Research Article

Bioaccumulation of zinc by *Portunus pelagicus*: Nuclear application techniques that use radiotracer ⁶⁵Zn to study influence of concentration of Zn in seawater

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Article information	Abstract
	Zinc (Zn) is an essential trace metal, but also a potential toxicant to aquatic organisms. Zinc is one of the toxicants from various industrial and domestic activities whose waste is directly discharged into water. One of the marine organisms used as an indicator of pollution is the blue swimming crab (*Portunus pelagicus*). Bioaccumulation studies of Zn in *Portunus pelagicus* from Jakarta Bay were conducted in a radiotracer laboratory. Laboratory experiments were done to study the uptake and release of ⁶⁵Zn radioisotope in *Portunus pelagicus*. The experiments conducted were biota collection, acclimatization, bioaccumulation, and elimination. This study was to determine the relationship between the amount of ⁶⁵Zn released into the aquatic environment and its effect on Zn bioaccumulation ability and radiation dose received by *Portunus pelagicus*. Biokinetics parameters, such as concentration factors CF, uptake rate constant k_u, elimination rate constant k_e, and concentration factor steady state (CF_{ss}) were investigated. The results showed that an increase in concentration would decrease the rate of absorption and rate of elimination, and that the concentration factor CF values varied from 7.82 - 60.62 ml.g⁻¹ in *Portunus pelagicus*. The concentration factor at steady-state Zn was 25.55-59.47 ml.g⁻¹, which is influenced by the concentration after 6 days of exposure. The depuration rate was observed to be high, with a value of retention 34.33 - 78.88 % of ⁶⁵Zn absorbed by *Portunus pelagicus*, which was absent 7 days after exposure.

1. Introduction

Jakarta Bay is located in the waters of the Java Sea to the north of Jakarta province, where 13 rivers flow through the city of Jakarta and into Jakarta Bay. The waters off Jakarta's northern coast are a highly dynamic and densely populated area and, with anthropogenic influences, the semi-

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enclosed Jakarta Bay has received a heavy load of domestic, organic, industrial, and heavy metals pollution and oil spills, which tend to increase every year. The waters of Jakarta Bay and the Thousand Islands have been polluted (Kunzmann et al., 2018).

One of the contaminants is heavy metal. Heavy metals have long been used as basic materials and catalysts for various types of industries. Waste that enters marine waters reduces water quality and causes pollution. Heavy metals that enter the waters experience dilution and are distributed by turbulence and ocean currents (Zuraida et al., 2018). Environmental pollution from heavy metal contamination is a serious problem and is receiving great attention because of the adverse effects it causes throughout the world (Briffa et al., 2020).

Zn is a heavy metal that living things need in small amounts, and is dangerous if the amount is excessive, including in the environment. Zinc can cause pathological effects in both excess and deficiency. Toxic levels of zinc have been shown to cause lethargy, neurotoxicity, and gliotoxicity. High-level zinc causes neuronal death in cortical cell tissue cultures (Bartzatt, 2017). The biggest source of contamination is Zn dissolved in water from human activities. Zinc metal is mainly used in the steel product galvanizing industry because of its anti-corrosion properties in the atmosphere and can be mixed with various other metals with different properties. Zinc is used in various industries, namely galvanizing, coating, die-cast alloys, extruded products, dry batteries, oxides, and chemicals (Sabnavis et al., 2018).

Due to human activities, Jakarta Bay has a high input of chemical, biological, and radioactive pollutants from land and sea (Prihatiningsih et al., 2016). Data from the Indonesian Central Bureau of Statistics in 2022 states that, in the coastal land area of Jakarta Bay, there are approximately 1,683 industries, consisting of large, medium, and small industries. Most of these industries, especially small industries, do not have proper waste treatment plants. This has resulted in a decrease in river water quality and has ultimately polluted the aquatic ecosystem of Jakarta Bay.

