher, which is due to the nature of heavy metals in water, which settle for a period of time and then accumulate on the bottom of the water. (Prayoga, Hariyadi and Effendi, 2021) The main factor in the distribution of heavy metals into waters originating from river flows is tides in the water.. (Kucuksezgin, Uluturhan and Batki, 2008) The heavy metal content in sediments was found to be much higher than that dissolved in the water column. Because heavy metal elements have a higher density than water, they are likely to precipitate (Tchounwou *et al.*, 2012) The type of sediment influences the accumulation of heavy metals in the sediment. The sediment in the waters of the river Gonjol Pandansari Demak is a type of mud that appears black. This condition also demonstrates the presence of high levels of heavy metals. (Wardani, Prartono and Sulistiono, 2020). Some metals are commonly found in the form of particulates. The formation process begins with the binding of a number of heavy metals in the top layer by suspended particulates from the water column. When the particulates reach a density greater than that of water, gravity will deposit them and they will become part of the surface sediment. (Hamad *et al.*, 2012) Metals in solution will be absorbed to some extent by particulate materials. Particulate matter in the water will be deposited and become part of the surface sediment in a subsequent process. Gravity can play a role in the deposition process.(Remaili *et al.*, 2018) Some metals that are bound to sediments and particles that settle back into the water are remobilized and diffuse upward. Metal concentrations in the sediment will be affected by bioturbation.. (Lu *et al.*, 2019) Because the effect of bioturbation is the release of metals from the sediment into the water above the sediment surface, fluctuations in heavy metal levels in the sediment are thought to be influenced by the activity of enrichment and metal release in the sediment.(He *et al.*, 2017)

The Distribution of Heavy Metals Pb and Cu in Upstream and Estuarine Water

Rainfall and tides influence the chemical and physical properties of water in rivers and estuaries. As a result, the description of the analysis results differs from the results of the sampling during low and high tide conditions. The results of the analysis of the levels of heavy metal Tin (Pb) in the water, as shown in table 3, showed values ranging from 0.04 mg/L to 0.13 mg/L with an average value of 0.08 mg/L from three sampling times and at low tide conditions. showed a range of 0.05 mg/L – 0.18 mg/L with a mean value of 0.09 mg/L from three samplings. Meanwhile, at high tide in the estuary, the value ranges from 0.05 mg/L to 0.22 mg/L, with an average of 0.12 mg/L, and at low tide, the value ranges from 0.06 mg/L to 0.32 mg/L, with an average of 0.15 mg/L. The results of the analysis of heavy metal levels of Copper (Cu) in the water, as shown in Table 3, indicate a value in the range of 0.03 mg/L – 0.18 mg/L with an average value of 0.08 mg/L indicating a value in the range of 0.04 mg/L – 0.11 mg/L with a mean value of 0.07 mg/L. Meanwhile, at high tide in the estuary, the value ranges from 0.04 mg/L to 0.31 mg/L, with an average of 0.14 mg/L, and at low tide, the value ranges from 0.05 mg/L to 0.42 mg/L, with an average of 0.17 mg/L.

Table 3. Results of analysis of metal levels of Tin (Pb) and Copper (Cu) in water

Condition	Snippets	Unit	Upstream		Estuary		Information
			Pb	Cu	Pb	Cu	
High tide	1	mg/L	0.13	0.18	0.22	0.31	Dry Weather
	2	mg/L	0.07	0.03	0.09	0.04	Rainy weather
	3	mg/L	0.04	0.03	0.05	0.06	Rainy weather
Minimum		mg/L	**0.04**	**0.03**	**0.05**	**0.04**	
Average		mg/L	**0.08**	**0.08**	**0.12**	**0.14**	
Maximum		mg/L	**0.13**	**0.18**	**0.22**	**0.31**	
Standard deviation		mg/L	0.05	0.09	0.09	0.15	
Low tide	1	mg/L	0.18	0.11	0.32	0.42	Dry Weather
	2	mg/L	0.05	0.04	0.06	0.05	Rainy weather
	3	mg/L	0.05	0.06	0.07	0.05	Rainy weather
Minimum		mg/L	**0.05**	**0.04**	**0.06**	**0.05**	
Average		mg/L	**0.09**	**0.07**	**0.15**	**0.17**	
Maximum		mg/L	**0.18**	**0.11**	**0.32**	**0.42**	
Standard deviation		mg/L	0.08	0.04	0.15	0.21	

