Lead in the marine environment: Concentrations and effects on invertebrates

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Abstract (150 words)

Lead (Pb) is a non-essential metal naturally present in the environment and often complexed with other elements (e.g. copper, selenium, zinc). This metal has been used since ancient Egypt and its extraction has grown in the last centuries. It has been used until recently as a fuel additive and is currently used in the production of vehicle batteries, paint, and plumbing. Marine ecosystems are sinks of terrestrial contaminations; consequently, lead is detected in oceans and seas. Furthermore, lead is not biodegradable. It remains in soil, atmosphere, and water inducing multiple negative impacts on marine invertebrates (key species in trophic chain) disturbing ecological ecosystems. This review established our knowledge on lead accumulation and its effects on marine invertebrates (Annelida, Cnidaria, Crustacea, Echinodermata, and Mollusca). Lead may affect different stages of development from fertilization to larval development and can also lead to disturbance in reproduction and mortality. Furthermore, we discussed changes in the seawater chemistry due to Ocean Acidification, which can affect the solubility, speciation, and distribution of the lead, increasing potentially its toxicity to marine invertebrates.

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Keywords: Lead, metallic trace elements, contamination, marine invertebrates

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Introduction

Marine invertebrates are recognized as good bioindicators in ecotoxicology (Rainbow 2002; Chiarelli et al. 2014, 2019). Thus, various species in distinct subphyla or phyla (Annelida, Cnidaria, Crustacea, Echinodermata, and Mollusca) are commonly used in ecotoxicological investigations. For example, many studies focused on marine contamination use filter-feeding organisms (as mussel, *Mytilus galloprovincialis*, Azizi et al. 2018; and oyster, *Crassostrea gigas*, Liu et al. 2021), grazers (as gastropods, *Patella caerulea*, Aydm-Öhnen and Öztürk 2017), detritivores (as sea urchins, *Paracentrotus lividus*, Rouane-Hacene et al. 2018) and predators (as crabs, *Carcinus* spp, Leignel et al. 2014) to investigate the effects on key species in the trophic chain.

Metallic trace elements (MTE) are naturally present in the environment and their major sources are volcanic eruptions and rock erosion. Nevertheless, their extraction and use in industry lead to their massive release into the atmospheric, terrestrial and aquatic environments. MTE pollution has been a major concern since the industrial revolution because of increasing anthropogenic activities, making them the most studied pollutants (Ali et al. 2019). Among these MTE, lead (Pb) has been widely studied for its impacts on terrestrial fauna and humans (Celis et al. 2015; Gomot-De Vaufleury 2000; Assi et al. 2016). Nevertheless, its toxicity to aquatic organisms, and more specifically marine animals, remains little studied and its negative impacts on fauna are probably highly underestimated.

Lead (Atomic number : 82; atomic mass : 207.2; CAS number : 7439-92-1) is a bluish-grey metal that is naturally present in the Earth's crust (Carocci et al. 2015) at an average concentration of 20 mg/kg of soil and including 4 major isotopes: 204Pb, 205Pb, 207Pb et 208Pb. The last three forms result from the radioactive decay of thorium and two isotopes of uranium (Flora et al. 2006). Lead can either be organic, inorganic, or metallic and it is mainly found in the environment in the form of salts ($PbCO_3$, $Pb(NO_3)$, $PbSO_4$), hydroxylated ($Pb(OH)_2$) or ionized (Pb^{2+}). Lead speciation in the form Pb^{2+} is very important in determining the bioavailability and behaviour of this metal. This major form is mainly complexed with an organic or inorganic ligand. Complexation with an organic ligand can be related to cations such as Ca^{2+}, Mg^{2+}, and Zn^{2+} (Capodaglio et al. 1990; Carocci et al. 2015). The inorganic phase of lead is dominated by two ligands, which are chloride and carbonate, thus allowing the metal to form complexes (Woosley and Millero 2013):

$$\mathrm{Pb^{2+} + nCl^{-} \leftrightarrow PbCl_{\mathit{n}}^{2-\mathit{n}}\ ou\ Pb^{2+} +\ nCO_3^{2-} \leftrightarrow PbCO_3^{2-2\mathit{n}}}$$

[Where n is the number of chloride ions (between 1 and 3) or a carbonate ion]

