

Bioaccumulation of heavy metals in *Donax obesulus* from the Lambayeque coast, Peru

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Abstract: *Donax obesulus* is a bivalve widely mined collected and consumed widely by the local coastal population of the Lambayeque region, Peru. *Donax obesulus* lives in the coastal marine environment and presents anthropogenic disturbances, including pollutants such as heavy metals (cadmium, copper, and lead). The objectives were to quantify and evaluate the concentrations of copper, lead, and cadmium in edible tissues of *Donax obesulus*. The samples were collected from three different beaches on the Lambayeque coast (from south to north: Lagunas, San José, and El Gigante) between May and July 2013, and a physical-chemical treatment was performed to detect the heavy metal concentrations by inductively coupled plasma optical emission spectroscopy (ICP-OES). The results showed that copper presented a cumulative trend in tissues of *Donax obesulus* with the highest concentration collected at San José beach in July (10.1 µg g⁻¹). In contrast, the concentrations of lead (> 0.3 µg g⁻¹) and cadmium (> 0.2 µg g⁻¹). *Donax obesulus* presents bioaccumulation of copper, lead, and cadmium.

Keywords: *Donax obesulus*, bioaccumulation, heavy metals, copper, cadmium, lead.

Resumen: *Bioacumulación de metales pesados en Donax obesulus del litoral de Lambayeque, Perú*

Donax obesulus es un bivalvo ampliamente extraído, recolectado y consumido ampliamente por la población costera local de la región de Lambayeque, Perú. El ambiente costero-marino donde vive *Donax obesulus* presenta perturbaciones antropogénicas, incluyendo contaminantes como metales pesados (cadmio, cobre y plomo). Los objetivos de esta investigación fueron cuantificar y evaluar las concentraciones de cobre, plomo y cadmio en tejidos comestibles de *Donax obesulus*. Donax obesulus las muestras fueron recolectadas en tres playas diferentes de la costa de Lambayeque (de sur a norte: Lagunas, San José y El Gigante) entre mayo y julio de 2013. Las muestras fueron sometidas a una prueba toxicológica. Se les realizó un tratamiento físico-químico, realizado para obtener las concentraciones de metales pesados mediante espectrometría de emisión óptica de plasma acoplado inductivamente (OES) (ICP). Los resultados mostraron que el cobre presentó una clara tendencia acumulativa en *Donax obesulus* con la mayor concentración en la playa de San José en julio (10.1 µg g⁻¹). En cambio, las concentraciones de plomo (> 0,3 µg g⁻¹) y cadmio (> 0,2 µg g⁻¹) fueron escasas en todas las áreas de muestreo durante todo el curso de la investigación. A lo largo del tiempo que abarcó la investigación. La bioacumulación de cobre, plomo y cadmio que se encuentra en *Donax obesulus* podría NO vulnerar la seguridad alimentaria de sus consumidores.

Palabras clave: *Donax obesulus*, Bioacumulación, metales pesados, moluscos, medio marino costero, contaminación.

Introduction

Many industrialized countries do not adequately manage their waste or discard it. This fact is reflected in the increasing discharge of industrial and urban effluents to the ocean seas and rivers. A great

diversity of pollutants ends up in the environment being harmful to wildlife, causing ecological and sanitary problems (Liu *et al.*, 2019). One of the biggest problems issues of aquatic contamination pollution is caused by heavy metals, these contaminants are passively assimilated by different organisms during their feeding process (Goretti *et al.*, 2016). The impact of heavy metals on the environment is a serious and growing problem worldwide because of their availability to be accumulated by many aquatic organisms.

The bivalve genus of mollusks *Donax* includes bivalves distributed vertically aligned into the sand on in exposed beaches, of tropical and temperate coasts around the world, such as is the case of the northern coast of Peru (Warner *et al.*, 2016, 2019). These bivalves assimilate heavy metals during the filtration of their food, giving way to bioaccumulation and subsequent biomagnification in the food chain (Shahsavani *et al.*, 2017).

