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Plankton diversity and its heavy metal content in Ratai Bay of Pesawaran district, Lampung, Indonesia

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ABSTRACT

Artisanal and small-scale gold mining (ASGM) activity in Way Ratai River produces heavy metal wastes; therefore, further information regarding heavy metal concentrations in the water was needed, especially in plankton samples. Furthermore, the determination of plankton diversity was also carried out in the waters of Way Ratai to determine the bioconcentration factor (BCF). Eight sampling sites were chosen along the river reaching the coast of Way Ratai. The research was conducted in November 2020 and March 2021. Ten heavy metals, Ag, Cd, Co, Cr, Cu, Fe, Mn, Pb, and Zn that are commonly found in mining areas, were determined in the water and plankton samples by using ICP-OES. The results indicated that the highest concentration found was Fe in plankton samples (0.725 mg/L in the river and 1.294 mg/L on the coast). Meanwhile, contents of Cd, Cu, Fe, Mn, and Zn in the river exceeded the predetermined water quality standards, while Ag and Pb metals were not detected. The Cd, Cr, Cu, Pb, and Zn content in seawater also exceeded quality standards. The highest BCF value (12.96) was found for Fe at station G, whereas the lowest BCF value (0.13) was found for Ag at stations G and H.

Key words: bioconcentration factor, fresh-coastal water, heavy metal, Pesawaran district, plankton diversity, Way Ratai

HIGHLIGHTS

- Plankton and water samples were collected from the river and coastal areas of Way Ratai.
- 77 and 36 types of plankton were found in freshwater and coastal water, respectively.
- Seven heavy metals, namely Fe, Mn, Zn, Cd, Cu, Cr, and Pb, were found to exceed the quality standard set by the Government Regulation of the Republic of Indonesia and the Regulation of the Minister of the Environment.

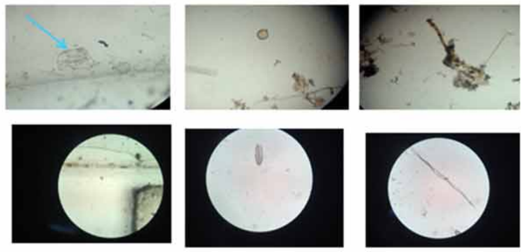
GRAPHICAL ABSTRACT

Pesawaran District

Ratai Bay

Sample Station	Metal (mg/L) Ag	Cd	Co	Cr	Cu	Fe	Mn	Ni	Pb	Zn
A	nd	nd	0.011	nd	0.017	0.318	0.052	nd	nd	0.065
B	nd	nd	0.008	nd	0.007	0.548	0.105	nd	nd	0.030
C	nd	0.005	0.007	nd	nd	0.616	0.068	0.025	0.038	0.028
D	nd	0.007	nd	nd	nd	0.291	0.087	0.018	nd	0.036
E	0.007	0.007	0.020	nd	nd	0.090	0.023	0.064	nd	0.078
F	0.011	0.005	0.035	nd	nd	0.196	0.037	0.080	0.080	0.049
G	0.005	0.009	0.017	nd	0.008	0.726	0.070	0.035	0.058	0.054
H	0.005	0.006	0.018	nd	nd	1,248	0.179	0.082	0.103	0.040

Heavey metal content



Example of plankton observed

INTRODUCTION

Pesawaran Regency, especially the area around Way Ratai, has a high risk for heavy metal exposure due to the artisanal small-scale gold mining (ASGM) activities. These activities have existed for a long time from the higher areas to the lower land of Way Ratai, and are vulnerable to exploitation by mining companies with or without permits. Based on observations conducted by the Directory of Mining and Energy Companies in Lampung, there are several active mining and excavation process units in Pesawaran. The gold mining process unit is one such unit located around the river banks of the Way Ratai River (Knedi & Rochayatini 2018).

Similar to industrial areas, most of the ASGM areas contribute to pollutant wastes, such as toxic compounds of heavy metals (Abidjulu 2008; Parhizkar *et al.* 2021; Taufiq *et al.* 2022). Heavy metal contamination is very harmful to the environment in the natural ecosystem and to humans. Some heavy metals can be transformed into persistent metal compounds with high levels of toxicity. Furthermore, these heavy metals accumulate in organisms and enter the food chain, and ultimately affect human health (Mahipal *et al.* 2016). Once the heavy metals enter water ecosystems, rivers, or oceans, they become major pollutants, due to their characteristics such as persistency, high toxicity, and bioaccumulation properties (Wang *et al.* 2018; Li *et al.* 2020). Some studies also indicated that some physicochemical parameters exceed permitted concentrations in the polluted river caused by discharging chemicals such as Hg, Cd, and cyanide used in gold mining (Martin *et al.* 2020).