The main lead-bearing mineral is galena (PbS) but we also distinguish other ores such as anglesite ($PbSO_4$) and cerussite ($PbCO_3$) (Flora et al. 2006). Therefore, lead has been widely used since Ancient Egypt era (5000-3000 BC) (Nriagu 1983) because of its own melting point, its malleability, and its corrosion resistance. For example, it has been used for the manufacture of kitchen utensils and decorative items, in plumbing or the tableware factory (Flora et al. 2006). However, its use has dramatically increased since the 18th century with the implementation of the metal industry. Lead has been exploited for the manufacture of pipes, pigment, and as a biocide in antifouling paints but also for ceramics and building materials. Finally, lead has also been used as an additive in fuel (tetraethyl and tetramethyl), in some batteries, electrical components, and sometimes in drugs and cosmetics (Flora et al. 2006). The largest increase of lead release in the environment took place between 1950 and 2000, corresponding to the use of tetralethyllead as an additive in gasoline. This increasing use of lead generates an increasing accumulation of this nonbiodegradable metal in soils, air and drinking water, making it a major concern for organism health. Because of the persistence and toxicity of lead to humans and the environment, national and international organizations have imposed strict regulations over the past 20 years on the use of lead in the industry (Annibaldi et al. 2009), including leaded gasoline, lead paint, lead welding in tin cans, or pesticides based on lead arsenate ($Pb_3(AsO_4)_2$). An example of these regulations is the reduction of leaded gasoline, which began in the 70's in the US and the 90's in Europe (Annibaldi et al. 2009). It was completely phased out in 2000 in France and in 2002 in Spain and Italy (Annibaldi et al. 2009). Another example concerns child's products in the US, where an act set up in 2008 defined the lead limit for all of children's products at 100 parts per million, unless it is not technologically feasible, in this case, the lead limit is 300 parts per million (Consumer Product Safety, Improvement Act of 2008). These regulations caused the replacement of lead mainly by plastic for cable sheaths by tin for welding of drinking water system. Steel and zinc are also usual substitutes of lead (Brown et al. 2019). Furthemore, these various regulations have allowed a drastic reduction of lead emissions in the environment (Carocci et al. 2015).

However, despite the efforts made to reduce lead emissions, they remain present. Nowadays, the primary sources of lead production include mines and ore smelting, while secondary sources are recycled materials such as batteries and lead pipes (Flora et al. 2006). We also still find lead sources near incinerators and foundries, some paints for military or industrial use (Carocci et al. 2015; Flora et al. 2006). Moreover, total atmospheric emissions of lead vapour have increased in the past 15 years due to the increased demand for electrical energy and the uses of coal and natural gas (Carocci et al. 2015). A report published by the British Geological Survey in 2020 informs on global mineral production between 2014 and 2018 and indicates that China was the first country producer of lead from 2014 to 2018 (**Table 1,** https://www.bgs.ac.uk/mineralsuk/statistics/worldStatistics.html). However, between 2014 and 2018, China's mine lead production decreased by 12.6%. As for the USA and Russia, respectively in second and fifth place behind China, they produced almost 8.7 and 10.6 times less lead than China.

I/ Lead contamination in marine environments

Lead enters the marine environment *via* precipitation, dry deposition, soil leaching, municipal and industrial waste discharges as well as runoff from fallout deposits from streets and surfaces (Carocci et al. 2015). Industrial activities represent an important source of lead in seawater. For instance, anti-fouling paints used on boats to prevent the growth of organisms on them, are a significant emission source of lead in the marine environment (Bhattacharyya et al. 2013). Once the lead is in the ocean, it undergoes long-range transport through ocean currents. For instance, Celis et al. (2015) demonstrated that it could reach polar areas.

