Journal of Academic Biology and Biology Education
Vol. 2, No. 1, June 2025, pp. 97~105
ISSN: 3062-9705, DOI: 10.37251/jouabe.v2i1.2073

Comparative Effectiveness of Tamarind (*Tamarindus indica*) and Starfruit (*Averrhoa bilimbi*) Solutions in Reducing Lead (Pb) Levels in Shellfish (*Polymesoda erosa*)

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Article Info

Article history:

Received Mar 23, 2025
Revised Apr 7, 2025
Accepted Jun 9, 2025
Online First Jun 23, 2025

Keywords:

Lead (Pb)
Polymesoda erosa
Starfruit
Tamarind

ABSTRACT

Purpose of the study: This study aims to determine the effectiveness of soaking duration in tamarind (*Tamarindus indica*) and starfruit (*Averrhoa bilimbi*) solutions in reducing the concentration of heavy metal lead (Pb) in shellfish (*Polymesoda erosa*) before and after treatment.

Methodology: This study employed a descriptive method using Atomic Absorption Spectrophotometry (AAS) to measure Pb levels in *Polymesoda erosa* clams. Samples from Paotere Market, Makassar, were soaked in tamarind or starfruit solutions for 30, 60, and 90 minutes, then analyzed using AAS at a wavelength of 217.0 nm.

Main Findings: Soaking *Polymesoda erosa* in tamarind solution for 30, 60, and 90 minutes reduced Pb levels by 30.69%, 40.92%, and 50%, respectively. Soaking in starfruit solution for the same durations reduced Pb levels by 19.32%, 65.91%, and 93.18%, respectively. Starfruit solution soaking for 90 minutes achieved Pb levels below the permissible consumption limit.

Novelty/Originality of this study: This study introduces the comparative use of tamarind (*Tamarindus indica*) and starfruit (*Averrhoa bilimbi*) solutions to reduce Pb levels in *Polymesoda erosa*. It reveals starfruit's superior effectiveness due to higher citric acid content, offering a simple, natural, and low-cost method for heavy metal reduction in seafood, potentially improving food safety in coastal communities.

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1. INTRODUCTION

The sea is where rivers flow, which has the potential to collect pollutants. These pollutants generally originate from industrial and human activities [1], [2]. The increase in industrial activity and human activity is always accompanied by an increase in waste. Waste containing toxic and hazardous chemicals is carried by rivers, which ultimately flow into the sea, contaminating the environment [3], [4]. The entry of waste into waters can cause water pollution, contaminating rivers and seas and accumulating in the food chain [5], [6]. Heavy metals can enter and accumulate in the bodies of aquatic biota such as shrimp, squid, and shellfish [7]-[9].

Mollusks, especially shellfish, are aquatic commodities with significant economic value, as evidenced by their production volume. *Polymesoda erosa*, locally known as the kepah clam, is a bivalve species inhabiting tidal zones of mangrove forests in tropical and subtropical regions. This shellfish plays an important role in coastal ecosystems as a benthic filter feeder and serves as a valuable food source for local communities. Its high economic value stems from its demand in local markets and its role as a source of animal protein.

As a relatively sedentary marine organism, the kepah clam exhibits growth patterns and physiological adaptations closely linked to its habitat, which also make it susceptible to bioaccumulation of pollutants, particularly heavy metals, from surrounding sediments and water [10], [11]. Heavy metals are one of the most dangerous pollutants for human health [12], [13]. The World Health Organization (WHO) and the Food and Agriculture Organization (FAO) recommend avoiding seafood contaminated with heavy metals [14]-[16]. Heavy metals have long been recognized as highly toxic and accumulate in human organs [17], [18]. In some cases, they can even cause death.

Heavy metals that enter waters will pollute the sea. In addition to polluting the water, heavy metals will also settle at the bottom of the water, having a residence time of up to thousands of years. Heavy metals will concentrate in the bodies of living things through bioaccumulation and biomagnification processes through several pathways: through the respiratory tract, the digestive tract, and through the skin [19], [20]. The high heavy metal content in shellfish is due to the low mobility of shellfish and their persistence in specific habitats, namely in sediments or the seabed, allowing for more intensive bioconcentration and bioaccumulation processes [21], [22]. Accumulation of heavy metals, such as lead (Pb), often occurs in raw shellfish and causes poisoning in people who consume them due to its high toxicity [23], [24].