Regarding the heavy metals released into the aquatic ecosystem, the health of aquatic biota is directly influenced by the heavy metals, which also have an incline to bioconcentration in different plant and animal tissues (Ernawati 2014; Ndome *et al.* 2014; Riani *et al.* 2014; Luo *et al.* 2022), where through the food chain it is possible for the poisonous heavy metals to be absorbed into human tissues. Other than that, many people living around the river bank of Way Ratai are still depending on the water resources of the river itself, such as for agriculture and households. Meanwhile, discharge water as well as brine containing heavy metals degrades water quality and thus water cannot be directly used for potable water via desalination and industrial applications (Panagopoulos 2022). Therefore, it is necessary to determine the heavy metal concentration in the water of Way Ratai; besides, there have been no related studies conducted in this area.

Planktons are microorganisms floating along the water currents (Dianthani 2003), making them easily exposed to heavy metals from the surrounding area. Planktons have a great opportunity to accumulate heavy metals because their surface area is larger than their volume ratio, so it has a high accumulation ability in a relatively short time. Therefore, plankton is used as a determining factor for heavy metal transportation in water (Whitfiled 2001). In addition, plankton being a primary source of food is also one of the main components in the food chain system and food web in the waters. The diversity of plankton needs to be considered as it can also be used to determine the water quality level. Therefore, the aims of this study were to determine the diversity of plankton in the Way Ratai as well as its heavy metal content and the heavy metal concentration in the water, from which their bioconcentration factor (BCF) in the plankton could be calculated.

MATERIALS AND METHODS

Study area

This research was conducted in November 2020 and March 2021 during the rainy season and samples were taken twice during that time period. The sampling of water and plankton was carried out at eight observation stations, with four stations in the river flow around Bunut Seberang village, Way Urang village, Kephong Jaya village, Sanggi Pematang Awi village, and four stations from the estuary to the coast of Teluk Ratai (Figure 1, Table 1). The first station (station A) was taken at the site where the ASGM (in the riverbank) activity has been done, then the following station was taken afterwards (from stations A to C) with a distance of approximately 500 m, but stations C to D were taken at a much greater distance. The identification of plankton was carried out at the Zoology Laboratory of the Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung, Indonesia. The sample preparation was carried out at the Biomolecular Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, University of Lampung, while the sample analysis of heavy metal material was carried out at the Integrated Laboratory and Technology Innovation Center (LTSIT), University of Lampung.

Tools and materials

Plankton net with a mesh of 25 μm was used to collect plankton. Cool box, 600-mL plastic bottle, zipper plastic, filter paper, tissue, test tube, measuring cup, dropper, object glass, microscope, and thermometer were used to collect and prepare samples. Global Positioning System (GPS) was used to determine the coordinates of the sampling location, and ICP-OES

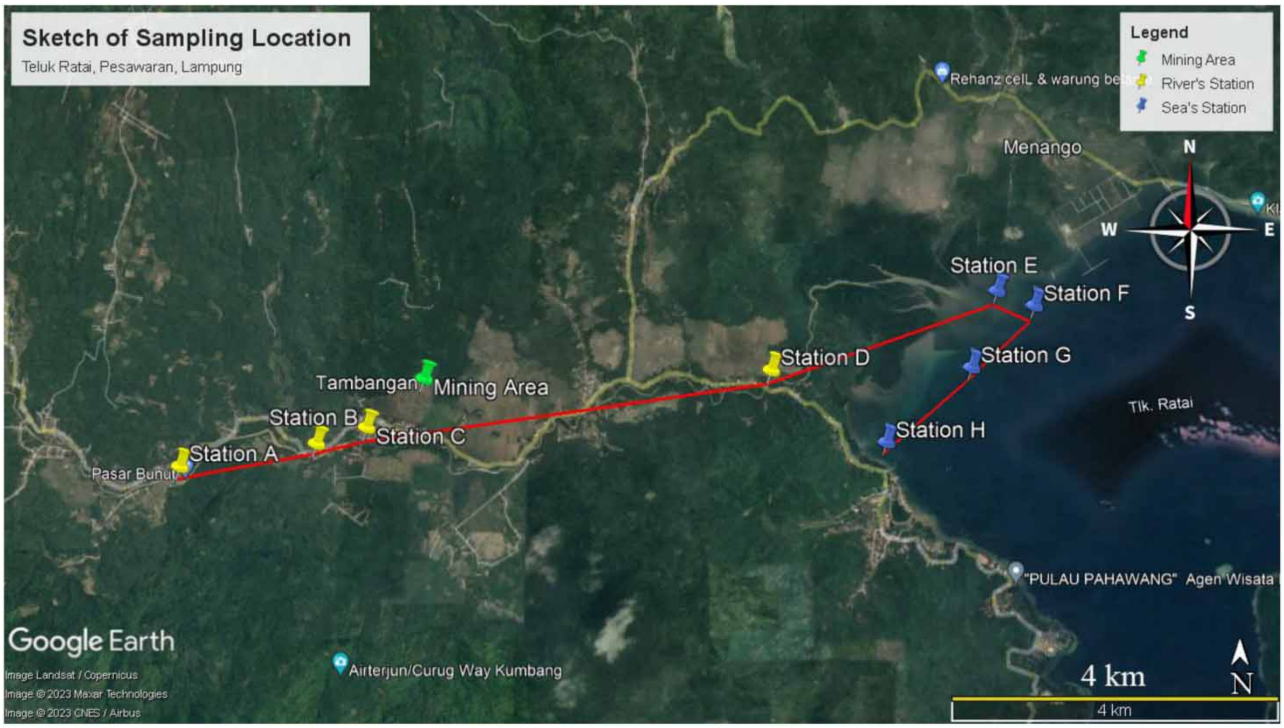


Figure 1 | Sketch of the sampling location.