The bioavailability and toxicity of lead in water depend on the pH, water hardness, organic material concentration, and the presence of other metals (Branica and Konrad 1977). In seawater, lead is in its ionic forms or complexed with organic ligands and this metal can precipitate when its solubility limit is exceeded (Flora et al. 2006; Angel et al. 2016). Lead is mostly precipitated in lead acetate [$Pb(C_2H_3O_2)_2$] or cerussite ($PbCO_3$). In aquatic systems, sediments adsorb a very large part of the lead, while only a minor fraction remains dissolved in water, due to the complexation of the inorganic phase of lead by ligands. Furthermore, organic compounds like tetraethyllead ($C_8H_{20}Pb$, colorless oily liquid) or tetramethyllead ($C_4H_{12}Pb$) are bioavailable to organisms (Flora et al. 2006). Moreover, Pb can complex with dissolved organic matter (DOM) and it has been suggested that the nature of the DOM could have different effects on Pb bioavailability and toxicity (Sánchez-Marín et al. 2011; Sánchez-Marín and Beiras 2012). For example, according to Sánchez-Marín et al. (2007), Pb complexed with humic acids induces an increase of Pb uptake for *Mytilus edulis* gills and an increase of Pb toxicity for the embryos of *Paracentrotus lividus*. However, the effects of humic acids on Pb toxicity would be more important than the effects of fulvic acids or DOM extracted from the Suwannee River also tested on *P. lividus* embryos concerning toxicity (Sánchez-Marín and Beiras 2012) and *Mytilus edulis* concerning gills uptake (Sánchez-Marín et al. 2011). The results of these studies suggest that the effects of DOM on Pb toxicity and bioavailability depend on DOM type and more precisely on physicochemical properties of DOM types (Sánchez-Marín et al. 2011; Sánchez-Marín and Beiras 2012).

Many studies have focused on the quantification of dissolved forms of lead concentrations in offshore and coastal waters (**Table 2**) (Patterson et al. 1976; Paulson and Feely 1985; Fowler 1990). Concentrations of these dissolved and particulate forms of lead in coastal waters are very heterogeneous ranging from 0.001176 µg/L (Beagle Channel, Patagonia, Argentina; Conti et al. 2012) to 1015 µg/L (Gulf of Gabes, Tunisia; Drira et al. 2017). The Gulf of Gabes, situated in the Southeast of Tunisia, has been reported heavily polluted because of the increase in urbanization, industrialization, tourism, and intensive fishing activity (Drira et al. 2017). Lead concentrations in offshore waters are ranging from 0.0035 (American Samoa, South Pacific Ocean) to 0.150 µg/L (Western Mediterranean Sea; Copin-Montegut et al. 1986). The large discrepancy between coastal and offshore levels in lead may be related to a dilution gradient in lead sources and emissions from terrestrial to offshore environments (Davis 1993; Espejo et al. 2019).

In order to limit the negative effects of lead on organisms in the marine environment, water quality guidelines indicating the concentration thresholds not to be exceeded for a given pollutant in a given compartment have been developed in different countries, such as Australia and New Zealand. For lead, the recommended value in marine waters to protect 95% of the species present in the study area is 4.4 µg/L (https://www.waterquality.gov.au/anz-guidelines/guideline-values/default/water-quality-toxicants/search). This threshold is established from 25 data of LOEC (Low Observed Effect Concentration), NOEC (No Observed Effect Concentration) and EC_{50} (Half-maximal Effective Concentration) divided into the following taxonomic groups: algae, annelids, crustaceans, and molluscs. However, this water quality guideline (https://www.waterquality.gov.au/anz-guidelines/guideline-values/default/water-quality-toxicants/search) has been established using tropical and temperate marine species and thus could not be valid for all marine organisms.

II/ Lead concentrations in marine invertebrates

As said earlier, when lead particles are present in seawater, a fraction is released in soluble form which can enter the marine food web. Filter feeders or scavengers are prone to bioaccumulate important levels of lead (**Table 3**). Nevertheless, for the same species, differences of several orders of magnitude can be observed among studies (**Table 3**); that may be related to contamination levels in surrounding waters as well as biological factors (e.g. age, diet, metabolism, sex) and abiotic parameters (temperature, salinity, pH…). Pb accumulation seems preferential in kidney and muscle (Jakimska et al. (2011). Noël et al. (2010) showed lead concentration values ranging from 0.040 to 0.247 µg/g of dry weight for sea urchin *Paracentrotus lividus* collected in the same location. And finally, lead concentrations can fluctuate importantly among life stages, such as in the horseshoe crab *Limulus polyphemus* with levels ranged from 0.02 to 0.59 µg/g of dry weight from the stage egg to embryo respectively (Bakker et al. 2017). The Pb content in invertebrate tissues depend of seasons and anthropogenic activities, as showed by Scudiero et al. (2014) in mussels from Campania coast (Italy).