The amount of heavy metals that accumulate in the body tissues of aquatic animals that are still suitable for human consumption is determined by a standard, with the maximum limit for Pb contamination in food reaching 1.0 mg/kg (WHO). According to the Indonesian National Standard (SNI) 7387:2009, the heavy metal lead enters the body through respiration and food. Consuming large amounts of lead directly causes tissue damage. In infants and children, excessive exposure to lead can cause brain damage [25], [26]. The maximum limit for Pb contamination in food, especially shellfish (bivalves), is 2 mg/kg.

Tamarind, scientifically known as *Tamarindus indica L.,* is a tropical plant and a legume [27], [28]. Tamarind pulp contains 8-14% tartaric acid, 30-40% sugar, citric acid, and potassium bicarbonate, giving it a very sour taste. Starfruit (*Averrhoa bilimbi*) has traditionally been used as a food additive and medicinal ingredient [29], [30]. Starfruit contains formic acid, citric acid, ascorbic acid (vitamin C), saponins, tannins, glucosides, flavonoids, and several minerals, especially calcium and potassium in the form of potassium citrate and calcium oxalate [31].

Various studies have utilized natural acids, such as tamarind, to reduce heavy metal (Pb) levels in marine biota. For example, Amelia et al. found that soaking gonggong snails in a 10% tamarind solution for 60 minutes reduced Pb by 47.1% [32]. Meanwhile, a study by Santi et al. achieved an efficiency of up to 98.89% in reducing Pb in red mussels when soaked in a 25% tamarind solution for 60 minutes [33]. On the other hand, research on Palu anchovies showed that 5% tamarind only reduced 1.64%, while 100% starfruit reduced 1.39% [34]. However, no studies have been found that specifically examine the effectiveness of soaking using tamarind and starfruit solutions on the skin (*Polymesoda erosa*). This gap is important to fill in order to know the effectiveness of reducing Pb in kepah, including optimizing concentration, soaking time, and direct comparison between tamarind and starfruit, information that is not currently available.

The urgency of this research lies in the high risk of Pb contamination in shellfish consumed by coastal communities, which can directly threaten public health, especially in areas where mussels are the primary protein source. Current methods for mitigating Pb contamination often rely on chemical treatments or lengthy depuration processes, which are less accessible and more expensive for local fishermen and small-scale seafood industries. The use of readily available, inexpensive, and safe natural acids such as tamarind and starfruit offers a practical alternative that can be applied directly at the community level to reduce Pb contamination before consumption [35], [36]. Given that mussels are relatively sedentary benthic organisms with a high bioaccumulation capacity, determining an effective and affordable detoxification method is crucial to ensure food safety and maintain consumer confidence.

The novelty of this study lies in its specific focus on evaluating and comparing the effectiveness of tamarind and starfruit solutions in reducing Pb levels in *Polymesoda erosa*, a species for which no similar comparative detoxification study currently exists. The use of easily obtained, cheap and safe natural acids such as tamarind and starfruit is a practical alternative that can be applied directly at the community level to reduce Pb pollution before consumption. This research therefore provides new empirical evidence that can contribute to the development of practical, locally adapted, and scientifically validated detoxification techniques for Pb in shellfish. This research was conducted to determine the effect and comparison of soaking using a solution of tamarind and star fruit in reducing the content of the heavy metal Pb (lead) in kepah clams so that they can be consumed by the public.

2. RESEARCH METHOD

This research is quantitative with a descriptive approach. This method is intended to describe existing phenomena, whether they are occurring now or in the past [37], [38]. This method is a validation or testing method, namely testing the influence of one or more other variables. The influencing variable is grouped as the independent variable, and the influenced variable is grouped as the dependent variable. The dependent variable in this study is the concentration of the heavy metal lead (Pb) in mussels. The independent variable in this study is the soaking time of mussels in tamarind and starfruit solutions.