Table 1 | Sampling coordinates

Station	Location	South latitude	East longitude
A	Bunut Seberang Village River	5°36′39.5″S	105°05′31.5″E
B	Khepong Jaya Village River	5°36′24.0″S	105°06′46.0″E
C	Way Urang Village River	5°36′30.4″S	105°06′26.0″E
D	Sanggi Village River Pem. Awi	5°36′24.0″S	105°06′29.0″E
E	Estuary	5°35′29.0″S	105°11′01.0″E
F	Ratai Bay Coast	5°35′35.0″S	105°11′16.0″E
G	Ratai Bay Coast	5°36′00.0″S	105°10′50.0″E
H	Ratai Bay Coast	5°36′30.0″S	105°10′15.0″E

equipment (Variant 715-ES) made in Rocklin, California, USA was used to determine the heavy metal content of the samples. The materials used for preparing plankton samples were 4% formalin, ice cubes, bi-distilled water, and 65% HNO_3 from EMD Millipore Corp., Germany.

Research procedure

Research design

The method used in the research was a purposive sampling method based on the consideration of the distance to the location of ASGM activities and waste input as well as the ease of sampling. Water and plankton sampling techniques were carried out randomly at two points at each station with a distance of 500 m from the first point (station A).

Sample collection and preparation

The water and plankton samplings were carried out by determining *the coordinates of the sampling location using GPS*. Sampling was carried out at a depth of 50 cm from the water surface at all points by using bottle samplers. Sampling on the river water surface was carried out in the morning, while seawater samples were taken in the afternoon. The plankton samples were collected using a plankton net (with 20-μm mesh size) by filtering 100 L of surface water, and the filtered sample was then put into a 600-mL sample bottle followed by the addition of three drops of 4% formalin and stored in a cool box. Meanwhile, river and seawater samples were taken using a 600-mL bottle and then stored in a cool box.

Identification of plankton was carried out by taking plankton samples using a dropper pipette, and dropping them on an object glass, and then observing them using a binocular microscope. Plankton have been identified based on morphological characteristics in accordance with the plankton identification book (Davis 1955).

25 mL of plankton samples (taken from the previous 600-mL sample plankton) were destroyed by adding three drops of 75% HNO_3 and then filtered using a filter paper (Whatman No. 41). Filtered samples were then put into a film bottle to further analyze their heavy metal content.

Meanwhile, 10 mL of water sample (after being filtered through a filter paper Whatman No. 41) was put into a digestion flask using a 10-mL pipette. Then, 0.2 mL of HNO_3 and 0.1 mL of HCl were added and homogenized. The mixture was filtered again if precipitate forms. Then the sample was heated using a hot plate at 95 °C for 30 min. The sample was then cooled and transferred into a 10-mL volumetric flask using a 10 mL pipette. The sample was added with distilled water until the sample volume reached 10 mL and was transferred into a high-density polyethylene (HDPE) bottle. Samples were ready to be analyzed using the ICP-OES.

Heavy metal analysis using ICP-OES

The prepared samples were analyzed using ICP-OES (following the method described by Ghosh *et al.* 2013) to determine the concentration of lead (Pb), cadmium (Cd), copper (Cu), chromium (Cr), nickel (Ni), zinc (Zn), silver (Ag), cobalt (Co), iron (Fe), and manganese (Mn) with LOD (0.004 mg/L for Ag and Co; 0.003 mg/L for Cd and Zn; 0.005 mg/L for Cu; 0.009 mg/L for Cr; 0.037 mg/L for Pb) and LOQ (0.013 mg/L for Ag and Co; 0.008 mg/L for Cd; 0.030 mg/L for Cr; 0,032 mg/L for Cu; 0.072 mg/L for Mn; 0.010 mg/L for Zn). LOD and LOQ are presented in Table 5.

Data analysis

The following heavy metal parameter was observed, namely Ag, Zn, Pb, Cd, Ni, Cr, Cu, Co, Fe, and Mn. The heavy metal quality standard set by the Ministerial Regulation of Indonesian Ministry of Environment and Forestry year 2014 (PERMEN LH No.5/2014) concerning the wastewater quality standard, Australian and New Zealand Environment and Conservation Council (ANZECC), Agriculture and Resource Management Council of Australia and New Zealand (ARMCANZ) 2000, Government Regulations of Indonesia number 22 year 2021 (PP RI No. 22/2021), Environmental Protection Agency (EPA) of 1987 was used as the quality standard.

Calculation of bioaccumulation of heavy metals in plankton

The study of the ability of plankton to accumulate heavy metals in the waters of the Way Ratai River was analyzed using a BCF. The BCF was calculated using a formula (Potipat *et al.* 2015) as follows:

$$\mathrm{BCF} = \frac{C_{\,biota}}{C_{\,ambient\ media}}$$

where c_{biota} refers to the heavy metal concentration in plankton (mg/L); $c_{ambient\ media}$ refers to the concentration of heavy metals in water (mg/L).