The potentially high levels of lead bioaccumulation in marine invertebrates present a potential ecotoxicological risk for human food consumption (Sioen et al. 2008; Cabral-Oliveira et al. 2015). In Europe, the maximum concentrations of lead allowed in commercialized bivalves, such as mussels or oysters, is 1.5 mg/kg of fresh weight by the Commission Regulation (EC) no. 1881/2006 (European Commission 2006; Cabral-Oliveira et al. 2015). However, this limit is not always respected as shown by Cabral-Oliveira et al. (2015) who assessed concentrations of lead of 4.6 mg/kg of fresh weight in mussel *Mytilus galloprovincialis* on the Portuguese coast. In 2015, the World Health Organization (WHO) established the level of tolerable weekly intake for lead to 25 µg/kg of body weight. Abdallah (2013) showed that the estimated weekly intake of lead for bivalve *Ruditapes decussatus* along the Alexandria coast of the Mediterranean Sea exceeds the level indicated by the WHO. These examples show the importance of evaluating levels of lead in the environment and in edible seafood, in order to limit chronic exposure to metals for human populations.

III/ Effects of lead on marine invertebrates

As mentioned before, lead does not possess any biological function in organisms (vegetal or animal). In contrast, it is known to induce damage to the central nervous system, kidneys and hematopoietic system in vertebrates (Flora et al. 2006). Its toxicity mechanism is probably the most studied among the MTE and its effects have already been reviewed by Flora et al. (2006, 2012). Nevertheless, although studied in superior organisms, it seems that no reviews have been written about lead effects on different groups of marine invertebrates. The acute effects (Effective Concentration at 50% [EC_{50}]) of lead have mainly been measured in bivalves and sea urchins (**Table 4**). Indeed, lead contamination has negative consequences on various phyla and these effects are also diversified even though studies mainly focused on the effects of this metal on early life stages. If we compare the EC_{50} in various marine organisms (**Table 4**) and lead environmental coastal concentrations (**Table 2**), we can see that EC_{50} values for molluscs and echinoderms (except for Sea urchin *Arbacia punctulata*) are lower than the highest concentration found in the literature (1015 µg/L, Gulf of Gabes, Tunisia; Drira et al. 2017). Therefore, we can hypothesize that these marine organisms and potentially others could be affected by current lead environmental concentrations in coastal areas. Thus, despite the efforts made to decrease lead emissions during the past decades, this metal is still representing an ecotoxicological risk today for marine ecosystems.

III.1. Effects on Echinodermata

Echinoderms represent a relevant model system for investigating environmental pollution (Chiarelli et al. 2019). It has been evidenced that different Sea urchin species are able to survive in polluted environments and accumulate high levels of metals in their tissues via physiological uptake of nutrients (Burić et al. 2015; Chiarelli et al. 2019) . Heavy metals mainly affect fertilization, skeletogenesis, gut elongation, growth or tolerance to temperature stress (Kobayashi and Okamura 2004; Roccheri et al. 2004; Anselmo et al. 2011; Burić et al. 2015). The larval skeleton is highly sensitive to environmental stressors and so is often used as a marker of metal pollution in ecotoxicology (Matranga et al. 2013; Martino et al. 2018). Therefore, Sea urchins provide a valuable and attractive model to evaluate the toxicity of pollutants (Chiarelli et al. 2019). The Sea urchin *Paracentrotus lividus* is an echinoderm species used in toxicological studies concerning trace metals. Furthermore, its early stages are of interest because they present qualities such as an important amount of gametes or external fertilization (Chiarelli et al. 2014). Geraci et al. (2004) found that short-term exposure of *P. lividus* embryos to lead causes stress to this organism translated by the increase of *Hsp70/72* expression, a marker of cellular stress. However, a longer exposure, which continued until the pluteus stage, induces the decrease of these protein levels(HSP70) from the blastula stage. Also, this study showed that an exposure to lead during development induced the decrease of the protein HSC70 at the blastula and gastrula stages, followed by an increase of its level at the pluteus stage (Geraci et al. 2004). Geraci et al. (2004) also observed that exposure of *P. lividus* embryos to Pb caused irregular morphology at the gastrula and pluteus stages. Furthermore, Fernández and Beiras (2001) reported that lead is responsible for disrupting the embryo development of Sea urchin *Paracentrotus lividus*, and particularly the growth of its larvae, with a negative correlation between lead concentrations and development (inhibition and arrest). For example, at 500 µg/L, the embryo reaches the stage of pluteus larvae, while at 1000 µg/L its embryogenesis stops at the gastrula stage. Moreover, at 250 µg/L, the length of the larvae decreased by 5.3% and 13% at 500 µg/L. This work then provided a 48h-EC_{50} value of 509 µg/L for this species (Fernández and Beiras 2001). Another study found that lead caused abnormal development of *Evechinus chloroticus* larvae, with skeletal anomalies appearing at concentrations of 10 and 20 µg/L of lead, as well as stunted growth and even growth arrest at the pluteus larvae from 50 µg/L of lead (7% growth inhibition) (Rouchon and Phillips 2016). Effects of lead on growth of Echinodermata have also been assessed on *A. punctulata* embryos with a 4h-EC_{50} value from 32 to 500 µg/L measured by Nacci et al. (1986), and also on *S. purpuratus* larvae with a 72h-EC50 of 74 µg/L (Nadella et al. 2013). The negative effects of lead on Echinodermata do not only concern the growth of these organisms but also their reproduction processes. Indeed, Warnau and Pagano (1994) focused on the effects of $PbCl_2$ on the sperm fertilization of *Paracentrotus lividus* and on offspring quality. No significant effect was observed on the fertilization rate; nevertheless, offspring quality seems to be weathered by lead concentrations. We can observe from **Table 4** that, in Echinodermata, the EC_{50} values range concerning the development of larvae of *E. chloriticus* and *S. purpuratus* for 72h overlap, showing that the sensitivity of these species to lead wouldn't be very different. However, **Table 4** also shows that EC_{50} values of all Echinodermata species presented are below the highest lead concentration measured in coastal environments (1015 µg/L; Table 2), which could indicate that these species could be affected by current lead concentrations and particularly at their early life stages.