Mussel samples were obtained from the Paotere market in Makassar City and analyzed in the Analytical Chemistry Laboratory, 1st Floor, Faculty of Science and Technology, Alauddin State Islamic University of Makassar. The data collection method used the SSA (Atomic Absorption Spectrometry) test. Mussel samples were taken from the Paotere market in Makassar City and then tested in tamarind and starfruit solutions at different soaking times. The tools used in this study were plastic bags, Erlenmeyer flasks, Teflon bombs, beakers, hot plates, measuring flasks, freezers, ovens, and water baths. The materials used in this study were clam shells, HNO_3, $HClO_4$, aquabidest, tamarind solution, and starfruit.

The procedure began with the collection of *Polymesoda erosa* clam samples directly from Potere Market, Makassar City. The samples were placed in airtight plastic bags to prevent metal contamination during transport to the laboratory. The experimental treatments involved soaking the clam meat in starfruit (*Averrhoa bilimbi*) and tamarind (*Tamarindus indica*) solutions for durations of 30, 60, and 90 minutes, each in two replications. A control group was soaked in distilled water (aquadest). The treatment layout consisted of: Lo (no solution), L1 (tamarind solution), and L2 (starfruit solution), with soaking times W0 (0 minutes), W1 (30 minutes), W2 (60 minutes), and W3 (90 minutes). Prior to treatment, standard Pb solutions were prepared by pipetting 0, 0.1, 0.2, 0.5, 1, and 2 mL of a 100 mg/L Pb stock solution into 100 mL volumetric flasks, followed by dilution with demineralized water to the mark and homogenization. Initial Pb concentrations in the clam samples were measured before treatment.

For the soaking process, 5 grams of wet clam meat were weighed and placed into Erlenmeyer flasks containing either tamarind or starfruit solutions for the designated times. The samples were then drained using plastic sieves and rinsed for further analysis. Sample preparation involved heating the soaked samples on a hot plate at 100°C with the addition of 5 mL concentrated nitric acid (HNO_3) until the tissue was fully digested and the solution became clear. Then, 1 mL of perchloric acid ($HClO_4$) was added, and the mixture was filtered through Whatman filter paper into a 100 mL volumetric flask. The solution was then brought to volume with sterile bidistilled water and homogenized. The final measurement of Pb concentration was conducted using Atomic Absorption Spectrophotometry (AAS) at a wavelength of 217.0 nm, and the resulting concentration values in mg/L were converted to mg/kg.

The results of the measurement of a series of standard Pb solutions, then a standard/calibration curve is created which is a plot between the absorbance value (y-axis) and the standard concentration (x-axis). Based on this standard curve, a linear equation can then be obtained, namely y = ax + b. The results of the absorbance measurement of the sample solution are then entered into the linear equation above to obtain the concentration of the sample solution. Meanwhile, the Pb concentration in the sample in mg/kg wet sample weight is calculated according to the following equation.

$$M = \frac{C \times V \times fp}{B} \quad \ldots(1)$$

where M is the Pb content in the sample (mg/kg), C is the concentration obtained from the calibration curve (mg/L), V is the volume of the sample solution (L), fp is the dilution factor, and B is the sample weight (kg). The reduction in Pb concentration due to treatment uses the following formula:

$$MA\text{-}B = MA - MB \quad \ldots(2)$$

where MA-B is the concentration of Pb reduction due to treatment (mg/kg), MA is the concentration of Pb before treatment (mg/kg), MB is the concentration of Pb after treatment (mg/kg).

3. RESULTS AND DISCUSSION

Analysis of Pb metal content in mussels in this study was conducted using the calibration curve method. In this method, a series of standard solutions of known concentrations are prepared and their absorbance is measured using an atomic absorption spectrophotometer. The obtained absorbance is then plotted against the concentration of the standard solution to obtain a standard/calibration curve. Based on this curve, a linear equation is then determined. The absorbance of the sample solution of unknown concentration is then measured with the instrument under the same conditions. The obtained absorbance is then entered into the linear equation obtained to obtain the determined sample concentration. The standard curve for measuring the absorbance of Pb metal standard solutions at various concentrations is shown in Figure 1.