Statistical analysis of heavy metal concentration in different locations

Statistical analysis was conducted in order to determine the difference in heavy metal concentrations in plankton and water among seven locations of the study site using ANOVA followed by Tukey at a 5% level of difference (SAS/STAT).

RESULTS AND DISCUSSION

Diversity of plankton in the water of Ratai Bay

Based on the study results conducted in the waters of the Way Ratai River and the coast of Ratai Bay (Tables 1 and 2), 14 classes were identified. Also, 77 types of plankton were detected, of which 37 types belong to phytoplankton and four belong to freshwater zooplankton. Around 36 types of seawater phytoplankton were found in the samples collected from the Way Ratai Coast and estuary.

Tables 2 and 3 show the number of plankton species identified from water samples of Way Ratai River and estuary Ratai Bay Coast. In this study, the total number of classes of plankton (phyto and zooplankton) found in freshwater was nine classes while those in estuary-coast were six classes.

Plankton diversity in the Way Ratai River Stream

The sampling of plankton was carried out at different times, and plankton collection in the river was carried out in the morning. Plankton at station A was found to be the least compared to other stations. There were five classes including Bacillariophyceae, Chlorophyceae, Euglenophyceae, Rhizopoda, and Litostomatea with one species each. The last two (Rhizopoda and Litostomatea) belong to zooplankton. The low diversity of plankton species at station A seems to be influenced by

Table 2 | Plankton types in the Way Ratai River (freshwater)

Station	Name of location	Class of plankton
A	Bunut Seberang Village (Way Ratai river	Bacillariophyceae (1 species), Chlorophyceae (1 species), Euglenophyceae (1 species), Rhizopods* (1 species)
B	Kepong Jaya Village	Litostomata* (1 species). Bacillariophyceae (7 species), Chlorophyceae (4 species), Cyanophyceae ([illegible] species), Euglenophyceae (2 species)
C	Way Urang Village	Rhizopods* (1 species). Bacillariophyceae (5 species), Chlorophyceae (2 species), Cyanophyceae (1 species), Euglenophyceae (1 species)
D	Way Ratai Stream in Sanggi Village, Pematang Awi	Ulvophyceae (1 species), Zygnematophyceae (1 species). Bacillariophyceae ([illegible] species), Chlorophyceae (5 species), Cyanophyceae (1 species), Euglenophyceae (2 species), Dinophyceae (1 species), Lobosea* (1 species), and Rhizopods* (1 species)

Note: * indicates zooplanktons.

Table 3 | Plankton types in Way Ratai Estuary and Ratai Bay Coast

Station	Name of location	Class of plankton
E	Way Ratai Estuary	Bacillariophyceae (1 species), Dinophyceae (3 species), Raphidophyceae (1 species), Prymnesiophyceae (2 types_
F	Ratai Bay Coast	Dinophyceae (12 species), Prymnesiophyceae (1 species), Dictyochophyceae (2 species)
G	Ratai Bay Coast	Bacillariophyceae (2 species), Dinophyceae (3 species), Mediophyceae (2 species)
H	Ratai Bay Coast	Bacillariophyceae (3 species) Dinophyceae (4 species), Mediophyceae (1 species)

many factors, especially the water factor. The watershed at station A has several gold processing points (for ASGM activities) as well as a dumping ground for tailings, where the waste from the mining process causes the river to become cloudy. High turbidity inhibits the penetration of sunlight required for the photosynthesis process of phytoplankton. A good environment is a very important factor for phytoplankton density and for the level of water fertility. The fertile waters will have a good impact on the environment; therefore it can support the life of many different organisms through food webs. Meanwhile, at stations B, C, and D which are far from the tailing places, 34 types of phytoplanktons and 2 types of zooplanktons are found. From the number of plankton species counted in this study, the farther the location away from the ASGM process the more the number of plankton species found. For instance, 20 plankton species are found in station D (the farthest from station A), while in station A only five plankton species are found.

In general, based on samples obtained from the waters of the Way Ratai River, phytoplanktons were mostly found compared to the zooplankton group. This was also indicated in the River Ravi in Pakistan, where plankton taxa disappeared in highly polluted sampling areas (Rauf *et al.* 2019). This study (Rauf *et al.* 2019) also indicated that the number of phytoplankton found was greatly in abundance compared to the zooplankton. The various types of existing phytoplanktons indicated their role as primary producers in the waters. Biologically, a much smaller number of zooplankton species were found in the waters compared to the number of phytoplankton and this might also be due to the difference in their life cycles. The phytoplankton life cycle takes place in a faster time than zooplankton, where the division of phytoplankton cells is faster so that within a few days it will reach a very high density. On the other hand, zooplankton in its development takes more time, because generally to reach adult conditions, zooplankton must go through several stages of life such as eggs, larvae, and juveniles.