The purification processes in bivalves serve to eliminate microorganisms, but they do not eliminate toxic chemical agents (Anacleto *et al.*, 2015). Within among heavy metal elements pool, cadmium is a non-essential trace element toxic at low concentrations to aquatic organisms, however, is bioaccumulated in bivalves, without any specific function known in these marine beings, so far (Merad *et al.*, 2018, Shi *et al.*, 2016). On the other hand, (Merad *et al.*, 2018), while copper, in its oxidized form, induces genotoxicity in bivalves (Chelomin *et al.*, 2017).

Besides its consumption, currently, some marine bivalves are considered as potential accurate biocontrol tools, capable of monitoring heavy metal pollution in aquatic environments, detecting such as bioavailable lead along the coasts, for instance (Cariou *et al.*, 2017). Cadmium, one of the main acidifiers in the ocean, bioaccumulates in bivalves, until now its function has not been well studied in these marine beings (Shi *et al.*, 2016). This skill becomes converting them, in addition to being a source of food, into a useful method to know the state of health of a coastal-marine ecosystem.

The significance of knowing the concentration of heavy metals in mollusks does not only based on relies on the possibility of being able to market them under a standard of food control and safety since there is a significant population that depends on these resources. (Shahsavani *et al.*, 2017), but also for their potential use as biomonitors. The objectives aim of this research were to quantify and evaluate the concentrations of copper, lead, and cadmium in edible tissues of *D. obesulus* (Bivalvia: Donacidae) from the coast of the Lambayeque, Peru.

Methods

Study area

A descriptive cross-sectional study was performed, the population was constituted by adults of *D. obesulus* (Bivalvia: Donacidae), coming from the intertidal zone of the following beaches: Lagunas (07°02'49"S, 79°44'48"W), San José (06°45'25"S, 79°58'46"W), and El Gigante – Mórrope (06° 29'22''S, 80° 23'38''W) from Lambayeque (Figure 1) during the months' May to July in 2013. Adults were collected and weighed 250 g of edible tissue. A morphometry measure including size

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structure total length (valve length) and total weight with a weight-length relationship was made to describe the individuals.

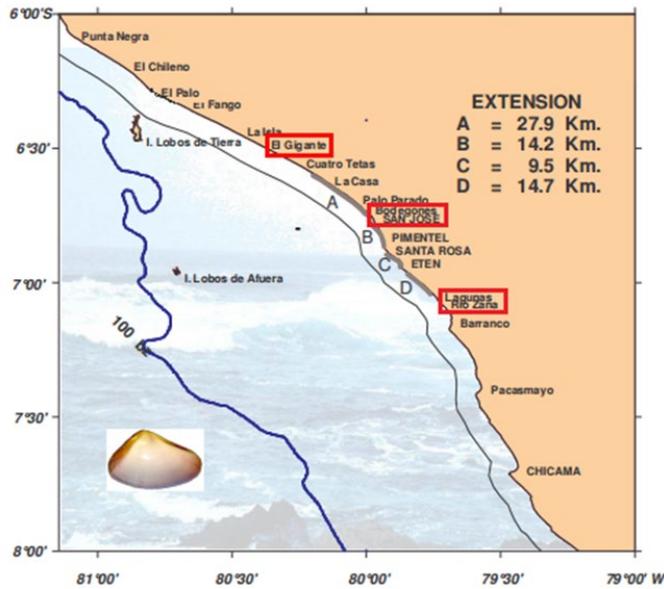


Figure 1. Location of sampling areas (in red) of the Lambayeque coast.

Heavy metal measurement

It was taken randomly only 50 g from the first sample quantity were randomly to obtain a uniform mass separated and liquefied in a homogenizer, obtaining uniform mass. Approximately 1 g of the homogenized sample was separated and taken for the chemical digestion process (acid attack). The soft tissue digestion, each sample was respectively poured into a Teflon test tube with 4.5 to 5 ml ultrapure water. Two repetitions were performed for each sample. A tube was filled with ultrapure served as a control tube.

It was added 9 mL 98 % nitric acid (HNO₃) and 3 mL of 97 % hydrochloric acid (HCl) respectively for each Teflon tube for the digestion process. To accelerate this step, catalyze the digestion, a microwave programmed with a method for animal tissues digestion was used: using a temperature ramp of 170 °C at 800 watts/7 minutes (indicated for fish, mollusks, and crustaceans). The result of the digestion was filtered (filter paper) into 100 mL flasks. Flasks were further flushed with ultra-pure water.