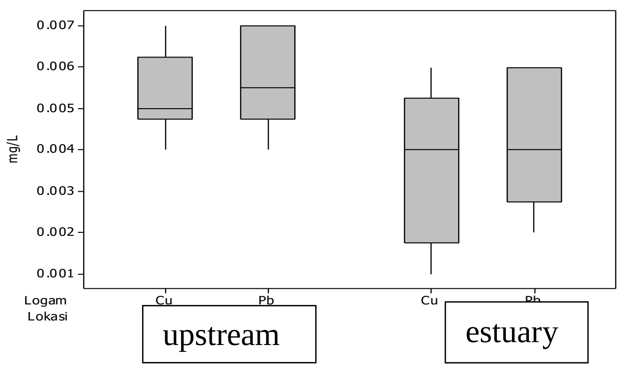

Figure 3: Concentration of heavy metals dissolved in water in a box plot

Cu and Pb levels in the estuary are higher than in the upper river. The same thing happened with the heavy metal Pb, which had a higher concentration in the estuary. There are concentration differences in heavy metal Pb with Cu at upstream and estuary locations. At the upstream and estuary locations, the concentration of heavy metal Cu is greater than the concentration of heavy metal Pb. This is because other nutrients are sent from the sea, which is directly adjacent to the research location, to the Muara location, where the heavy metal content is higher than in the Upstream. Because the estuary connects fresh water from rivers and salt water from the sea. Because fresh water from rivers has a lower density than sea water, it will float on top of the sea water. Each estuary has a distinct personality or nature. This variation is primarily due to changes in tidal and river water input, which affects a more effective mixing process in the water column. Because seawater is mixed up and fresh water is mixed down, the levels of heavy metals Pb and Cu in the estuary are higher than the levels of heavy metals Pb and Cu upstream. Pb and Cu levels in the water around the Gonjol river in the Pandansari area are thought to be caused by industrial activity in factories upstream of the river. Heavy metals are also produced by industries such as furniture painting, canning, the beverage industry, and printing near river mouths, which produce Pb and Cu metals.

The content of Cu metal in the upstream river is much lower during high tide conditions because the area is a closed area with no relatively large water movement, causing heavy metals to be deposited at the bottom of the water more quickly. As a result, the value of Cu metal in water differs between the two locations. (Tian *et al.*, 2020)

Cu belongs to the group of essential metals, and while it is required by organisms as a coenzyme in the body's metabolic processes at low levels, its toxic nature appears only at high levels. Aquatic biota is extremely sensitive to excess Cu in the bodies of water in which it lives. A dissolved Cu concentration of 0.01 ppm in sea water can kill phytoplankton. The death was caused by Cu toxicity, which inhibited the activity of enzymes involved in phytoplankton cell division. Crustacean species will die after a 96-hour grace period if the Cu concentration is between 0.17 and 100 ppm. (Prayoga, Hariyadi and Effendi, 2021) In the same period of time, mollusk biota will die if the dissolved Cu content in the water body in which the biota lives is between 0.16 – 0.5 ppm, and Cu levels of 2.5 – 3.0 ppm have been able to kill the fish. (Hindarti and Larasati, 2019) . Heavy metal levels of Pb at the research station were highest with low tide conditions in the first observation, with an average value of 0.32 mg/l, and lowest with high tide conditions in the third observation, with an average value of 0.04 mg/l. Meanwhile, Cu levels in water showed the highest results with low tide conditions in the first observation with an average value of 0.042 mg/l and the lowest value with high tide conditions in the third observation with an average value of 0.03 mg/l, according to the research station.

Pollution cases demonstrate that the release of heavy metals from human activities is significantly greater than that produced by natural processes (Briffa, Sinagra and Blundell, 2020). Because metallic elements are naturally present in very low concentrations in the sea, it can be seen that the sources of heavy metals originating from the sea have lower concentrations than those originating from industrial and household activities in the watershed. Furthermore, the cause of river estuaries has a relatively high level of heavy metals because the river estuary is the estuary for materials carried by the river's flow of water, As a result, it is assumed that the material carrying heavy metal waste will congregate at the river's mouth and will be mixed and diluted by sea water before being carried away by currents to the sea. The water circulation system is closely related to heavy metal levels that vary between the estuary and upstream of the river. Water exchange occurs due to the entry of river water containing heavy metals and sea water through the tide at the mouth of the Gonjol river, which has the condition of an open estuary area and the beach is characterized by a sand bottom. While the Gonjol River's base is a closed area characterized by an abundance of *Avicennia sp* and *Rhizopora* sp. In areas with closed water circulation, Pb metal will deposition and demineralize in the river bed layer. As a result, the Pb metal content was reduced significantly.(Suchara, 2019) The amount of pollution load carried by a pollutant (heavy metal) that enters a water area is highly dependent on the activity that liberates pollutant compounds (heavy metal) in the water area. Furthermore, the high load of pollutant (heavy metals) entering the waters can be supported by the density of the population. Thus, differences in heavy metal levels at the research sites are thought to be caused by differences in the background conditions of each research location, which include natural characteristics of an area, population levels, and human activities.