There has been significant water pollution in Jakarta Bay, resulting in many heavy metals and other contaminants entering the bay's waters, sediments, and fisheries. Pollution in Jakarta Bay causes the degradation of fish resources, environment, and habitat (Nugraha et al., 2020). Degradation of fish resources appears in the form of low fish abundance and biomass, smaller fish catches, and tissue damage in green mussels, blue swimming crabs, and fishes. Environmental degradation appears in the form of the appearance of algae blooms from the diatom group, low macro zoobenthos abundance index, decreased water quality, and habitat degradation in the form of shrinking mangrove areas due to conversion to cultivation areas and agricultural land. Heavy metals are dissolved in water, distributed into sea waters, and precipitated in sediments, and enter the food chain so that they accumulate in the organs of living things (Riani et al., 2014). Heavy metal contamination in fish is influenced by the age of the fish, its tissue fat content, and how it is eaten (Asante et al., 2014).

Jakarta Bay is one of the biggest fishing grounds for Blue Swimming Crabs (*Portunus pelagicus*) in the Java Sea with its small-scale fishery (Jayawiguna et al., 2017). Based on a report from the Ministry of Maritime Affairs and Fisheries in 2021, the catch data for *Portunus pelagicus* from Jakarta Bay was 206,037 kg. *Portunus pelagicus* is an important species in Indonesia that is caught nearshore on the seafloor. *Portunus pelagicus*, a portunidae family member, is a commercially important portunid crab found on Java's north coast, including Jakarta Bay. These crabs are regarded as a delectable seafood item that is a significant commodity in the international seafood market. Various kinds of heavy metal contaminants can accumulate in *Portunus pelagicus*. Aquatic organisms such as *Portunus pelagicus* absorb metals through their gills, which enter the bloodstream. Transfer of metals then enters the apical membrane and, through the cell, interacts with intracellular ligands by crossing the basement membrane. Metal bioaccumulation is characterized by direct proportionality of the accumulated concentration and exposure time, while the concentration of metals in the body of organisms is determined by the intracellular metal-binding ligands and the exchange of complexes formed (Azizi et al., 2018).

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Research on heavy metal bioaccumulation has been widely carried out across the world, especially since the outbreak of mercury pollution in Minamata, Japan. The research carried out has been environmental monitoring for a certain period of time and has not been continuous. A sampling of abiotic components (water and sediment) and biotic components (phytoplankton, zooplankton, invertebrates, and various types of fish) has been analyzed for heavy metal content. This method obtained various bioaccumulation factor values and did not take into account exposure to feed and particulates (Riani et al., 2017).

Research publications related to the bioaccumulation of Zn metal are urgently needed by the public in general and by environmental NGOs to add knowledge in understanding the nature of pollutants, and the mechanism of pollutants from pollutant sources to enter the environment and organisms, which then enter the human body. The development of heavy metal bioaccumulation methods is currently carried out in the laboratory by modeling the conditions of aquatic ecosystems. Radioactive tracers are used as labeling of pollutant simulations to make it easier to detect, and research is carried out continuously. Several studies of heavy metal bioaccumulation in various biota using radiotracer techniques have been carried out (Kuranchie-Mensah et al., 2018; McDonald et al., 2020; Pouil et al., 2018; Prihatiningsih et al., 2016).

Studies of heavy metal bioaccumulation are generally carried out in situ, including Zn bioaccumulation in biota, water, and sediments. However, this study does not represent a steady state. Zn bioaccumulation studies by a nuclear technique using the isotope ^{65}Zn (T1/2= 244.26 days and 0.39 % uncertainty value) have been carried out in pomfret (*Colossoma macropomun*); sepat fish (*Trichogaster trichopterus*) (Al Mustawa et al., 2022); horse mussels (*Modiolus micropterus*) (Budiawan et al., 2021); rock clams (*Saccosrea glomerata*) (Lee et al., 2014); freshwater shrimp (*Paratya australiensis*) (McDonald et al., 2020), etc. However, research on Zn bioaccumulation in *Portunus pelagicus* from Jakarta Bay by a nuclear technique using the ^{65}Zn isotope has never been carried out.