Using a high-performance detector (características), the values obtained were interpreted by the computer and translated into concentrations of cadmium, lead, and copper present in the processed *D. obesulus* sample. Faltan detalles de la tecnica y equipo usados, niveles de detección, etc.

Statistical analysis

To determine statistical differences in cadmium, lead, and copper concentrations in edible bivalve mollusks, data were expressed as mean and standard deviation (SD) using InfoStat software. The comparison of the mean values between areas (for the same station) was estimated by Student's t-test. The effect of the station and the sampling area was tested with a significance level of p <0.05. Dataset is available in <https://doi.org/10.6084/m9.figshare.14850540.v1>.

Results

In Lagunas beach, the largest number of individuals collected was recorded in July, with a total of 424 specimens, which is the minimum size in May, with a length of 11.55 mm, in addition to being the smallest size found in the investigation (Figure 2). The maximum measured length corresponded to July with 33.46 mm. The average length on the beach ranged from 25.34 mm to 28.75 mm (Table 1).

Table 1. Valvar length (mm) of the individuals found from May to July in Lagunas beach, Lagunas district, Chiclayo province, 2013.

<i>Donax obesulus</i>	Beach Lagunas		
	May	June	July
Number	301	333	424
Minimum	11.55	20.68	13.92
Maximum	31.50	31.67	33.46
Average	26.87	28.75	25.34
Standard deviation	2.85	1.63	4.34
Coefficient of variation	10.59	5.68	17.15

271 individuals were collected at the San José beach. The minimum size found corresponds to 11.78 mm and the maximum 41.3 mm during the beginning of the sampling in May, being, in turn, the largest size recorded in the sampled specimens (Figure 3). The mean valve length was between 24.48 mm and 28.04 mm, being the group of individuals with the smallest size, which corresponded to all the research time, especially in July, reaching only 24.48 mm (Table 2).

Table 2. Valvar length (mm) of the individuals found during May to July on the San José beach, San José district, Chiclayo province, 2013.

<i>Donax obesulus</i>	Beach San José		
	May	June	July
Number	271	278	283
Minimum	14.75	11.78	13.9
Maximum	41.3	32.56	33.99
Average	28.04	26.79	24.48
Standard deviation	3.48	3.72	4.11
Coefficient of variation	12.40	13.89	16.77

The largest number of individuals was recorded in El Gigante beach during all the research work with a total number of 472 specimens, being in the first month of sampling the month with the highest number of individuals (Figure 4). The minimum size found was 15.03 mm in June and the maximum size was 36.94 mm in the same month. their average ranged from 28.69 mm to 30.76 mm, representing the largest group of individuals from the three areas sampled during the study time (Table 3).

Table 3. Valvar length (mm) of the individuals found from May to July at El Gigante beach, Mórrope district, Lambayeque province, 2013.

<i>Donax obesulus</i>	Beach El Gigante		
	May	June	July
Number	472	318	290
Minimum	18.55	15.03	18.64
Maximum	35	36.94	35.52
Average	28.69	30.76	30.44
Standard deviation	2,22	3.48	3.10
Coefficient of variation	7.74	11.31	10.18

It can be corroborated that there was a correlation of R = 0.9629 between the weight and lengths of *Donax obesulus* sampled from May to July, observing a negative allometric growth of b = 2.5674 as observed in Figure 5.

On the San José beach, it was found that the copper bioaccumulation practically tripled, going from 3.7 µg g⁻¹ to 10.1 µg g⁻¹ in wet weight during the duration of the investigation, as presented in table 4. The lowest concentration of this element was 2.0 µg g⁻¹ in wet weight at El Gigante beach in May, with this sampling area presenting the lowest copper concentrations during the investigation period. Lead