Heavy Metal Pb and Cu Distribution in the Upper and Estuary of the River

Table 4 shows the results of an analysis of the heavy metal content of Pb in tissue; in the upstream, it ranges from 0.13 mg/kg to 0.27 mg/kg, with an average value of 0.20 mg/kg, while in the estuary, it ranges from 0.04 mg/kg to 0.29 mg/kg, with an average value of 0.17 mg/kg. Table 4 shows the results of an analysis of heavy metal levels of Copper (Cu) in tissues, with upstream values ranging from 0.07 mg/kg to 0.16 mg/kg, with an average value of 0.09 mg/kg. The values in the estuary range from 0.02 mg/kg to 0.13 mg/kg, with an average of 0.06 mg/kg..

Table 4. Results of analysis of metal concentrations of Lead (Pb) and Copper (Cu) in tissue (dry basis)

Condition	Snippets	Unit	Upstream		Estuary	
			Pb	Cu	Pb	Cu

High tide	1	mg/Kg	0.17	0.08	0.09	0.02
	2	mg/Kg	0.27	0.09	0.29	0.07
	3	mg/Kg	0.19	0.07	0.21	0.05
Low tide	1	mg/Kg	0.13	0.11	0.04	0.06
	2	mg/Kg	0.18	0.16	0.29	0.13
	3	mg/Kg	0.27	0.07	0.24	0.06
Minimum		mg/Kg	0.13	0.07	0.04	0.02
Average		mg/Kg	0.20	0.10	0.19	0.07
Maximum		mg/Kg	0.27	0.16	0.29	0.13
Standard deviation		mg/Kg	0.06	0.03	0.11	0.04

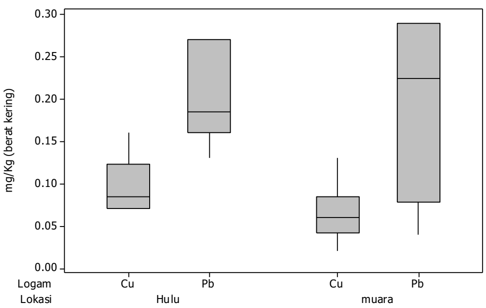

Figure 4. Box Plot of Heavy Metal Content in Tissue

Heavy metal levels of Pb in the body tissue of Wideng (Episesarma sp.) showed the highest results in the upstream with an average value of 0.20 mg/kg, despite the fact that one of the data from the sampling results in the estuary had a value of 0.29 mg/kg. What matters, however, is the sum of three sampling repetitions. The estuary had the lowest value, with an average of 0.07 mg/kg. Meanwhile, Cu metal in wideng body tissue (Episesarma sp.) showed the highest results in the upstream with an average value of 0.16 mg/kg and the lowest value in the estuary with an average of 0.02 mg/kg. These findings suggest that the wideng caught upstream accumulated more Pb and Cu from pollutant sources than those caught in the estuary, where fresh water meets sea water, either directly through predation and digestion or indirectly through metabolism and molting. Because the estuary was relatively more closed and heavily influenced by fresh water, the lower levels of Pb and Cu in the body tissue of the wideng crab caught in the Gonjol River's estuary area were related to the tidal washing system. Heavy metal absorption by aquatic organisms begins with rapid absorption in cell membranes, followed by a rate of uptake that is taken up by diffusion and then bound by proteins, either through food digestion or molting.(Rehman *et al.*, 2021) The accumulation of heavy metals Pb and Cu in animal tissues is caused by the biota's life cycle, digestive system, and growth process. The process of taking food can predict the accumulation of heavy metals in crab soft tissues. The food will be forwarded to the digestive tract and undergo a metabolic process after being filtered by the gills. Nutrients that are required will be directly absorbed by the tissues and forwarded to the cell membrane, whereas substances that are not required will be excreted along with the results of digestion and urine. Heavy metals that are bound in food substances are absorbed during the absorption process. Variations in heavy metal levels in soft tissues are thought to be caused by differences in heavy metal levels from the source as well as organism-specific factors such as age, body size, growth rate, feeding rate, and individual ability to accumulate, excrete, and detoxify heavy metals. (Bonsignore *et al.*, 2018)

Metal Content in Wideng Crab Tissue Correlates with Heavy Metals Pb and Cu in Sediment

Based on the regression analysis of the Pb metal content data at the Upstream location, the value of r = 0.395 with the regression equation is $Y = 205.08x^2 + 79.285x – 24,932$, where X is the Pb content in the sediment and Y is the Pb content in the wider network (Figure 4). This means that there is no close relationship between heavy metal Pb found in crabs and those found in sediments upstream. (fig 4)