Based on the identification results, it can be seen that the most abundant phytoplankton group at each station belongs to the Bacillariophyceae class. The abundance of Bacillariophyceae (namely diatoms) in waters is presumably due to their ability to adapt to the environment, they are cosmopolitan, found in fresh, brackish, and marine waters worldwide, and also in damp terrestrial habitats (Mann *et al.* 2017). They have survived in various extreme water conditions and have high reproductive power compared to plankton from other classes (Odum 1998). In addition, they are the most species-rich group of autotrophic algae. The phytoplankton of the Bacillariophyceae class has a very fast response to the addition of nutrients and is able to adapt to the environment (Nybakken & Bertness 2005). This makes the Bacillariophyceae class the most commonly found in all sampling locations.

Plankton diversity in Ratai Bay Coastal waters

Based on the identification results (Table 3), it can be seen that the most abundant phytoplankton groups at each station in the coastal waters of Ratai Bay are Dinophyceae and Bacillariophyceae classes. By using a plankton net with a size of 20 μm, three main groups, namely diatoms (Bacillariophyceae), dinoflagellates (Dinophyceae), and blue algae (Romimohtarto & Juwana 2009) were mostly found. Dinophyceae (dinoflagellates) based on the identification results were found in all coastal stations (F–H). However, at station E (river estuary), Bacillariophyceae and Dinophyceae classes were found with fewer species compared to other stations, indicating that the estuary area has fewer species compared to sea waters. The number of species in the downstream area or brackish water is generally much less than that of the plankton inhabiting nearby freshwater or marine habitats (Dianthani 2003). The types of phytoplankton that belong to the Mediophyceae class exist only at stations G and H, which are marine waters, while in stations E and F, this type of Meddiophyceae was not found. This presumably is because phytoplanktons of the Mediophyceae class are rarely found in freshwater or stagnant water; this type is usually found in environments that have electrolyte-rich waters (Israwati *et al.* 2018).

Heavy metals in plankton

The following are the results of heavy metal analysis in plankton using ICP-OES.

Based on Table 4, the results showed that there is some detection of heavy metals in the planktons either collected from freshwater or brine water. However, only chromium was undetected in the plankton of this study. No standard quality of the heavy metal concentrations in plankton has been made so far; therefore, this data could be used as a baseline study to determine the degree of heavy metal pollution of the study site due to the ASGM activity by using plankton as the determining factor for heavy metals transport in the water. This study could elaborate on the study by Dobaradaran *et al.* (2018) who investigated the fate of metal pollutants in aquatic food webs through zooplankton at a lower tropic level. The higher metal concentration in plankton could be seen in different sites of study. For instance, the concentration of Ag was only detected on E, F, G, and H sites (coastal water), from which the further the site location from the ASGM site, the higher concentration of Ag in the plankton ($p < 0.05$). Fe on A, B, and C sites (river sites), G, and H sites (coastal sites); Mn only on the H site (coastal site), yet among those study sites, the higher concentration of Fe was found in the H site ($p < 0.05$). This phenomenon also was shown for Ni and Pb. Ni on E, F, and H sites (coastal sites), from which the highest concentration was also found in H site ($p < 0.05$). Pb on C site and mostly on F, G, H sites (coastal sites) from which the H site also showed the highest concentration, even though the $p > 0{,}05$. Zn on C site and E and G sites (coastal sites), yet the concentration did not show any statistical difference among the study sites ($p > 0.05$). This difference in heavy metal concentration found in plankton could be caused by several factors, one of which was due to the high metal accumulation process by plankton from the polluted water by ASGM activities (in the A site). Other than that, the higher accumulation of some heavy metals in plankton in coastal areas could also be related to high erosion because of high surface runoff and steep slopes along the same study sites (Yuwono *et al.* 2023). Based on the consideration that plankton is a single-celled organism with a large surface area compared to its volume ratio, therefore, it has a high accumulation ability of organic and inorganic substances in a relatively short time (Kullenberg 1987).

However, it is possible that there are many other metal contributors apart from tailings (ASGM) activities, for example, community activities around the riverbank and waters. Based on the survey that has been carried out, the Way Ratai River is used by the community in daily activities such as washing, and bathing, and in several locations there were piles of garbage found around the banks of the Way Ratai River. These piles of garbage were seen in stations C and D, which could trigger heavy metal pollution from the river to the Ratai Bay area, especially for Fe, Pb, and Zn. While in sea waters, the concentration of Cd metal at all coastal sites (E, F, G, H sites) has exceeded the quality standard limit but not for Cu. The possibility of exceeding the Cd concentration in plankton could be accounted from coastal activity, such as