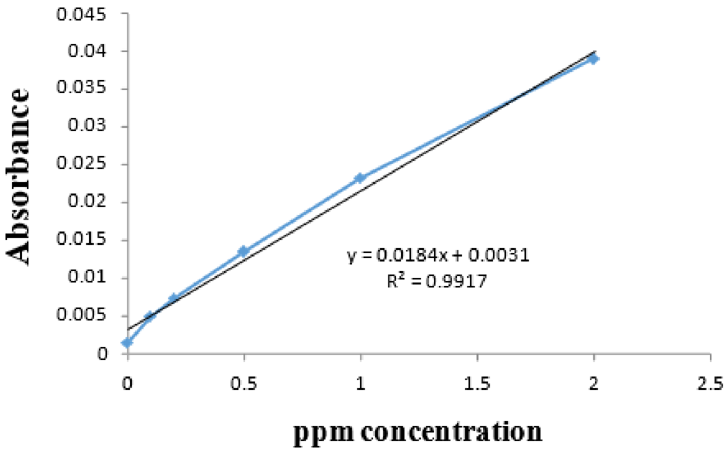

Figure 1. Standard curve of Pb metal

The standard curve in Figure 1 shows that the measured absorbance is a function of the concentration of the standard solution. The absorbance of the standard solution increases with increasing concentration, indicating a linear relationship between concentration and absorbance. This is supported by the correlation coefficient of 0.9917, which is close to 1. The linear equation obtained from this measurement is y = 0.0184x – 0.0031, where y is the absorbance and x is the concentration (ppm).

Based on the obtained standard curve, the absorbance of the sample solution was then measured. The average Pb content in the clam samples, with varying soaking times in tamarind solution and without soaking, is presented in Table 1. The concentrations listed are the average concentrations from the study, which was repeated twice.

Table 1. Concentration of the heavy metal lead (Pb) in kepah clams soaked in tamarind solution

Soaking Time	Pb Concentration (mg/kg)	Pb Concentration Reduced (mg/kg)	Pb Concentration Reduction (%)
0 minutes	4.782	-	-
30 minutes	3.314	-	30.69
60 minutes	2.825	1.468	40.92
90 minutes	2.391	1.957	50.00

The data in Table 1 above illustrates that the concentration of Pb metal in the unsoaked clams (control) was higher than that soaked in tamarind solution. The Pb concentration in the unsoaked clam meat was 4.782 mg/kg, while the Pb concentration in the bamboo clam meat soaked for 30, 60, and 90 minutes was 3.314 mg/kg, 2.825 mg/kg, and 2.391 mg/kg, respectively. The decrease in Pb metal concentration with the length of soaking time, as shown in Table 1 above, is presented in Figure 2.

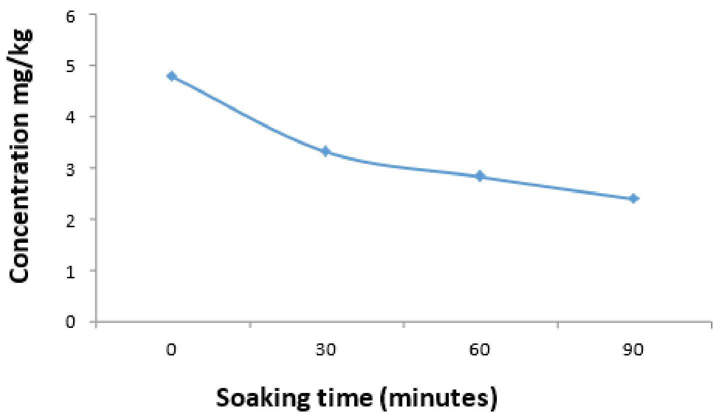

Figure 2. Concentration of the heavy metal lead (Pb) in kepah clams soaked in tamarind solution

The obtained standards were then measured for absorbance of the sample solution. The average Pb metal content in the clam samples at various soaking times in the starfruit solution and without soaking is presented in Table 2.