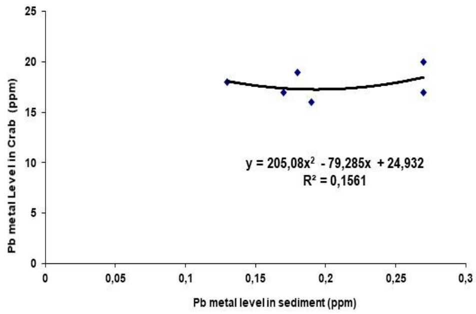

Figure 5. Pattern of regression relationship between Pb content in sediment and wideng crab tissue. (Upstream location)

Based on the regression analysis of the Pb metal content data at estuary, the value of r = 0.4340 with the regression equation is $Y= -0.005X^2 – 0.1681X – 1.195$, where X is the Pb content in the sediment and Y is the Pb content in the wideng crab tissue. This means that there is no close relationship between the heavy metal Pb in the sediment and the heavy metal Pb content in Wideng crab tissue at the estuary site. Estuary river is situated on a heavy metal dump that is directly adjacent to the river. Because of the nature of the Wideng crab, there aren't many of them in the estuary because if the nutrients in the water are depleted, the Wideng crab dies (fig 6)

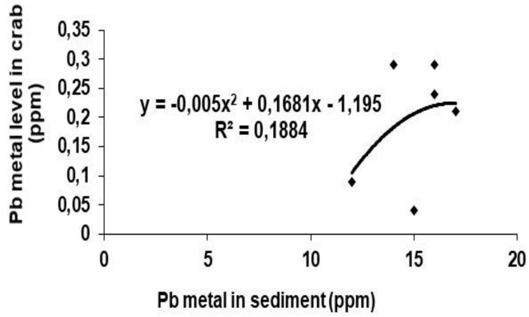

Figure 6. The pattern of regression relationships between Pb levels in sediments and those in wideng crab tissues (estuary location)

Meanwhile, for Cu metal in the Upstream, the value of r = 0.3912 with the regression equation Y = -0.0025X2 + 0.157X - 2.353, where X and Y are Cu content in sediment and network widening, respectively (Figure 6). This means that there is no direct relationship between the influence of heavy metal Cu in the sediment and heavy metal Cu levels in the Wideng crab tissue. Heavy metal Cu is deposited in crustacean shells because the Wideng Crab requires heavy metal Cu for shell hardening. In this study, tissue from the Wideng crab was taken, and the content of heavy metal Cu was obtained in a small amount compared to heavy metal (Pb)

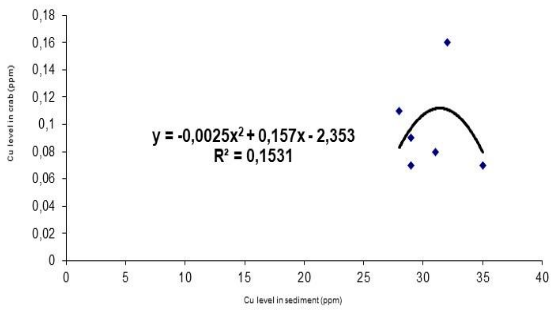

Figure 7. Pattern of regression relationship between Cu content in sediment and wideng crab tissue. (Upstream location)

The correlation test value of Cu metal in estuary to sediments with Wideng has a correlation value of r = 0.8968 with the regression equation $Y = 0.0068X^2 – 0.3736X + 5.1867$, where X and Y are the Cu content of the sediment and the widening tissue, respectively (Figure 8). It means that the heavy metal content of Cu in the sediment is closely related to the heavy metal content of Cu in the Wideng network. The obtained correlation is positive, indicating that if the heavy metal content of the Wideng crab is high, high levels of heavy metal in the sediment will follow.

The results of the analysis revealed that heavy metal Pb levels in sediments in the upstream and estuary affected heavy metal levels in crab tissue more than heavy metal Cu levels in the upstream and estuary due to crabs absorbing more heavy metal Cu for the formation of cuticles/scales in crustaceans.

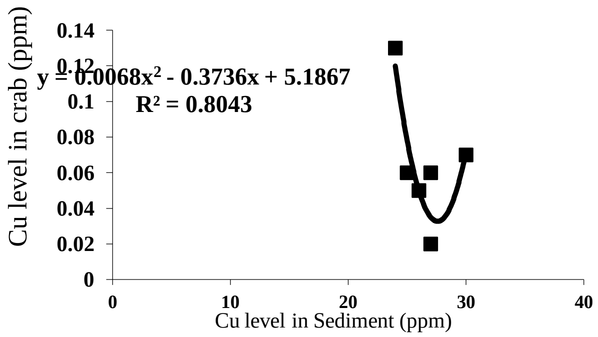

Figure 8. Pattern of regression relationship between Cu content in sediment and wideng crab tissue. (Estuary location)

Pb and Cu Heavy Metal Rates in Wideng Crab Tissue Correlation with Metals in Water

The correlation test for Pb metal content in upstream waters and wideng crab tissue yielded a value of 0.7338, and the regression equation is $y = -10.576x^2 + 1.6379x + 0.1669$. (Figure 9) where X is the amount of Pb in the water and Y is the amount of Pb in the tissue This means that there is a close relationship between heavy metal levels in the waters and heavy metal levels in the wideng crab tissue in the estuary. The obtained correlation is negative, which means that if the heavy metal content of the Wideng Crab is low, the heavy metal content of the water will be high, and vice versa.