Based on the above description, a bioaccumulation study was conducted by using a ^{65}Zn radiotracer to observe the accumulation and elimination of zinc metal in *Portunus pelagicus*, an important fish economically and which is widely consumed by the people of Indonesia. In addition, the opportunity was provided to investigate a subject that has not been widely studied in the field radionuclide bioaccumulation on other marine biota, the relationship between release, accumulation ability, and dose received by organisms.

2. Methods

Zinc bioaccumulation follows the same principle as pollutant entry into living organisms. The method used in this study is based on the absorption and elimination of contaminants using a biokinetic model. The research method for Zn bioaccumulation in *Portunus pelagicus* with the isotope ^{65}Zn refers to Budiawan (Budiawan et al., 2021), with several modifications in the laboratory.

2.1 Biota

In October 2022, *Portunus pelagicus* specimens weighing 106.7 - 124.2 g were collected from Tanjung Kait in Jakarta Bay. Samples of *Portunus pelagicus* were placed in plastic containers filled with seawater and added oxygen, then brought to the laboratory of the Research Center for Radioisotope Technology, Radiopharmaceuticals and Biodosimetry (BRIN), Serpong, South Tangerang.

2.2 Acclimatization

Acclimatization was carried out to provide adaptation time for the research object in the form of living organisms in the research environment so that the biota was not stressed and could be used for experiments. *Portunus pelagicus* were acclimated to laboratory conditions (open circuit, 200 L

aquarium, seawater renewal: 30 % h^{-1}; T: 27±0.5 °C; salinity: 35 ‰; pH: 8.0±0.1) for 7 days before the experiment. *Portunus pelagicus* was fed chopped shrimp meat during this time.

2.3 Radioactive tracer

The single-compartment model was used to determine the kinetics ^{65}Zn contaminant uptake and depuration. Natural Zn from the Merck brand was used to create radioisotope ^{65}Zn at the Research Center for Radioisotope Technology, Radiopharmaceuticals, and Biodosimetry-BRIN. The activity of the radioisotope concentration was counted by a carrier with the ORTEC Solid-State Photon Detector HPGE (High-Purity Germanium) spectrometry system, which was linked to a multichannel analyzer and a computer running the *Maestro* software. The radioactivity concentration in the biota was determined by comparing the concentration of the phantom in ^{65}Zn, namely 1584 Bq. Radioactivity was determined by comparing it to a known standard (1584 Bq activity ^{65}Zn) and the phantom geometry of *Portunus pelagicus*. To calculate the efficiency and physical radioactive decay, the measurements were corrected. Using counting time, the counting error was determined to be less than 5 %.

2.4 Seawater exposure pathway

Following the completion of the acclimatization process, 4 aquariums with a capacity of 10 L were prepared, each filled with 8 L of seawater. Furthermore, each aquarium contained 3 *Portunus pelagicus* of varying sizes. Three *Portunus pelagicus* weighing 106.7 - 124.2±0.1 g were placed in a 10 L aquarium with 0.5 ppm ZnCl$_2$ and 2,846×10^{-15} ppm ^{65}Zn. Every day, the ^{65}Zn activity in *Portunus pelagicus* and 1 ml of seawater was measured using the HPGE ORTEC Solid-State Photon Detector Spectrometry. The concentration factor (*CF*) is the ratio of the pollutant concentration in the biota body to the concentration in the water. As shown in the following Eq. (1):

$$CFt = \frac{Ct}{Cw} \tag{1}$$

CFt is the activity concentration factor in a certain time (t), and *Ct* is the concentration of the ^{65}Zn radiotracer in the organism versus time, also known as biota activity concentration. *Cw* is the activity concentration of ^{65}Zn pollutants in the water. The contaminant uptake procedure lasted 7 days. Similar experiments were conducted with ZnCl$_2$ concentrations of 1, 1.5, and 2 ppm, with each of the 3 *Portunus pelagicus* placed in a separate aquarium. The amount of ^{65}Zn tracer introduced was the same.