Table 2. Concentration of the heavy metal lead (Pb) in kepah clams soaked in starfruit solution

Soaking Time	Pb Concentration (mg/kg)	Pb Concentration Reduced (mg/kg)	Pb Concentration Reduction (%)
0 minutes	4.782	-	-
30 minutes	3.858	0.924	19.32
60 minutes	1.630	3.152	65.91
90 minutes	0.326	4.452	93.18*

Description: *: Meets standards for consumption

The data in Table 2 above illustrates that the concentration of Pb metal in the unsoaked clams (control) was higher than that soaked in the starfruit solution. The Pb concentration in the unsoaked clam meat was 4.782 mg/kg, while the Pb concentration in the bamboo clam meat soaked for 30, 60, and 90 minutes was 3.858 mg/kg, 1.63 mg/kg, and 0.326 mg/kg, respectively. The decrease in Pb metal concentration with the length of soaking time, as shown in Table 2 above, is presented in Figure 3.

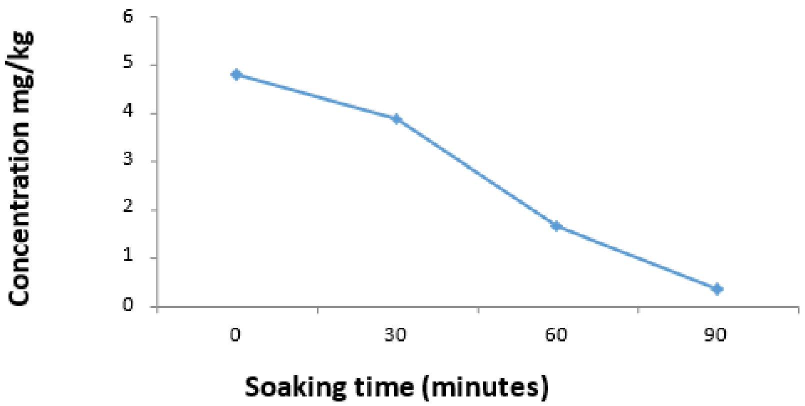

Figure 3. Concentration of the heavy metal lead (Pb) in kepah clams soaked in starfruit solution

Based on the decision of the Director General of the Food and Drug Monitoring Agency (BPOM), the maximum limit of Pb metal contamination in shellfish is 1.5 mg/kg (Decree of the Minister of Maritime Affairs and Fisheries, 2004). Likewise, the maximum Pb content limit in food set by the FAO is 2 ppm. The results of determining Pb content using an Atomic Absorption Spectrometer showed that Pb metal had exceeded the permitted threshold of 4.782 mg/kg, considering that Pb is one of the toxic metals for organisms. This is caused by the accumulative nature of non-essential metals such as Pb in shellfish body tissue because the nature of these metals tends to form complex bonds with organic matter. The high Pb content in the body tissue of the clam is because this type of organism cannot excrete Pb metal properly, so it will accumulate continuously in the body tissue of clams living in the Badung River Estuary Reservoir, which has been contaminated with Pb at quite high concentrations. Heavy metal levels in water bodies gradually increase due to human activity, resulting in their absorption and accumulation in fish tissue (bioaccumulation). Heavy metal pollution of the environment is a process closely related to human use of these metals. Industrial production processes requiring high temperatures, such as oil refining, coal mining, oil-powered power generation, and metal casting, produce significant amounts of pollutant waste, particularly metals that are relatively volatile and water-soluble (in ionic form), such as lead (Pb). Pollution can also be caused by cement factories, steel smelting waste, and waste ashing.

Pb can enter shellfish through the food chain, gills, and diffusion through the skin surface. Pb accumulation in green mussels occurs through absorption from water, particles, and plankton. Furthermore, high Pb concentrations in shellfish tissue are closely related to the high Pb content in the water and sediment. Besides rivers, heavy metals in water can also enter the air, particularly lead (Pb) used in fuel blends. The increasing pace of development in all sectors has resulted in increased air pollution through motor vehicle emissions. The role of the government and all stakeholders is crucial in creating a clean environment and reducing the ongoing rate of pollution.

Soaking clam meat in tamarind and starfruit solutions reduces lead (Pb) levels. The longer soaking time reduces the Pb concentration. The percentage reduction in Pb levels in clam meat is presented in Tables 1 and 2.

Table 4.1 shows that the Pb concentrations that can be reduced during soaking clam meat in tamarind solution for 30, 60, and 90 minutes are 1.468 mg/kg, 1.957 mg/kg, and 2.391 mg/kg, respectively. The magnitude of the reduction in the concentration of Pb metal that can be reduced has a percentage of 30.69%, 40.92% and 50% respectively from the initial concentration (control) of Pb metal in the mussel meat.