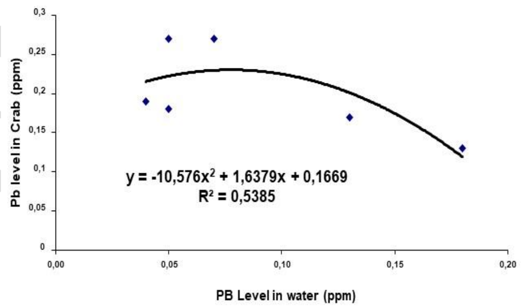

Figure 9. Pattern of regression relationship between Cu content in sediment and wideng crab tissue. (Upstream)

The correlation coefficient for heavy metal Pb in water to crabs in the estuary area is r = 0.949, with the equation y = -3.361x2 – 2.0971x + 0.3654, where x and y are the levels of Pb in water and crab tissue, respectively broadening (figure 10). The obtained correlation is negative, which means that if the heavy metal content of the Wideng crab is high, the heavy metal content of the water will be low and vice versa. This indicates that there is a close relationship between the heavy metal Pb in the water and the heavy metal Pb content in the Wideng tissue. The obtained correlation is negative, which means that if the heavy metal content of the Wideng crab is high, the heavy metal content of the water will be low and vice versa. This implies that there is a close relationship between the heavy metal Pb in the water and the heavy metal Pb content in Wideng crab tissue. Because a large amount of heavy metal Pb is deposited in the tissue, the heavy metal content obtained in the estuary area Wideng crab tissue can be compared to the heavy metal content of Pb in the water. The graph shows that as the heavy metal content of the water increases, so does the heavy metal content of the Wideng Crab tissue . This is because the Wideng Crab absorbs more food from the sediment because it is detritus..

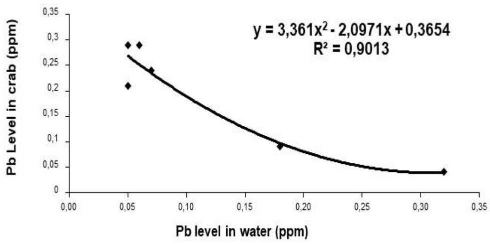

Figure 10. Pattern of regression relationship between Pb content in sediment and wideng crab tissue. (Estuary)

The correlation regression correlation of heavy metal Cu concentration in the upstream in water and crabs has a positive value of r = 0.244 with the regression equation y = -2.7975x2 - 0.4827x + 0.0845 where x and y are Cu levels in water and wideng crab tissue, respectively (Fig. 11). This demonstrates that there is no close relationship between water and heavy metal accumulation in wideng crab tissue. As a result, an increase in the accumulation of heavy metals in the water is not followed by an increase in the concentration of heavy metals in the wideng crab tissue.. This is because the heavy metal Cu accumulates in the wideng crab's shell because it is required for the formation of chitin..

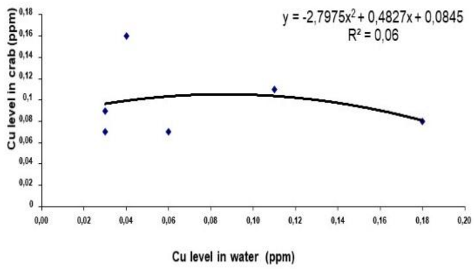

Figure 11. Pattern of regression relationship between Cu content in sediment and wideng crab tissue. (Upstream)

The correlation regression correlation of heavy metal Cu in the estuary and the wideng crab tissue is positive (r) = 0.658, with the regression equation $y = 1.6176x^2 – 0.8066x + 0.1138$, where x and y are Cu levels in water and wideng crab tissue, respectively (Fig. 12). This demonstrates that there is no close relationship between Cu concentration in water and heavy metal accumulation in wideng crab tissue, so an increase in heavy metal accumulation in water does not result in an increase in heavy metal concentration in wideng crab tissue..