III.2. Effects on Mollusca

As for the effects in Echinodermata species, the impacts of lead on molluscs have been assessed on various species, also focusing on larval development. Beiras and Albentosa (2004) demonstrated for clam *Meretrix meretrix*, that embryos reached the D-shaped form ("normal form") when they were exposed to 197 µg/L lead. Meanwhile, only gastrula and blastula stages were maintained after exposition to 1016 µg/L lead. The same authors reported that mussel *Mytillus galloprovincialis* seemed to be the most sensitive among all bivalve species showing the inhibition of embryogenesis at an EC_{50} value of 221 µg/L. Fathallah et al. (2013) also studied the inhibition of embryogenesis on *Ruditapes decussatus*, and found that embryogenesis was affected at 256 µg/L with a 50% decrease in the number of D-shaped larvae. Similarly, Xie et al. (2017) observed that a lead concentration of 8948.4 µg/L induced abnormalities in 88.7% larvae (no D-shape) in oyster *Crassostera gigas*. Indeed, lead's effects are mainly observed at the beginning of embryogenesis in the animals (Wang et al. 2009). The most sensitive larvae species to lead seems to be *Mytilus trossolu*s and the less sensitive seems to be *C. gigas* (**Table 4**). Furthermore, all of the molluscan species presented in **Table 4** have an EC_{50} value lower than the highest current concentration of lead measured in coastal environments, meaning that all of these species, at least at their early life stages, could already be affected by these lead concentrations.

III.3. Effects on Annelida and Cnidaria

The polychaete annelid *Hydroides elegans* has its fertilization rate reduced by 71.2% in the presence of 100 µg/L of lead after 20 min of exposure (Gopalakrishnan et al. 2008). Furthermore, tentacle retraction of the cnidarian *Aiptasia pulchella* in presence of $PbCO_3$ has been studied as a sublethal parameter to estimate EC_{50} values for lead (Howe et al. 2014, **Table 4**). At 96h, a massive loss of its symbiont, *Symbiodinium pulchrorum* (Dinoflagellate), has been observed on 80 to 90% of anemone exposed to lead concentration of 688 000 µg/L.

III.4. Effects on Crustacea

Ecotoxicological studies showing the LC_{50} Pb for various marine organisms revealed that the metal toxicity depends on the lead form used in the experimentation (Lead nitrate, tetramethyllead, tetraethyllead) (**Table 5**). For example, tetraethyllead was more toxic to the shrimp *Crangon crangon* (96h-LC_{50} of 100 µg/L) than tetramethyllead (96h-LC_{50} of 270 µg/L) (Maddock and Taylor 1980). Similarly, for the same form of lead used, there are species-specific differences in the LC_{50} values. Some of the species present LC_{50} Pb values below the higher lead concentration measured in the coastal environment, such as annelid *Hydroides elegans*, arthropod *Crangon crangon*, and mollusc *Mytilus edulis*, showing that they could be affected by current lead environmental concentrations (**Table 5**).