2.5 Measurement of ^{65}Zn activity in biota

4 aquariums of 16 L seawater were contaminated with radioisotope ^{65}Zn in the form of ZnCl$_2$ 0.5; 1; 1.5; 2 ppm in an aerated aquaria system with closed circulation (T: 27 °C, salinity: 35 ‰). During the contamination period, the ^{65}Zn activity measurement process was carried out every day. To measure radioactivity with an HPGE ORTEC detector, *Portunus pelagicus* was placed in a 250 ml plastic container and placed in a holder. Each time measurement had to be performed under the same conditions, which included the container's distance from the detector and the geometry of the container used. Each *Portunus pelagicus* was measured for 3 minutes.

2.6 Depuration

Following the bioaccumulation process, *Portunus pelagicus* was transferred to a new aquarium with 8 L of fresh seawater (T: 27 °C; salinity 35 ‰), free of contaminants. The biota was still fed on a daily basis. Every day for 7 days, the depuration of ^{65}Zn radionuclide was measured. The measurement procedure was the same as during the bioaccumulation experiment.

2.7 Data analysis

No *Portunus pelagicus* died during the acclimatization, bioaccumulation, or depuration experiments. Radionuclides released into the marine environment are concentrated in marine

biological species. The use of concentration factors is an easy way to explain the accumulation of radionuclides in biota relative to radionuclide concentrations in seawater. Then, the concentration factor becomes a tool for modeling the distribution and transfer of radionuclides in the aquatic environment and for predicting radioactivity in organisms. Many environmental parameters can modify the biokinetics of radionuclide accumulation and elimination in marine biota, but concentration factors remain an easy way to describe the process of radioactive and stable isotope concentrations in aquatic organisms (Carvalho, 2018). This experiment determined the level of Zn uptake and elimination in *Portunus pelagicus* using the radiotracer technique.

When the measured kinetics stabilized, the biokinetic model assumed the form of a simple linear regression, Eq. (2), or a simple first-order exponential kinetic model, as shown in Eq. (3):

$$CF_t = k_u t \qquad (2)$$

$$CF_t = CF_{ss}(1 - e^{-k_e t}) \qquad (3)$$

Where CF_t (g.mL^{-1}) is the concentration factor at time t (d), CF_{ss} (g.mL^{-1}) is the concentration factor at steady state, k_u (g.mL^{-1}d^{-1}) is the rate constant of absorption, and k_e (d^{-1}) is the rate constant of depuration (Budiawan et al., 2021). The kinetics of ^{65}Zn release are shown by the percentage of radioactivity concentration (activity concentration at time t divided by the activity concentration measured at the beginning of the depuration multiplied by 100). Simple linear regression was used to calculate release kinetics with and without semi-log data transformation. The following is an explanation using a simple model of exponential depuration Eq. (4):

$$A_t = A_0 e^{-k_e t} \qquad (4)$$

Where A_0, A_t, and k_e are the remaining concentration activities (%) at time t (0) and t and k_e are the elimination rate constant (d^{-1}).

3. Results and discussion

Portunus pelagicus ability to accumulate Zn is represented as a *CF*. The *CF* is the Zn activity in *Portunus pelagicus* divided by the Zn activity in water. The uptake and release kinetics of ^{65}Zn in the *Portunus pelagicus* is shown in **Figures 1** and **2**. The results showed that the higher the concentration of Zn, the lower the absorption kinetics; this was indicated by the smaller concentration factor (*CF*) value (**Figure 1**), and the uptake constant (k_u) (**Figure 3**) showed the same thing. Bioaccumulation is a process of accumulation of chemicals in an organism that occurs if the intake rate exceeds the rate of excretion (Popek, 2017). Chemicals are introduced into the organism through exposure to the abiotic environment (soil, water, air) or as dietary intake (trophic transfer).