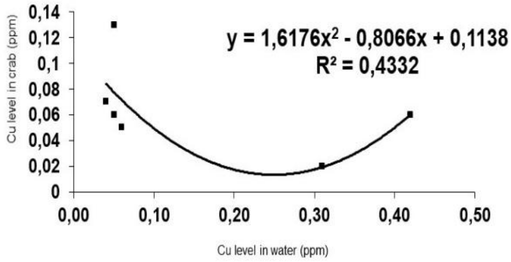

Figure 12. Pattern of regression relationship between Cu content in sediment and wideng crab tissue. (Estuary location)

Correlation of Pb and Cu Heavy Metal Content in Sediment and Water

The regression analysis results on the two types of heavy metals, namely Pb and Cu, based on the research location, upstream and estuary, are as follows. The correlation coefficient value (r) = 0.7130 was based on the results of the regression analysis of the Pb metal content data at the upstream location of the river, with the regression equation y = 0.0023x2 + 0.8265x – 7.2901 where x is the Pb content in the water and y is the Pb content in the water (Figure 13). The obtained correlation is negative, indicating that if the heavy metal content in the water is high, the heavy metal content in the sediment is relatively low. This means that heavy metal levels in the water are closely related to heavy metal levels in the sediment. The trend graph shows a peak followed by a decrease, which is caused by the influence of tides and conditions in the Upper River where there is no flow for water exchange.

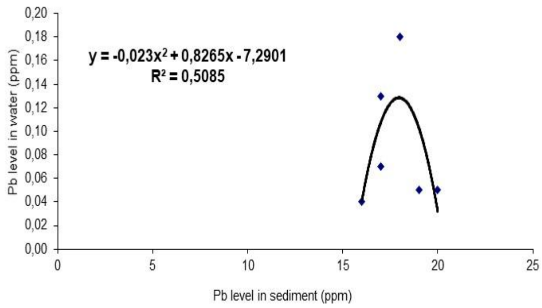

Figure 13. Pattern of regression relationship between Pb levels water and sediment (Upstream)

The correlation coefficient value (r) = 0.4384 with the regression equation y= $-0.0095x^2$ + 0.2513 – 1.4892, where x and y are the levels of Pb in sediment and water, respectively, according to the results of the correlation test value on water with heavy metal sediment Pb in the estuary (See Figure 14). This demonstrates that in the estuary, there is no close relationship between water and sediment. As a result, an increase in heavy metal accumulation in the water will not be followed by an increase in heavy metal accumulation in the estuary's sediments. This is due to the fact that the estuary receives a plethora of different sources from the sea and surrounding ponds, making it impossible to estimate how much pollutant affects the estuary.

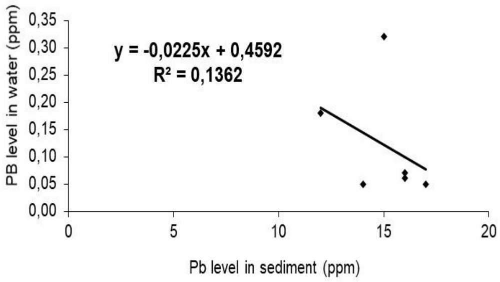

Figure 14. Pattern of regression relationship between Pb levels in water and sediment (estuary location)

Heavy metal regression correlation correlation Cu content in water and sediment upstream is positive (r) = 0.3971, as calculated by the regression equation y= -0.0033x^2 + 0.204x – 3.0277, where x represents cu content in water and y represents cu content in sediment (Figure 15). This demonstrates that there is no close relationship between heavy metal accumulation in water and sediment upstream of the river, so that an increase in heavy metal accumulation in water is not followed by an increase in heavy metal concentration in sediment.

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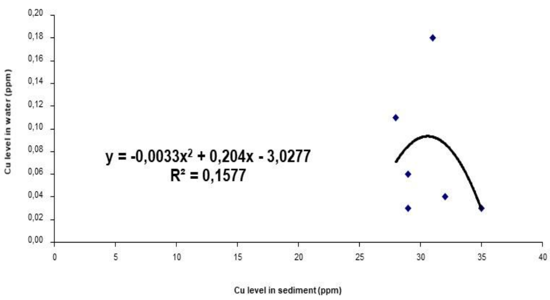

Figure 15. Pattern of regression relationship between Cu content in water and sediment (Upstream location)

The correlation coefficient between heavy metal concentrations of Cu in river mouths in sediments is (r)=0.3823, with the regression equation y= -0.0106x2 + 0.5527 – 7.0008 where x and y are Cu levels in sediment and water, respectively (Figure 16). This demonstrates that there is no close relationship between heavy metal concentrations in water and in sediment, so that an increase in heavy metal accumulation in water is not followed by an increase in heavy metal concentration in sediment..