IV/ Conclusions and future perspectives

Lead is a MTE found in some coastal areas but also in pelagic zones. This metal comes from natural (volcanic eruption, soil erosion) and anthropogenic (paint, fuel additives ...) sources. The lead entering the marine environment from these sources can then be accumulated in various marine invertebrates, which can then be used as bioindicators to monitor the evolution of lead contamination in several regions of the world. In addition to its accumulation in marine invertebrates tissues, lead causes also several negative effects on these organisms. These effects have been widely investigated on terrestrial animals but the bibliography concerning marine invertebrates is more restricted and needs to be extended to more marine invertebrate species to draw a more accurate picture of the impacts of this metal on these organisms. However, studies available show that, in marine invertebrates, lead can affect early life stages by inducing growth (inhibition or arrest of growth, morphological anomalies), and reproduction disruption (offspring quality, fertilization rate), which could lead to negative effects on populations and communities. Therefore, the follow-up of lead contamination levels in marine environments is of great importance to better assess the threats to the survival and vitality of marine invertebrates and more globally of marine organisms.

Since the beginning of the industrial revolution, human industrial activities have become major problem affecting marine ecosystems by multiple processes such as climate change and pollution. Factors associated with global warming mainly involve temperature and ocean acidification (OA), which could considerably modulate the impacts of pollution on coastal and estuarine ecosystems (Ivanina and Sokolova 2015). Fossil fuel and biomass combustion as well as cement production result in greater CO_2 assimilation by ocean (Gattuso et al. 1998). High levels of dissolved CO_2 in oceans induce the increase in CO_2 partial pressure (P_{CO2}) leading to pH and ocean carbonate chemistry changes (Ivanina and Sokolova 2015). When carbonate dissolved in the seawater, it reacts with water molecules to transform into carbonic acid (H_2CO_3). Then, carbonic acid dissociates into hydrogen and bicarbonate (HCO_3^-), which leads to a decrease in pH and carbonate ion concentration (CO_3^{2-}) (Gazeau et al. 2011).

In marine environments, dissolved metal levels are generally low because of the low solubility of the MTE in seawater and their adsorption in sediments. Despite this fact, changes in the seawater chemistry due to OA can affect the solubility, speciation, and distribution of the MTE in water and sediments, affecting potentially their toxicity to marine organisms (Ivanina and Sokolova 2015). Indeed, a metal is present in various forms in the marine environment and these various forms have a different availability for organisms. The acidification can, therefore, influence interactions between metals and organisms in two ways. In fact, the decrease in pH modifies the metal form occurrence and could make trace metals to be more toxic (Han et al. 2013). Metals complex with organic and inorganic ligands and as the pH decreases, these metals tend to dissociate from the complexes resulting in increased concentrations of free ions, which become therefore more bioavailable and toxic to organisms (Campbell et al. 1985). Concerning effects on the inorganic speciation, metals forming strong complexes with carbonate and chlorine, such as lead, could, therefore, be affected by the decrease in seawater pH (Millero et al. 2009). The scenario of the pH decrease according to Caldeira and Wickett (2003) will lead to a seawater pH, which could reach 7.7 in 2100 and 7.4 in 2250. If these predictions prove to be correct, as pH decreases, free forms of lead will increase by 10% as well as its complexation with chlorine leading to a 15% increase in the forms $PbCl^+$, $PbCl_2$, and $PbCl^-_3$ (Millero et al. 2009). Only a few studies have studied the effects of decreasing pH on lead toxicity. For instance, Han et al. (2013) studied the effects of pH (6.2, 7.7, and 8.2) on heavy metal (Cd, Cu, and Pb) toxicity in mussel *Mytilus edulis*. For these metals, a lower pH leads to a higher mortality rate in this mussel. Furthermore, at pH 6.2 and during a lead pre-treatment, a decrease in the synthesis of metallothioneins has been noticed (Han et al. 2013). Other studies on cadmium (Shi et al. 2016) and copper (Ivanina and Sokolova 2015) showed that a lower pH increases the solubility of these two metals. Therefore, we can hypothesize that lead could behave the same way and become more available to the marine organisms. Thus, OA could worsen the toxicity of MTE such as lead for marine organisms, in semi-enclosed areas (like bays or lakes) where the effect would be accentuated. However, more studies need to be done to assess whether or not the toxicity of lead in the marine environment would be greater with OA. It will be interesting to develop an Adverse Outcome Pathways Framework to collect mechanistic knowledge on synergic effects of OA and lead accumulation on different levels of biological organization in marine ecosystem. Thus, a global investigation including the estimation of lead bioaccumulation in tissues and biomagnification from photosynthetic producers (as diatoms), filter-feeding organisms to predators and scavengers would allow to understand the additive or synergic effects of OA (distinct pH tested) and lead on trophic network. Epigenetic modifications, biomarkers expression (transcriptomic and RT-qPCR), and biochemical responses carry out on distinct species models (producers, filter-feeding organisms, predators, and scavengers) could be informative to detect the incidence of OA and lead fluctuation on the integrity of the trophic chain. Therefore, ecotoxicogenomics (allelic selection...) and Genome Wide Assocation (SNP detection), and Ecotoxicoproteomic analysis (identification of metabolomic pathway alteration) could be very interesting to detect precisely the cellular disturbances.