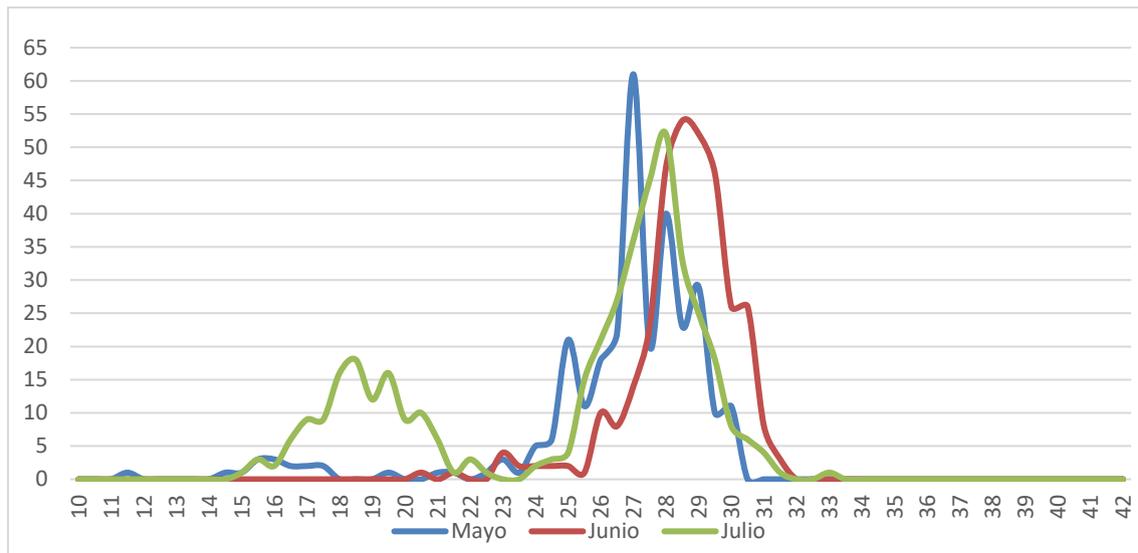


Figure 2. Distribution of length frequencies (valve length) of *D. obesulus* during May to July at Lagunas beach, Lagunas district, Chiclayo province, 2013.

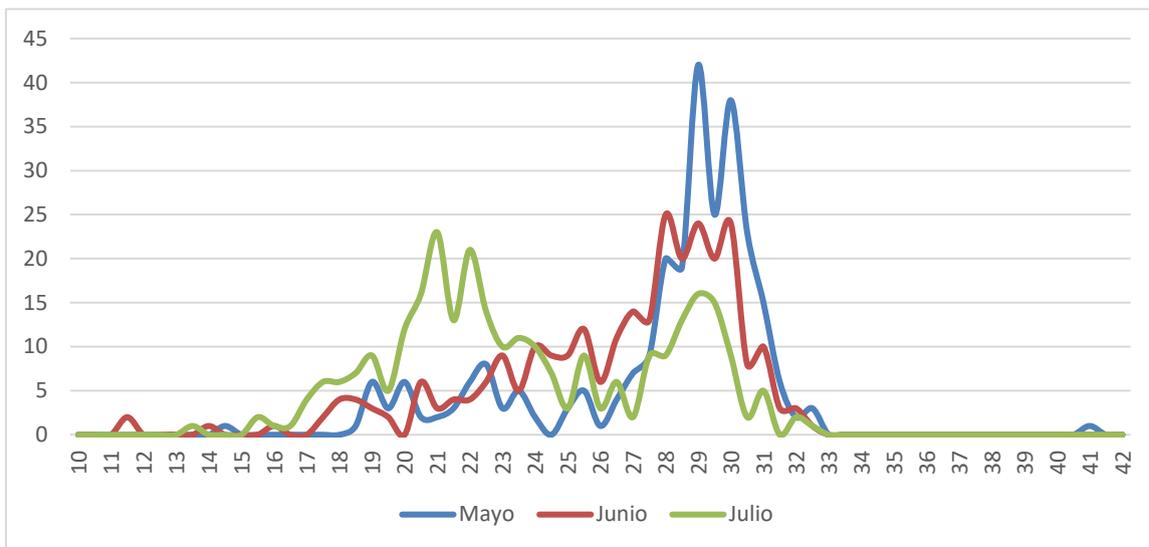


Figure 3. Distribution of length frequencies (valve length) of *D. obesulus* from May to July at San José beach, San José district, Chiclayo province, 2013.