Bioaccumulation of Zn metal based on the influence of concentration, from experimental results in the form of symbols and model calculation results in the form of lines, is shown in **Figure 1**. The estimated values of kinetic parameters with several organisms for comparison are shown in **Table 1**. Based on **Figure 1**, at 0.5 ppm Zn concentration, the *CF* on the primary day was 19.52 ml g^{-1}, and until the last day was 60.62 ml g^{-1}. The concentration of Zn 1 ppm on the 1st day was 13.45 ml g^{-1}, and until the 7th day was 38.27 ml g^{-1}. At a concentration of 1.5 ppm, the CF value on the 1st day was 11.15 ml g^{-1}, and the *CF* value on the 7th day was 34.47 ml g^{-1}. Meanwhile, the Zn concentration of 2 ppm on the primary day was 7.82 ml g^{-1}, and on the 7^{th,} day was 23.69 ml g^{-1}. These data indicate that the higher the concentration of Zn in the waters, the lower the accumulated Zn exposure in biota. *CF* values of Zn with various concentrations of ZnCl$_2$: 0.5, 1, 1.5, and 2 ppm at steady state reached on the sixth day were 59.47, 39.93, 35.46, and 25.55 g ml^{-1}, respectively (**Figure 1**). At the end of the experiment, the highest Zn uptake was observed at an initial concentration of 0.5 ppm and at a low of 2 ppm. The absorption and control mechanism of Zn metal
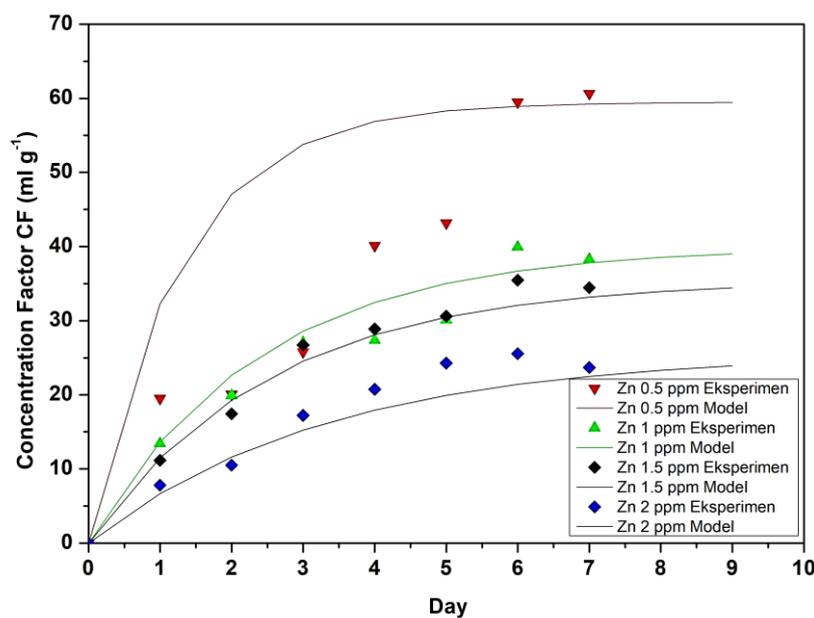
absorption in organisms is regulated based on the bioavailability of Zn in the environment. At low environmental levels of Zn concentrations, animals can absorb and retain Zn for important functions in tissues. Conversely, when Zn exposure levels are chronically elevated, aquatic animals can control bioaccumulation.