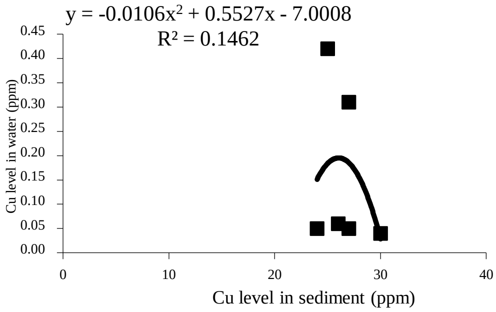

Figure 16. Regression relationship pattern between Cu content in water and sediment (Estuary location)

The interaction of Pb in the water and in the Wideng crab (estuary location) indicates that the heavy metal Pb has a greater chance of accumulating in the Wideng crab tisue at high tide. This is closely related to the wideng crab's metabolic system and predation. Metabolism occurs when water enters the body through the gills and is used as a source of respiration by taking oxygen.. Hemoglobin will bind oxygen and transport it to all body tissues. At the same time, the heavy metal Pb in the media will be carried into the bloodstream and deposited in various body tissues. The same thing happens when the wideng crab hunts for food; the media water enters the digestive tract and is deposited in several body tissues as a result of the process (Hutagalung 1994). All data generated on heavy metals Pb and Cu in the estuary at the same time between sediment and water have an effect on heavy metals in the body. Water containing trace amounts of heavy metals that have a negative impact on tissue. . The higher the heavy metals in the crab tissue, the lower the heavy metals in the water. Heavy metals present in body tissues are said to enter through food carried by the flow of water and be captured by these biota.

Factors of Concentration and Comparison of Pb and Cu Heavy Metal Levels with Quality Standards

If there are 100 ppm levels of heavy metal Pb in the water, the levels of heavy metal Pb in sediments in the upstream river are around 39 ppm at high tide. Meanwhile, at high tide, wideng crabs can accumulate heavy metal Pb concentrations of up to 88.430 ppm in sediment (Table 5). The accumulation of heavy metal levels of Pb and Cu in bioconcentration conditions at the river's mouth is higher than upstream. This is due to the fact that the bioaccumulation factor of heavy metals in the estuary is higher because the estuary is a confluence of seawater flows and river water, so metal concentrations are higher. The weight settles in the area, and the sediment undergoes bioconcentration. The bioaccumulation factor of heavy metals is greater at high tide than at low tide.

Table 5 . Heavy Metal Concentration Factors

Sample	W	A	S	FK			W	A	S	FK		
	(ppm)	(ppm)	(ppm)	AW	SW	SA	(ppm)	(ppm)	(ppm)	AW	SW	SA
Upstream (High tide)	0.202	0.080	17.833	0.397	88.430	0.004	0.097	0.080	30.667	0.828	317.241	0.003
Upstream (low tide)		0.093		0.463		0.005		0.070		0.724		0.002
Estuary (High tide)	0.193	0.120	15.000	0.621	77.586	0.008	0.065	0.137	26.500	2.103	407.692	0.005
Estuary (low tide)		0.150		0.776		0.010		0.173		2.667		0.007

Information :
W : Wideng Crab, SW : Sediment to Wideng, S : Sediment, FK : Concentration Factor, A : Water, SA : Sediment to Water. AW : Water against Wideng

Heavy metal bioaccumulation in aquatic animals is a complex process that is not fully understood (Wang and Rainbow, 2008) Certain materials (including metals) can accumulate in the body tissues of aquatic animals in four ways: through the flow of water in the gills, the process of eating and drinking, and the skin (Mohammed, Kapri and Goel, 2011) Metal accumulation in aquatic animals begins with taking (uptake) through the gills, which is then absorbed into all body tissues and stored / trapped inside. Several factors influence the process of 'uptake' of heavy metals and the amount that accumulates. These factors include metabolic rate, size and type, alkalinity, and pH. Furthermore, the demethylation process, temperature, level of contamination, time, source and form of heavy metals, and the organism's life stage all have a significant impact on the uptake process (Canli and Furness, 1993) approximately 70% of heavy metals ingested through food are absorbed into the body tissues of aquatic animals, while only 10% are absorbed through the gills.(Bosch *et al.*, 2016)

Water Quality and Heavy Metal Content

According to the findings of the analysis, the levels of heavy metals Pb and Cu in the water in the study area from upstream to the river's mouth, namely the Pandansari River estuary, place the research area in category C, which is moderately polluted by heavy metals Pb and Cu. According to Minister of Environment Decree 115 of 2003, guidelines for determining water quality can use the STORET method or the Index method, so the status of water quality at the Gonjol river estuary is tidal conditions.