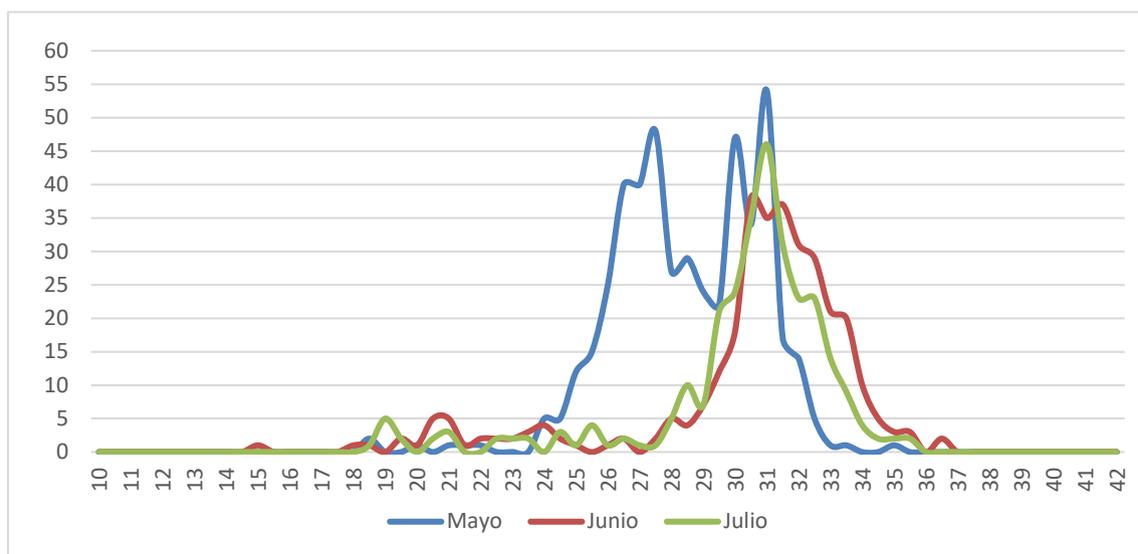


Figure 4. Distribution of length frequencies (valve length) of *D. obesulus* from May to July at Playa el Gigante, Mórrope district, Chiclayo province, 2013.

concentrations in the El Gigante, San José, and Lagunas beaches during the time investigated always showed the same concentration of 0.3 µg g⁻¹ wet weight. The analyzes carried out on the edible tissues of *D. obesus* showed that only in May (table 4) and only for the El Gigante beach in Morrope, the cadmium concentration was determined (0.4 µg g⁻¹ in wet weight) although in a concentration not very dissimilar to the other sampling areas, for which reason the presence is generally rejected. In the other months, the concentrations were even less than 0.2 µg g⁻¹ in wet weight, for all the months and all the sampling areas, with no significant differences. (P> 0.05) (Figure 5).

Table 4. Concentration levels of heavy metals expressed in µg g⁻¹ in wet weight, in edible tissue of *D. obesus* from the Lagunas, San José, and El Gigante beaches, on the Lambayeque Region coast, May to July 2013. Desviaciones de las replicas

Copper	Mayo	Junio	Julio
Lagunas	2.4	4.1	6.7
San José	3.7	5.4	10.1
El gigante	2.0	2.1	2.8
Lead			
Lagunas	0.3	0.3	0.3
San José	0.3	0.3	0.3
El gigante	0.3	0.3	0.3
Cadmium			
Lagunas	<0.2	<0.2	<0.2
San José	<0.2	<0.2	<0.2
El gigante	0.4	<0.2	<0.2

Discussion

In most organisms, copper is part of its proximal composition because it is an essential element, bioaccumulating to form certain types of proteins and enzymes. (Rainbow, 2002). This work reported a bioaccumulative trend, both for the different areas and sampling time. As Baršytė exposed, (1999) the large accumulation of copper in *Viviparus viviparus* from the Vilhemo channel, near Drevema, can be explained by its feeding mechanism.