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Table 1. Lead production (Ton/year) in different countries in 2014 and 2018

Sources of lead	Country	2014	2018	References
Lead mining				
	China	2 608 619	2 280 000	https://www.bgs.ac.uk/mineralsuk/statistics/worldStatistics.html
	USA	378 000	260 000	
	Peru	277 294	289 195	
	Mexico	250 462	230 869	
	Russia	196 000	215 000	
Refined lead production				
	China	4 704 000	5 112 850	https://www.bgs.ac.uk/mineralsuk/statistics/worldStatistics.html
	USA	1 020 000	1 300 000	
	Republic of Korea	634 700	801 000	
	India	475 000	620 000	
	Germany	380 000	315 000	

Table 2. Lead concentrations (µg/L) in offshore and coastal waters reported in the world before 2000.

Note: numbers indicated are average and range (in square brackets) and the symbol ± indicates the standard deviation

Regions	Lead concentrations (µg/L)	References
Offshore waters		
Baltic Sea	0.016 [0.0115; 0.0205]	Danielsson and Westerlund 1984
	0.041-0.083	Kremling 1987
	0.050	Brügmann 1988
North Sea	0.052	Kremling 1987
	0.031	Balls 1985b
Mediterranean Sea	0.030-0.150	Copin-Montegut et al. 1986b
Northwestern Atlantic Ocean	0.033	Schaule et Patterson 1983
	0.046	Nurnberg et al. 1983
	0.026 [0.022;0.030]	Boyle et al. 1986
Northeastern Atlantic Ocean	0.033	Copin-Montegut et al. 1986a, b
Arctic Ocean	0.0148 [0.0113;0.0183]	Mart et al. 1984
Indian Ocean	0.030	Danielsson 1980
Northeastern Pacific Ocean	0.014	Schaule and Patterson 1983
	0.005-0.015	Schaule and Patterson 1981
South western Pacific Ocean	0.0046 [0.0045;0.0047]	Flegal and Patterson 1983
South eastern Pacific Ocean	0.016	Nurnberg et al. 1983
Coastal waters		
Fjord Framvaren, Norway	0.073	Haraldsson and Westerlund 1988
Kattegat/Skagerrak, North Sea	0.050	Magnusson and Westerlund 1983
England	[0.030-0.265]	Balls 1985a
Rhône delta, France	0.077	Huynh Ngoc et al. 1988
Corsica, France	0.048	Lafabrie et al. 2007
Cape Cod, USA	0.021	Boyle and Huested 1983
South atlantic bay, USA	0.025 [0.015;0.035]	Windom et al. 1985
Monterey Bay, California, USA	0.0076	Schaule and Patterson 1981
Puget Sound, Seattle, USA	0.020-0.110	Paulson and Feely 1985
Southern California Bight, USA	0.025-0.150	Patterson et al. 1976
Al-Khobar, Persian Gulf, Saudi Arabia	0.04 [0.017-0.095]	Alharbi et al. 2017
Al-Khafji, Persian Gulf, Saudi Arabia	0.28 [0.09-0.43]	Alharbi and El-Sorogy 2019
Gulf of Aqaba, Saudi Arabia	0.202 [0.020-0.450]	Al-Taani et al. 2014
Land of Fire, Beagle Channel, Patagonia, Argentina	0.001176 ± 0.001243	Conti et al. 2012
South Coast, Australia	20.64 [0.4-55]	Chakraborty and Owens 2013
Ship breaking area of Sitakund Upazilla, Chittagong, Bangladesh	113 [0.06-0.15]	Hasan et al. 2013
Bay of Bengal, Bangladesh	452 [96.4-694]	Hasan et al. 2016
San Jorge Gulf, Antofagasta, Northern Chile	0.04 [0.02-0.09]	Valdes et al. 2011
Bay of Jinzhou, China	0.61 [0.21-1.39]	Wang et al. 2012
Bay of Jinzhou, China	1.16	Wan et al. 2008
Estuary of the Pear River, China	1.61 [0.8-3.08]	Zhang et al. 2012
Estuary of the Yellow River, China	0,51	Tang et al. 2010
Bohai Sea, China	1.1 ± 0.4	Wang and Wang 2007
Laizhou Bay, Bohai Sea, China	0.88 ± 0.32 [0.56-2.07]	Lü et al. 2015
Bohai Bay, Tianjin, China	7.18 ± 2.57 [3.63-12.65]	Meng et al. 2008
Bohai Bay, China	1.63 [1.25-2.02]	Zhang et al. 2010
Guangdong, China	1.32 2.55	Zhang et al. 2016
Damietta Port, Egypt	2.44 [1.33-4.12]	El-Gohary et al. 2017