Table 4 | Concentration of heavy metals in planktons

Sample station	Metal (X/SD mg/L)									
	Ag	Cd	Co	Cr	Cu	Fe	Mn	Ni	Pb	Zn
A	nd	nd	0.0073 0.0052	nd	0.0170 0.0113	0.3180 0.2290[ab]	0.0520 0.0395[a]	nd	nd	0.0650 0.0353
B	nd	nd	0.0058 0.0031	nd	0.0070 0.0014	0.5480 0.2503[ab]	0.1050 0.0381[ab]	nd	nd	0.0295 0.0034
C	nd	0.0050 0.0014	0.0053 0.0024	nd	nd	0.5655 0.0714[ab]	0.0680 0.0438[ab]	0.0157 0.0130[bc]	0.0355 0.0007	0.0280 0.0226
D	nd	0.0053 0.0024	nd	nd	nd	0.2910 0.0299[a]	0.0865 0.0247[ab]	0.0175 0.0021[bc]	nd	0.0360 0.0183
E	0.0070 0.0000[a]	0.0075 0.0007	0.0200 0.0098	nd	nd	0.0900 0.0961[a]	0.0230 0.0240[b]	0.0640 0.0226[ab]	nd	0.0404 0.0531
F	0.0110 0.0014[b]	0.0055 0.0007	0.0193 0.0020	nd	nd	0.1955 0.0007[a]	0.0370 0.0028[b]	0.0795 0.0275[b]	0.0580 0.0311	0.0259 0.0326
G	0.0045 0.0007[c]	0.0090 0.0042	0.0103 0.0094	nd	0.0080 0.0024	0.7260 0.2559[ab]	0.0705 0.0205[ab]	0.0345 0.0049[abc]	0.1105 0.0615	0.0535 0.0148
H	0.0050 0.0000[bc]	0.0060 0.0028	0.0180 0.0070	nd	nd	1,2480 0.0650[b]	0.1786 0.0162[b]	0.0815 0.0063[b]	0.0885 0.0728	0.0400 0.0162

Notation with different alphabetical order in the same column indicated significant different at $p < 0.05$. nd, not detected; A–D indicates river stations. E–H indicates coastal stations.

industrial and municipal effluents, landfill leaching, non-profit sources runoff, and atmospheric deposition, just like those found in different places (Azizi *et al.* 2018; Haeruddin *et al.* 2021).

The dangers posed by heavy metals also vary. Pb metal can cause acute and chronic poisoning. The toxicity of this metal is due to the disruption of hemoglobin formation due to the inhibition of the enzyme activity process by Pb^{2+} ions which are strongly bound to each other with sulfur groups in amino acids (Fardiaz 1992). In contrast to Zn metal which is an important component in enzymatic processes that can increase enzyme activity, Zn will also be dangerous and cause poisoning if in excessive concentrations (Mills 1989). Cd can cause toxicity in the human body if it accumulates over a long period of time (Martin & Griswold 2009). Ni is acidic and corrosive to the skin, exposure to nickel can cause skin itching, sores, and irritation (Djuanda 2007).

Mn content above the threshold could trigger many ailments (Aguilera *et al.* 2022), such as nerve damage, psychiatric disorders, symptoms of brain disorders, and abnormal behavior. One of the various minerals in the water is Fe; if the Fe content is present in large quantities it will cause environmental pollution and is toxic to humans (Martin & Griswold 2009). In testing the levels of heavy metals using ICP-OES there are several points that are not detected (nd). It is because the value of the metal is below the detection limit of the ICP-OES tool; therefore it cannot be detected by the tool.

Heavy metals in water

The value of heavy metal material per station in samples of river water and seawater in Way Ratai Waters, Pesawaran Regency can be seen in Table 5.

Table 5 | Concentration of heavy metals in water from each station

	Metal ($X \pm SD$) mg/L								
Sample station	**As**	**Cd**	**Co**	**Cr**	**Cu**	**Fe**	**Mn**	**Pb**	**Zn**
A	0.1030 0.0700	0.0090 0.0035	0.0690 0.0459	0.0130 0.0028	0.0560 0.0084	0.2440 0.0113[a]	0.0140 0.0049	nd	0.0370 0.0395
B	nd	0.0160 0.0028	0.0380 0.0311	0.0160 0.0014	0.0445 0.0176	0.3720 0.1060[a]	0.0215 0.0120	nd	0.0355 0.0021
C	nd	0.0205 0.0120	0.1450. 0.0120	0.0110 0.0021	0.0360 0.0070	0.4190 0.0749[a]	0.0170 0.0141	0.0290 0.0410	0.0460 0.0226
D	nd	0.0110 0.0028	0.0505 0.0120	0.0105 0.0021	0.0385 0.0035	0.6175 0.4023[a]	0.0595 0.0747	nd	0.1015 0.0700
E	nd	0.0010 0.0049	0.0780 0.0438	nd	0.0445 0.0063	0.2445 0.1138[ab]	0.0290 0.0028	nd	0.0430 0.0353
F	0.0350 0.0219	0.0100 0.0035	0.0370 0.0035	0.0150 0.0042	0.0430 0.0084	0.0295 0.0233[b]	nd	0.0400 0.0565	0.0180 0.0091
G	0.0380 0.0056	0.010 0.0035	0.0315 0.0106	0.0100 0.0007	0.0040 0.0070	0.0560 0.0197[b]	nd	0.0410 0.0515	0.0570 0.0056
H	0.0385 0.0106	0.0075 0.0021	0.0505 0.0205	nd	0.0515 0.0106	nd	nd	0.0515 0.0728	0.0585 0.0275
River water quality Standards #	0.003[4]	0.01[3]	0.20[3]	0.05[3]	0.02[3]	0.30[3]	0.10[3]	0.03[3]	0.05[3]
Seawater quality Standards#	0.50[1]	0.001[3]	0.40[1]	0.005[3]	0.008[3]	0.50[2]	0.10[2]	0.05[3]	0.008[3]
LoD	0.004	0.002	0.004	0.009	0.005	0.011	0.022	0.037	0.003
LoQ	0.013	0.008	0.013	0.030	0.032	0.459	0.072	0.123	0.010
% Recovery	83.7	104.5	92.7	106.7	102.2	91.5	105.7	88.74	105.80