Research of Usero et al., (2005), in the study of the concentration of heavy metals in mollusks on the south Atlantic coast of Spain, shows that copper was the most abundant metal in *Donax trunculus* with a concentration of 383 mg Kg⁻¹ dry weight and in *Chamelea gallina* with 90 mg Kg⁻¹ dry weight, following this study, the sampled areas are located near the mouth of the Huelva estuary, where the Tinto river and the Odiel river flow and both are highly contaminated by metals.

In the same sense, the bioconcentrations of lead found in the Patagonian region of Argentina by Gil et al., (2006) in *Mytilus edulis* and *Aulacomya atra* was from 1.82 to 8.07 µg g⁻¹ dry weight and from 1.64 to 6.85 µg g⁻¹ dry weight respectively, considering that such results are typical of low impact areas anthropic, slightly exceeding the maximum allowed for human consumption according to the European Economic Community (2001).

In similarity, Singh et al., (2012) found that in the tissues of *Donax faba* from the Panambur beach, the averages of the concentrations of copper and lead were 21.84 and 4.22 µg g⁻¹ dry weight respectively, and these means show that these concentrations are below of the maximum limits allowed.

Gil et al., (2006) found cadmium levels from 3.89 to 6.74 µg g⁻¹ dry

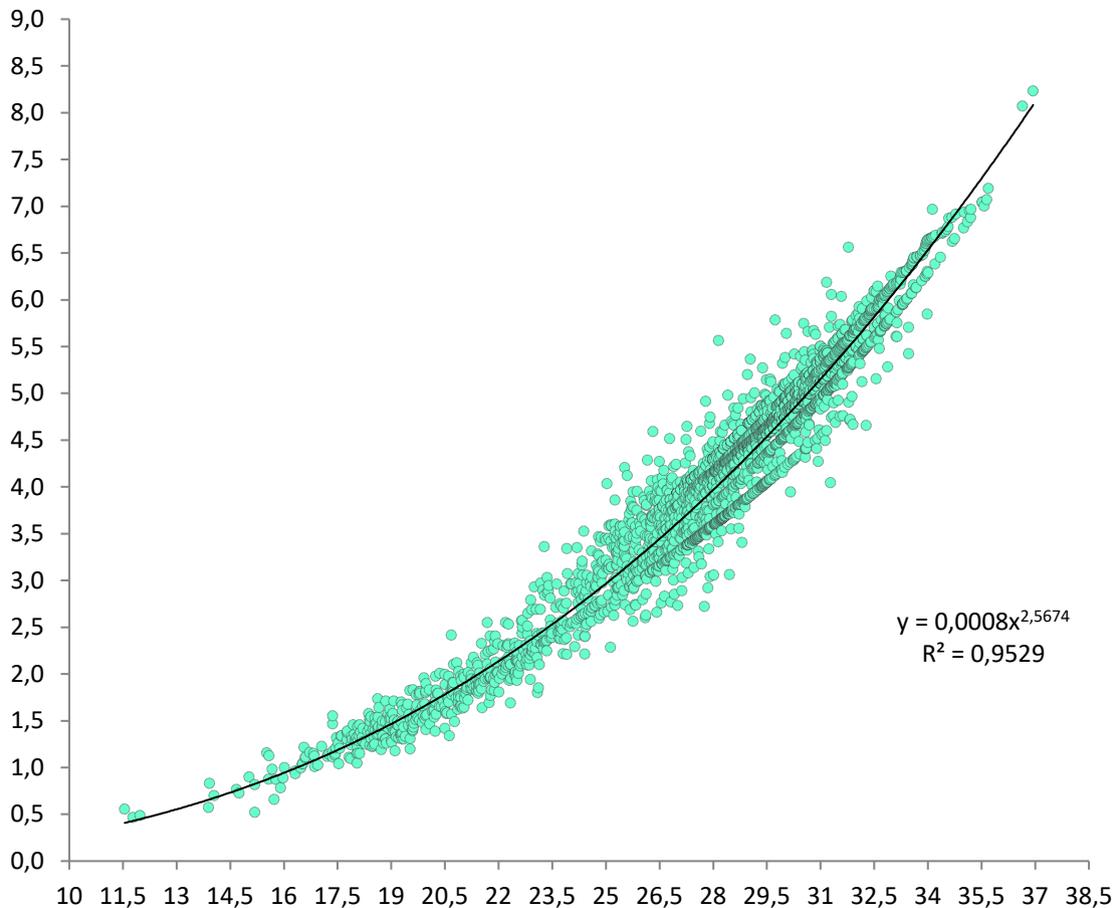


Figure 5. Weight-length relationship of *Donax obesus* from May to July at Lagunas, San José, and El Gigante beaches, Lambayeque Region, 2013.