Marine biotas in their metabolic processes are able to process all contaminants that enter their bodies. Each biota has a different ability to accumulate Zn. This is related to the ability to absorb Zn into the cell wall, and then through the cell membrane, to remove contaminants from the cell. Most metals accumulate mainly in the kidneys, gills, and liver (Kucukosmanoglu & Filazi, 2020; Squadrone et al., 2020). Zinc accumulates in fish gills, disrupting the oxygen supply to tissues and causing hypoxia, which leads to death. However, if the water pH falls, heavy metals may be mobilized and discharged into the water column, endangering marine organisms such as crustaceans and insects (Bonsignore et al., 2018).
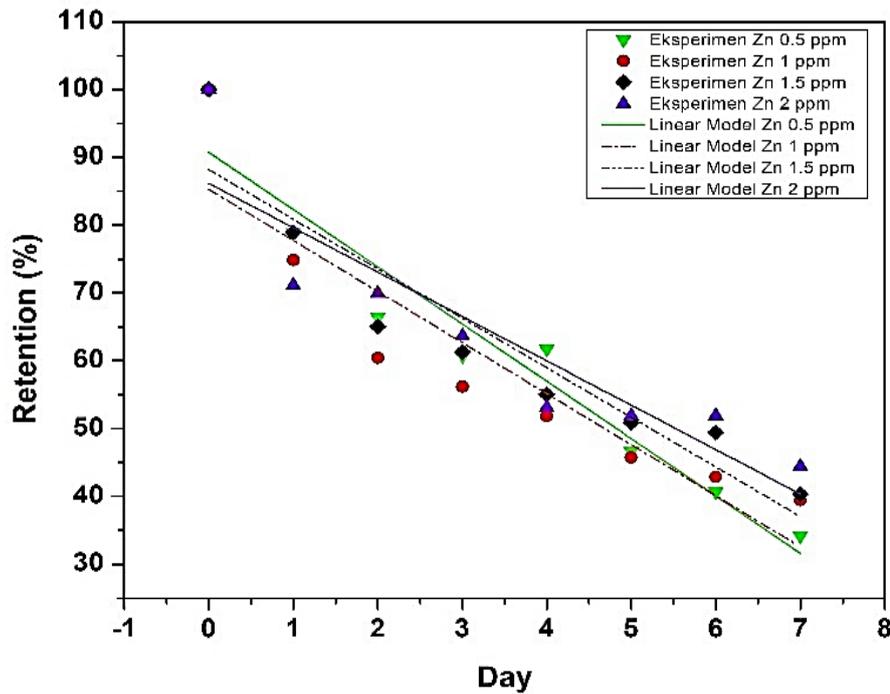
Table 1 Comparison of Zn, k_u and k_e, *CF* in the bioaccumulation of *Portunus pelagicus* with another biota.

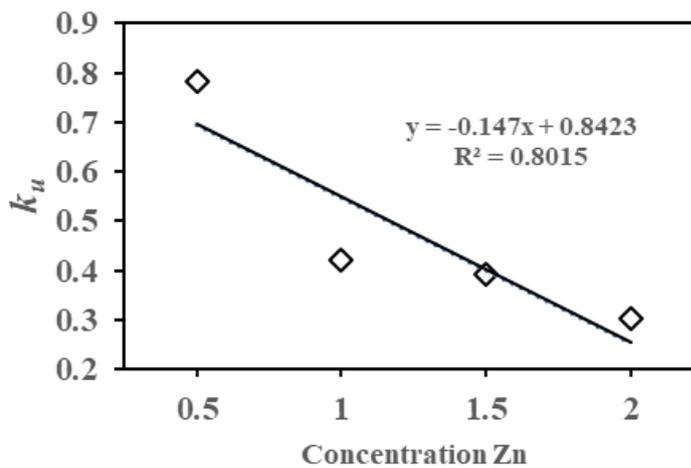| Biota	k_u (d^{-1})	k_e (d^{-1})	*CF* (mL.g^{-1})	Reference
Portunus pelagicus	0.47	0.08	19.43 - 37.99	Research
Colossoma macropomun	4.25	16.89	2.56 - 22.97	(Al Mustawa et al., 2022)
Trichogaster trichopterus	5.36	15.54	2.14 - 56.87	(Al Mustawa et al., 2022)
Modiolus micropterus	0.52	0.03	31.94 - 45.54	(Budiawan et al., 2021)
Glauconomya virens	-	-	11.14	(Budiawan et al., 2021)
Mytilus edulis	62.6	0.13	-	(Belivermiş et al., 2020)

Zn absorption was highest at a concentration of 0.5 ppm and decreased with increasing concentration. The correlation between Zn concentration and the ability of *Portunus pelagicus* to absorb contaminants (**Figure 1**) shows that the lower the Zn concentration, the greater the absorption kinetics; this is indicated by the linear regression $y = -0.147x + 0.8423$ with $R^2 = 0.8015$ (**Figure 3**), and the highest k_u value constant absorption is 0.7838 at a Zn concentration of 0.5 ppm (**Table 2**).

Figure 1 Kinetics of Zn absorption at concentrations of 0.5, 1, 1.5, 2 ppm.