Table 2 shows that the reducible Pb concentrations during immersion of clam meat in starfruit solution for 30, 60, and 90 minutes were 0.924 mg/kg; 3.152 mg/kg; and 4.456 mg/kg, respectively. The reductions in reducible Pb concentrations were 19.32%, 65.92%, and 93.18%, respectively, compared to the initial (control) Pb concentration in clam meat.

Tables 1 and 2 show an increase in the percentage of Pb reduction with the length of immersion. This increase is shown in Figures 4 and 5.

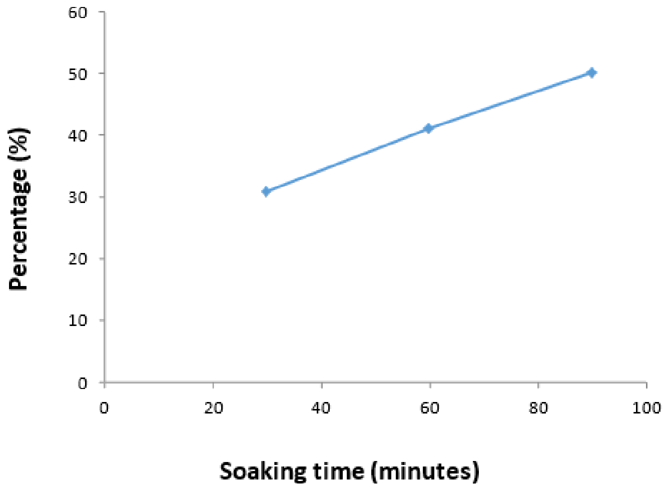

Figure 4. Decrease in Pb concentration with soaking time using tamarind solution

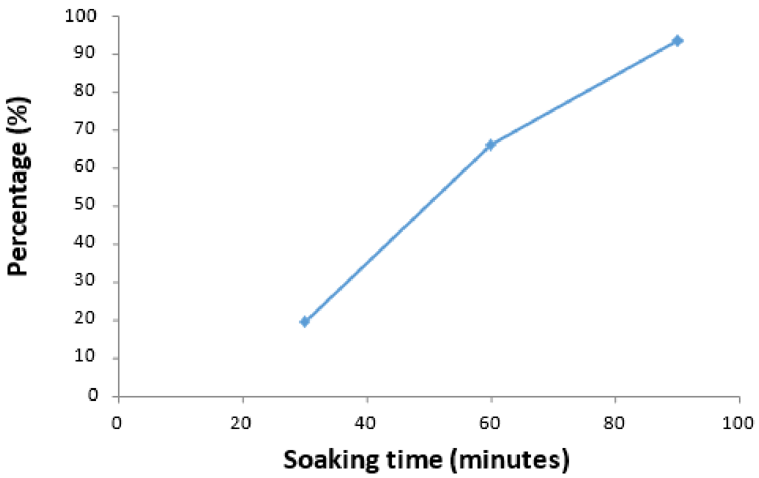

Figure 5. Decrease in Pb concentration with soaking time using starfruit solution

The percentage reduction in Pb levels with soaking time using tamarind and starfruit solutions can reduce the maximum limit of Pb metal concentration allowed in marine food ingredients. The highest percentage reduction was in the 90-minute tamarind solution soaking, namely 50%, while the 90-minute starfruit solution soaking was 93.18%. Soaking clams in starfruit solution was more effective than soaking in tamarind solution. This is because the citric acid content in starfruit is higher than that in tamarind. Citric acid can also function as a chemical compound that binds metals in the form of complex bonds. This property can minimize the adverse effects of metals in food ingredients. Thus, this compound can help reduce the levels of heavy metals such as lead in shellfish. Citric acid can bind lead metal through lone electron pairs owned by three carboxylate groups (-COOH) and from hydroxyl groups bound to carbon atoms. Citric acid is a sequestrant (metal-binding agent).

Citric acid has the chemical formula $CH_2COOH-C(OH)COOH-CH2COOH$ ($C_6H_8O_7$). The –OH and –COOH functional groups in citric acid enable citrate ions to react with metal ions to form citrate salts.