weight in *Mytilus edulis* and *Aulacomya sp.* at 2.36 to 6.74 µg g⁻¹ dry weight whose causes of origin are not from industrial or mineral sources in the vicinity from where the samples were taken, assuming that the origin is natural in a similar way as the concentrations found in *Donax obesulus* from the coast of the Lambayeque region.

Hamed and Emara (2006) in their study found that the maximum cadmium value (2.71 µg g⁻¹ dry weight) was recorded during the winter of 2003 in station 1 (Adabiya, first of the 7 sampling areas, northwest of the Gulf of Suez) while its minimum value (1.86 µg g⁻¹ ps) was found in the summer in the same sampling area the same year, attributing the origin to tributaries from industrial complexes and refineries, fertilizer and electricity companies, sewage and sewage from the city of Suez and finally from boats that use the Suez Canal for their journey.

On the contrary, it is the case of Liang et al., (2004) who reported that for cadmium in the Penglai and Qinhuangdao sampling areas (2.55 and 3.29 µg g⁻¹ ph respectively) in *Crassostrea taliwhanensis*, the concentrations of this metal were above the restrictive values established by the WHO (1987), showing that these bivalves do constitute a risk to human health. The same happened in the analyzes of Fang et al., (2003) in bivalves from the markets of the Pearl River Delta region in China, who exceeded the permitted levels of cadmium established by the PHMSO (1988), constituting a latent risk to human health.

Although the cadmium levels found in this study in *Donax obesulus* are small and leaflet analyzes were not performed, Huanxin et al. (2000) show that it bioaccumulates very well in both soft tissues and leaflets. This fact caught the researchers' attention, given that cadmium is not an essential trace element for any organism, and finding a concentration of the order of 42 times more in the leaflets than in the sediment, led them to deduce that this element is taken as a substitute for calcium, which is an essential element for living organisms, since they have a geochemical similarity, especially in the ionic radius (cf. Ca 9.7 and Cd 9.8 nm).

Through the biometric samples that were carried out, it was found that the largest number of *D. obesulus* individuals corresponded to sizes greater than the minimum size for capture established by the Ministry of Production of Peru (22 mm), which sizes varied between 22 mm up to 41.3 mm and is the highest average found of 30.76 mm and that was chosen to be part of the sub-sample for the toxicological test, and despite corresponding to a size that involves individuals whose age would exceed the average age of life of this species, the bioaccumulation of heavy metals they possess, did not represent alarming levels, as they are low, which do not put at risk neither the consumers of *D. obesulus* nor the normal growth, development and/or recruitment.

For future studies on the quality and environmental situation of the Lambayeque coast, the study time intervals should be extended so that other types of Spatio-temporal variations could be observed, and, if possible, concentration variations in the same organism intra-woven. Other species of mollusks that also form part of the diet of the inhabitants of the Lambayeque region, which have characteristics of bioaccumulative organisms, could also be used together to verify that bioaccumulation is registered not only in one organism but in all and to establish the capacity for use as a biomonitor.

As *D. obesulus* is a filtering bivalve and easily accessible for extraction and consumption, it is emerging as a suitable candidate for use as a biomonitor of medium and long-term contamination for the control and characterization of the environmental health status of the Lambayeque coast.

Conclusions

The copper concentration showed a cumulative trend throughout the investigation up to 10.1 µg g⁻¹ wet weight in July at El Gigante beach in Mórrope. They found levels of lead (> 0.3 µg g⁻¹ph) and cadmium

(> 0.2 µg g⁻¹ ph) correspond to meager levels present in the edible tissue of *D. obesulus*. The concentrations of copper, lead, and cadmium in the edible tissues do not exceed any of the Maximum Permissible Limits established by international government agencies and do not represent any problem from the health of diners.

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