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Figure 2 Kinetics of Zn released at concentrations of 0.5, 1, 1.5, 2 ppm.

Figure 3 Constant uptake linear Zn.

Table 2 [65]Zn absorption constant.

Zn (ppm)	k_u (d^{-1})
0.5	0.7838
1	0.4198
1.5	0.3925
2	0.3028

Concentration factor (*CF*) and uptake constant (k_u) data show that the value decreases with increasing Zn concentration. The heavy metals can dissolve in the aquatic environment and subsequently enter into the body of aquatic organisms depending upon certain conditions, e.g., pH, presence of complexing agents, etc. (Authman et al., 2015). In the course of the food chain, those

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metals then enter into the body of higher animals. Bioaccumulation of toxic heavy metals in the different tissues may harm animal health and cause damage to their normal physiological processes (Malik & Maurya, 2014). Heavy metal toxicity drastically affects the rate of survivability and reproductive capacity of organisms. Some of these have been reported to be highly carcinogenic, mutagenic, and teratogenic depending on the species, dose, and exposure time (Ngo et al., 2011). Exposure to acute concentrations of zinc sulfate in fish causes drastic degenerative histological changes in gills, liver, and kidneys, causing impaired organ function (Karanjkar & Deshpande, 2020).

The morphology of gills in marine biota is very important in its function in responding to environmental changes, and this is related to the physiological function of biota. Fish gills are responsible for a number of critical functions in addition to respiration: osmoregulation, excretion of nitrogenous waste, pH regulation, and hormone production (Herrero et al., 2018). Gills play an important role in gas exchange, acid-base regulation, osmoregulation, and excretion of nitrogenous waste (Foyle et al., 2020).

Based on the results of the bioaccumulation and depuration experiments of *Portunus pelagicus* for 14 days, k_u and k_e values were obtained (**Tables 2** and **3**). **Table 1** shows a comparison of *CF*, k_u, and k_e *Portunus pelagicus* values compared to other organisms. From **Table 1**, it can be seen that the *Portunus pelagicus* concentration factor (*CF*) has a moderate value and the smallest uptake constant (k_u) and elimination constant (k_e) value compared to other biota.

Table 3 ^{65}Zn elimination constant

Zn (ppm)	k_e (d^{-1})
0.5	0.0892
1	0.0752
1.5	0.073
2	0.0654

The uptake rate constant (k_u) is a value taken from the slope of the CF curve with respect to t (from t = 0 to t in equilibrium conditions). The relationship between the increase in Zn concentration and k_u is shown in **Figure 3**. The largest uptake rate constant was obtained at a Zn concentration of 0.5 ppm. The above curve shows that there is a correlation between the increase in Zn concentration and the uptake rate constant, as seen from the equation y = −0.147x + 0.8423 and the linear regression coefficient (R^2) = 0.8015.

The percentage of retention fraction in each concentration range showed a decrease in the percentage of retention during the 7 days of the contaminant release process (**Figure 2**). This discharge process produces a k_e value, or elimination rate constant. The k_e value represents the biota's ability to eliminate metal ion levels in its body. The elimination rate constant is calculated based on the slope of the graph of the percentage of retention in the biota's body against the release time. The highest contaminant release rate was obtained at 0.5 ppm Zn concentration. The experimental results show that the Zn release process is affected by the concentration of Zn in the water before it accumulates. This is obtained from the linear equation y = −0.0074x + 0.0941 and a linear regression coefficient (R^2) = 0.9154 (**Figure 4**).

Marine organisms have their own mechanisms when their environment changes due to pollutants. If environmental conditions change due to contaminants, marine organisms will spend a lot of energy to build and maintain shells in order to maintain the chemical health of body fluids; this is in accordance with research on crabs tested at heavy metal polluted sites with metabolomics analyses showing that metal pollution causes immune stress in crab gills, affecting energy metabolisms and inducing osmotic stress in crab samples (Yu et al., 2020). The pattern of heavy metal

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accumulation in the biota will vary depending on the habitat and eating habits of the biota, and also the influence of endogenous antioxidants and glucose metabolisms in the biota's muscles (Moniruzzaman et al., 2020).