Quality standard #: **1.** PERMEN LH No. 5 of 2014, **2.** EPA of 1987, **3.** PP RI No. 22 of 2021, **4.** ANZECC/ARMCANZ 2000. LoD/LoQ refers to the Limit of Detection and Limit of Quantification. Notation with different alphabetical order in the same column indicated significant different at $p < 0.05$. nd, not detected; A–D indicate river stations; E–H indicate coastal stations.

Analysis of heavy metal was also carried out on river water and seawater samples. The level of Ag at station A exceeded the quality standard based on ANZECC/ARMCANZ 2000 with a detection limit of 0.003 mg/L, unlike in stations B, C, and D, where Ag was not detected by ICP-OES. Yet, the concentration of Ag in water from different locations did not show any difference ($p > 0.05$), except for Fe. But Cd and Fe as well as Cu content at these three stations exceeded the predetermined river water quality standards that have been set by Indonesian Government (Government Regulation of the Republic of Indonesia Number 22 of 2021). However, only Fe indicated a significant difference among other locations (from the river to the coastal water, $p < 0.05$), the further the location from the ASGM site, the lower the concentration of Fe in the water, from station A to station H. This regulation covers the need for river water which is used for fish farming, freshwater, water husbandry to irrigate crops, and/or for other purposes that require the same water. While Mn content only exceeds quality standard class 1 at station D, where the water is used for drinking water quality standards or other designations that require the same water quality. Yet, we assumed that the Mn was released from the municipal activity of the D site, namely Sanggi Village – Pematang Awi.

At the coastal station (stations of E, F, G and H), we found that the metal content of Cd and Pb at stations F–H, Cr at stations F and G, Cu at stations E, F, H, and Zn at station G and H exceeded the heavy metal quality standards for seawater, yet none of these concentrations showed any significant difference ($p > 0.05$). While metals Ag, Co, Fe, and Mn at coastal stations were below the quality standard values set by Government Regulation of the Republic of Indonesia No. 22 of 2021, EPA of 1987, and Indonesian Ministerial Regulation (the Minister of the Environment & Forestry Number 5, of 2014). Cd and Pb content of the coastal water might be due to water transportation, since the Ratai Bay is also one of the busy bays for public water transportation from the inland of Sumatra to inhabitant small islands surrounding the bay.

Water quality decreases due to the presence of pollutants, in the form of organic and inorganic components, in which inorganic components, including heavy metals, are dangerous (Siaka 2008). Heavy metals are classified as pollutants because they are merely harmful when their concentrations are high, and they cannot be decomposed through biodegradation such as those organic pollutants. Heavy metals can be accumulated in the environment, especially in rivers and seas; they can be bound to organic and inorganic compounds through adsorption processes and the formation of complex compounds (Susiati *et al.* 2009). When compared between the heavy metal material in the plankton sample and the water sample, the heavy metal material in the plankton sample is much greater than those in the river and seawater samples, which is presumably caused by the accumulation of heavy metals in the water which experiences dilution due to tides, adsorption and absorption of both aquatic and sedimentary organisms. Other studies also indicated that the biomass of phytoplankton had a negative correlation with the heavy metal concentration in the water of Rongna Tiver, China (Luo *et al.* 2021).

However, there are some cases where the heavy metal material in the water is more than those in the plankton. As those seen in station D (river water), when compared to the heavy metal material in plankton, almost all heavy metals at station D (Ag, Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb) of the river water were greater than those inside plankton. This is due to an increase in the concentration of these metals at the location, resulting in a bioaccumulation process until it reaches a saturation point for plankton cells. It is indicated by the occurrence of excess accumulation of metal activity so that the plankton body is no longer able to process metabolism, such as those seen in phytoplankton affected by Cd (Permana & Akbarsyah 2021). Exposure to high Cd concentration in the water could lead to various impacts on the phytoplankton, including disruption of the cell, inhibition of biosynthesis, degradation of chlorophyll up to inhibition of cell division.

Based on Table 5, heavy metals have exceeded quality standards in both rivers and the sea caused by ASGM activities on land that dump tailings into rivers and the sea. At stations A, B, C, and E, the Pb metal material was not detected, because the Pb metal material at these stations had a small value and was still below the ICP-OES detection limit. The content of heavy metal Pb in the waters is relatively low because most of the metal ions are adsorbed and absorbed by the highly suspended solids (Arifin 2011).