Table.6 Status of water quality according to the STORET value system at the Gonjol river estuary tidal and low tide conditions and upstream at high tide and low tide conditions

No.	Parameter	Unit	Standard	Measurement Results			Score	Measurement Results			Score
				Max	Min	Avg		Max	Min	Avg	
Physics											
1	Temperature	Celcius	Normal	29.63	29.63	29.63	0	29.63	29.63	29.63	0
2	Brightness	MeterS	± 3	0.29	0.27	0.28		0.29	0.27	0.28	
Chemical	Water Temperature										
1	Pb	mg/l	0.03	0.22	0.05	0.12	-10	0.32	0.06	0.15	-10
2	Cu	mg/l	0.02	0.31	0.04	0.14	-10	0.42	0.05	0.17	-10
3	DO	mg/l	>3	3.18	2.12	3	0	3.18	2.12	3	0
4	pH		6-8.5	7	7	7	0	7	7	7	0
5	Salinity	0/00		33	32	32.5	0	33	32	32.5	0
6	PO_4	mg/l		0.7	0.6	0.65		0.7	0.6	0.65	
7	NO_3	mg/l		5	4	4.5		5	4	4.5	
	Total score						-20				-20
No.	Parameter	Satuan	Standard	Measurement Results			Skor	Measurement Results			Skor
				Max	Min	Avg		Max	Min	Avg	
Physics											
	Temperature	Celcius	normal ± 3	29.63	28.2	28.915	0	29.63	28.2	28.915	0
	Brightness	Meter		75.33	29.67	52.5		75.33	29.67	52.5	
Chemical											
1	Pb	mg/l	0.03	0.18	0.05	0.09	-10	0.18	0.05	0.09	-10
2	Cu	mg/l	0.02	0.11	0.04	0.07	-10	0.11	0.04	0.07	-10
3	DO	mg/l	>3	3.73	2.38	3.055	0	3.73	2.38	3.055	0
4	pH		6-8.5	7	7	7	0	7	7	7	0
5	Salinity	0/00		34	32	33	0	34	32	33	0
6	PO_4	mg/l		0.8	0.5	0.7		0.8	0.5	0.7	
7	NO_3	mg/l		6	4	5		6	4	5	
	Total score						-20				-20

Based on table 6 then the total score is -20, which means that the Gonjol river at station 1 is classified as category C, moderately polluted by heavy metals Pb and Cu. Heavy metals that enter and accumulate in the crab's body can be released through a release mechanism assisted by metabolic processes.

Based on the decree of the Director General of Drug and Food Control no. 03725/B/SK/VII/1989 of 1989 concerning the maximum limit of metal contamination in food for fish and its preparations can be seen in table 7, that the metal content in soft tissue of crabs, for Pb and Cu metals at all stations is below the threshold. (Mallongi, 2014) (Dirjen POM, 1989)

Table 7. Heavy metal levels of Pb and Cu in crab soft tissue compared to the decree of the Director General of National Agency of Drug and Food Control of Indonesia

Heavy Metal	Location	Station	Concentration (mg/kg)	Consumption Limit (mg/kg)	Information
Pb	Upstream	1	0,2		Below the threshold
	Estuary	2	0,17	2	Below the threshold
Cu	Upstream	1	0,09		Below the threshold
	Estuary	2	0,06	2	Below the threshold

According to the FAO, USFDA, and NAS-NRC maximum limit for heavy metal entry into the human body (Sivaperumal et al., 2005), the metal content in crab soft tissue for Pb and Cu metals is below the consumption threshold..

Table 8. Heavy metal concentration of Pb and Cu in Crab soft tissue compared with maximum limit for heavy metal entry into human body (Jaworska and Lemanowicz, 2019) (Sivaperumal *et al*. 2005)

Heavy metal	Location	Station	Concentration (mg/kg)	Comsumption limit (mg/kg)	Information
Pb	Upstream	1	0,2		Below the threshold
	Estuary	2	0,17	0,5	Below the threshold
	Upstream	1	0,09		Below the threshold
Cu	Estuary	2	0,06	30	Dibawah ambang batas

The levels of heavy metals in the sediments in the study area for Pb and Cu metals are still below the quality standard threshold at all stations.(Peng, 2015)

Table 9. Heavy metal content of Pb and Cu in sediment compared with quality standard value

Heavy Metal	Location	Station	Content at location (mg/kg)	Consumption Limit (mg/kg)	Information
Pb	Upstream	1	17,6		Below the threshold
	Estuary	2	14,6	30,24	Below the threshold
	Upstream	1	30,3		Below the threshold
Cu	Estuary	2	26,1	18,7	Below the threshold

DISCLAIMER

All research materials used in this study are commonly used materials, there is no conflict of interest between the author and any party for litigation, this research is only intended for the advancement of knowledge, and this research is financed by the author's personal efforts. DATA

AVAILABILITY

All relevant data has been registered with supporting file information. This research will help researchers to uncover critical areas related to The Characteristics Of Heavy Metal Correlation Pb And Cu Episesarma Sp (Wideng Crab) Tissue In Industrial Wastewater In Gonjol River, Demak

CONSENT

The research was carried out in accordance with research standards that apply in the Republic of Indonesia, written consent has been collected and kept by the author

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There

is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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