Zn absorption and release based on the effect of concentration are shown in **Figures 1** and **2**. The *CF* values of Zn with various concentrations of 0.5, 1, 1.5, and 2 ppm at stable state were 37.99, 34.62, 29.71, and 19.43 mLg^{-1}, respectively (**Figure 1**). The greatest bioaccumulation kinetics was at a Zn concentration of 0.5 ppm, which was shown by the value of $k_u = 0.7838$ d^{-1} (**Table 2**), while the greatest release kinetics occurred at a Zn concentration of 0.5 ppm, which was shown by the value of $k_e = 0.0892$ d^{-1} (**Table 3**). At the end of the experiment, the highest Zn uptake was observed at the concentration of 0.5 ppm. **Figure 3** shows the kinetics of Zn bioaccumulation with increasing time, where X is the Zn concentration, and Y is the Zn uptake coefficient at *Portunus pelagicus*, which decreases with increasing Zn concentration.

The percentage of retention fraction in each concentration range showed a decrease in the percentage of retention during the 7 days of the contaminant release process (**Figure 2**). This depuration process produces a k_e value, or release rate constant. The k_e value represents the biota's ability to eliminate metal ion levels in its body. The release rate constant is calculated based on the slope of the graph of the percentage of retention in the body of *Portunus pelagicus* against the release time.
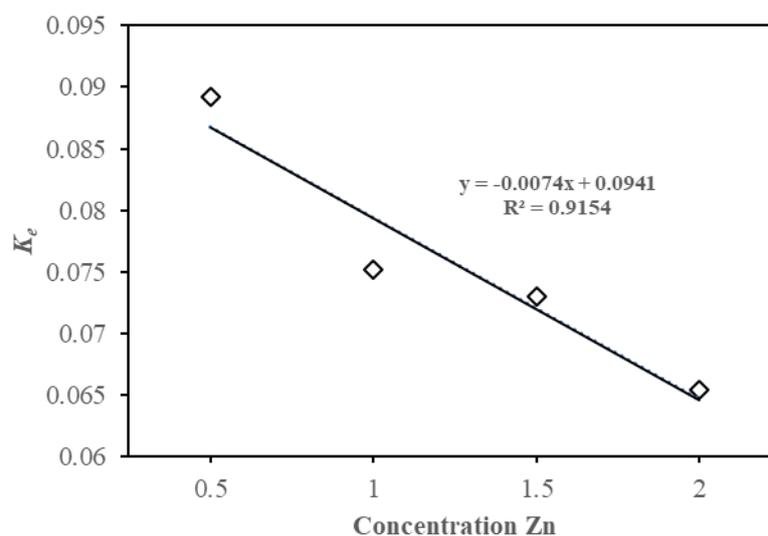
Figure 4 Linear elimination constant Zn.

Bioaccumulation and heavy metal detoxification experiments in estuarine crabs show that trace metals can be translocated to, and accumulated in, the hepatopancreas as the main organ of metabolism, and then eliminated under relieved conditions, with the appropriate reversibility of metallothionein (Truchet et al., 2020).

4. Conclusions

Blue swimming crab (*Portunus pelagicus*) is one of the featured marine products from Jakarta Bay, where the waters of Jakarta Bay are experiencing environmental degradation due to human activities. Research on ^{65}Zn bioaccumulation is needed to assess the uptake and degradation kinetics of *Portunus pelagicus* from Jakarta Bay, which is exposed to Zn through seawater. Absorption of ^{65}Zn by *Portunus pelagicus* through seawater displays a simple linear regression model which shows that the *CF* value of Zn with various concentrations of ZnCl$_2$: 0.5, 1, 1.5, and 2 ppm in a stable state are 59.47, 39.93, 35.46, and 25.55 g.ml^{-1}, respectively. The highest uptake constant (k_u) value was

obtained at the lowest Zn concentration of 0.5 ppm of 0.7838 d^{-1}, and the highest elimination constant (k_e) value was obtained at 0.5 ppm Zn concentration of 0.0892 d^{-1}.

Acknowledgement

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