BCF analysis

The values of the BCF for every heavy metal at each station are presented in Tables 6 and 7.

The higher the value of the BCF in an organism is, the higher the accumulation of heavy metals in that organism. Based on the category of BCF values, pollutant properties were classified into three sequence categories, namely the very accumulative category if the BCF value is greater than 1,000 (BCF > 1,000), moderate accumulation category if the value is between 100 and 1,000 (BCF 100–1,000), and low accumulation category if the value is less than 100 (BCF < 100) (Van Esch 1977). All BCF values of the plankton collected from the river and sea waters indicated that they lie in a low value category, predicting that the contamination level of such pollutants in plankton was low. It could be accepted, since the plankton mostly have very

Table 6 | Value of metal BCF in planktons

Station	BCF value Ag	Cd	Co	Cr	Cu	Fe	Mn	Ni	Pb	Zn
A	nd	nd	0.15	nd	0.33	1.30	3.70	nd	nd	1.75
B	nd	nd	0.21	nd	0.15	1.47	5	nd	nd	0.85
C	nd	0.23	0.05	nd	nd	1.47	2.51	nd	nd	0.60
D	nd	0.63	nd	nd	nd	0.47	0.77	nd	nd	0.35
E	nd	7.50	0.26	nd	nd	0.37	0.79	nd	nd	1.81
F	0.31	0.55	0.95	nd	nd	6.76	nd	nd	1	2.72
G	0.13	0.90	0.53	nd	2	12.96	nd	nd	1.90	0.95
H	0.13	0.75	0.35	nd	nd	nd	nd	nd	1.36	0.52
Average	0.19	1.76	0.35	nd	0.82	3.54	2.55	nd	1.42	1.19

nd, not detected.

Table 7 | BCF categories for heavy metals (Van Esch 1977)

BCF range	BCF category
>1,000	Very high
100–1,000	High
30–100	Moderate
<30	Low

short life cycles, unless other aquatic organisms which consumed those planktons at higher food trophic levels. For instance, at one of the food tropic levels and during their life cycle, the concentration of heavy metals could be accumulated in bivalves, even in their shells such as those seen in *Trachycardium lacunosum* (Karbasdehi *et al.* 2016; Dehghani *et al.* 2019). Besides that, the degree of bioconcentration of heavy metals in plankton depends on species and seasonality (Roy *et al.* 2010) and the complexity of the water ecosystem (Burada *et al.* 2014). Therefore, a time series study should be conducted for determining more precisely the BCF values of heavy metals found in the plankton samples in the study site.

The order of BCF values in the plankton of the Way Ratai was obtained based on the calculation results (Table 6) as follows: $BFC_{Fe} > Mn > Cd > Pb > Zn > Cu > Co > Ag$ metals. However, Cr and Ni metals could not be calculated for BCF, since Cr was not detected in plankton and Ni was also not detected in water samples. The highest metal concentration was found for Fe. Iron or ferrum (Fe) is one of the most common metals found in the earth's crust and is one of the minerals that is often found in large amounts of water. Fe metal is also one of the essential metals that are present in the plankton body, and is needed for plankton growth; therefore, the Fe content in plankton is greater than other metals. Yet, excessive Fe content can cause poisoning and disturb the environment (Martin & Griswold 2009).

The value of the BCF at several points was undetected because the concentration of metals in the water or plankton was below the limit of tool detection. This is likely because most of the metals at mining sites are only slightly dissolved in the water and some are in the form of deposits or particles. Other studies indicated that the low levels of heavy metals in the water did not mean that it did not have a negative impact on the waters, but rather the ability of these waters to dilute contaminants was quite high (Rochyatun *et al.* 2006). Besides, the waters in the area of study tend to change due to the influence of currents and tides which easily dissolved the metals and distributed them to other areas/locations. However, more study is needed to elaborate, such as determining the number of planktons as well as their heavy metal concentration in time series with seasonal changes. Some limitations might have occurred during collection of planktons for determining their heavy metal concentration; therefore, a more detailed study should be done along with differentiation between phytoplankton and zooplankton for their heavy metal content.

CONCLUSION

Based on the research that has been carried out, it is concluded that most of the heavy metal contents are found in plankton, with concentrations as Fe > Mn > Zn > Ni > Co > Cd > Ag > Pb, and only Cr could not be found in the plankton of the study site but exist in the water. Meanwhile, the heavy metal content of Ag, Cd, Cu, and Fe has exceeded the quality standards for freshwater. For those in coastal water, concentrations of Cd, Cr, Cu, Pb, and Zn have also exceeded the quality standard for saltwater. These indicate that the waters of Way Ratai have been polluted, which could be from many different resources, municipal or other activities. However, the BFC of every heavy metal was considered to be low since the average of BFC was lower than 30, with the consecutive BFC of each heavy metal being BFC_{Fe} > Mn > Cd > Pb > Zn > Cu > Co > Ag. This study could also be used as baseline data of the study site for determining the heavy metal concentration in the plankton of freshwater and brine water due to ASGM activity.

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DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

CONFLICT OF INTEREST

The authors declare there is no conflict.

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