The findings of this study reinforce the utility of natural materials and natural acid derivatives as effective agents in heavy metal (Pb) reduction in aquatic biota. For example, Mandal et al. introduced a tamarind-based chelating resin called TTAPA that effectively sequesters heavy metals from industrial wastewater, providing a solid foundation for understanding the role of tamarind in chemically binding Pb [39]. In addition, the use of activated carbon modified with lemon juice from rice husks has been proven to be effective in absorbing Pb in aqueous solutions, expanding the application range of natural acids in heavy metal adsorption [40]. Meanwhile, an alternative approach using Brachidontes variabilis shell powder showed that marine biota can function as Pb adsorbents up to 99.2% efficiency in freshwater environments, although it decreases in seawater due to interference from other ions [41]. These findings are directly relevant to your research using immersion in tamarind and starfruit solutions to reduce Pb in Polymesoda erosa, and support the theory that natural organic compounds—particularly citric acid and tamarind derivatives—can play an effective role in binding and reducing Pb accumulation in the tissues of marine biota.

The novelty of this study lies in its specific focus on evaluating and comparing the effectiveness of tamarind (*Tamarindus indica*) and starfruit (*Averrhoa bilimbi*) solutions in reducing lead (Pb) levels in mussels (*Polymesoda erosa*), a shellfish species with high economic value but is vulnerable to heavy metal exposure in estuarine habitats. Unlike previous studies that tested only one type of natural acid on specific marine biota, this study combined two types of natural acid solutions in a controlled experimental design, with varying immersion times (30, 60, and 90 minutes) to obtain a comparative picture of their effectiveness. This approach provides new, previously unreported empirical data for clams and offers a scientific basis for selecting a simple, inexpensive, and readily applicable heavy metal detoxification method at the coastal community level.

The implication of this research is the need for public awareness campaigns regarding the use of tamarind and starfruit solutions as alternative food additives to easily and simply reduce lead (Pb) content in shellfish processing. This study was only conducted under laboratory conditions with one solution concentration and without sensory evaluation or the effect on other heavy metals and long-term stability, so the results need to be verified at various concentrations and on a field scale. Recommendations for further research include increasing the soaking time to achieve better results. Furthermore, the effective dosage of tamarind and starfruit in reducing lead (Pb) levels is also being investigated. Further research is needed to determine the differences in protein nutritional value between unsoaked and soaked clams in tamarind and starfruit solutions.

4. CONCLUSION

The concentration of immersion of clams using tamarind solution for 30, 60 and 90 minutes respectively was 3.314 mg/kg; 2.825 mg/kg; and 2.391 mg/kg; while the concentration of immersion using starfruit solution for 30, 60 and 90 minutes respectively was 3.858 mg/kg; 1.63 mg/kg; and 0.326 mg/kg. The immersion time of tamarind and starfruit solution effectively reduced the levels of heavy metal lead (Pb) in clams, at the 30th, 60th and 90th minutes, respectively tamarind 30.69%; 40.92%; and 50%; and starfruit 19.32%; 65.91% and 93.18%. Recommendations for further research include increasing the soaking time to achieve better results. Furthermore, the effective dosage of tamarind and starfruit in reducing lead (Pb) levels is also being investigated. Further research is needed to determine the differences in protein nutritional value between unsoaked and soaked clams in tamarind and starfruit solutions.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the laboratory facilities and technical assistance provided, and thank all colleagues and laboratory staff whose support contributed to the completion of this research.

AUTHOR CONTRIBUTIONS

Conceptualization, A.M. and M.G.B.M.; Methodology, A.M. and M.G.B.M.; Software, A.M.; Validation, A.M., M.G.B.M. and S.T.; Formal Analysis, A.M.; Investigation, A.M.; Resources, M.G.B.M. and S.T.; Data Curation, A.M.; Writing – Original Draft Preparation, A.M.; Writing – Review & Editing, A.M., M.G.B.M. and S.T.; Visualization, A.M.; Supervision, M.G.B.M. and S.T.; Project Administration, M.G.B.M.; Funding Acquisition, S.T.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

USE OF ARTIFICIAL INTELLIGENCE (AI)-ASSISTED TECHNOLOGY

Not applicable.

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