Gulf of Suez, Red Sea, Egypt	0.56-3.17	Mirnategh et al. 2018
Gulf of Suez, Red Sea, Egypt	1.84-2.57	El-Moselhy and Gabal 2004
Gulf of Aqaba, Red Sea, Egypt	0.36 [0.29-0.43] 0.36 [0.33-0.39]	Shriadah et al. 2004
Estuary of Vigo, Spain	0.98 [0.17-2.05]	Pérez-Lopez et al. 2003
Bay of Malaga, Andalusia, Spain	[0.20-680]	Alonso Castillo et al. 2013
Saronic Gulf, Anavissos, Greece	2.85 [0.37-6.51]	Ladakis et al. 2007
Gulf of Chabahar, Arabian Sea, Iran	2.224 [0.98-4.52]	Bazzi 2014
Persian Gulf, Qeshm Island, Iran	15.4 [12-20]	Karbassi et al. 2018
Bay of Aughinish, Ireland	0.021-0.038	Reis et al. 2017
Venice, Italy	2.59 [0.1-0.59]	Giusti and Zhang 2002
Porto Torres, Sardinia, Italy	0.075	Lafabrie et al. 2007
Livorno, Tuscany, Italy	0.038	Lafabrie et al. 2007
Pasir Gudang, Malaysia	362 ± 210	Mahat et al. 2018
Tuaran, Sabah, Malaysia	5.56 [3.32-10.5]	Tan et al. 2016
Indian Ocean, Mauritus	57 [10-247]	Daby 2006
Montenegro	1.963 [0-3.66]	Dukic et al. 2019
Baltic Sea, Poland	0.0165 [0.004-0.088]	Pempkowiak et al. 2000
Kranji and Pulau Tekong, Singapore	0.009-0.062	Cuong et al. 2008
Port of Kaohsiung, Taiwan	0.2-0.7	Lin et al. 2013
South Coast of the Gulf of Gabes, Tunisia	765 [569-1015]	Drira et al. 2017
North Coast of the Gulf of Gabes, Tunisia	638 [386-961]	Drira et al. 2017
Ghannouch, Gulf of Gabes, Tunisia	467 [383-567]	Drira et al. 2017
Rize, Black Sea, Turkey	8.8 [6-13]	Baltas et al. 2017
Red Sea, Yemen	0.057 ± 0.011 0.044-0.07	Al-Shiwafi et al. 2005

Table 3. Lead concentrations (µg/g of dry weight, whole body) in various marine invertebrate species.

Note: numbers indicated are average, range (in square brackets) and symbol ± indicated the standard deviation

Phylum	Species	Lead concentrations (µg/g of dry weight)	Reference
Arthropoda	*Cancer pagurus*	1.23 ± 1.57	Connan and Tack 2008
	Fenneropenaeus indicus	2.20-23.10	Bhattacharyya et al. 2013
	Fenneropenaeus indicus	0.008 ± 0.01	Salam et al. 2019
	Portunus pelagicus	0.015 ± 0.01	Salam et al. 2019
Echinodermata	*Paracentrotus lividus*	0.065 [0.040 ; 0.247]	Noël et al. 2011
Mollusca	*Buccinum undatum*	0.043 [0.040 ; 0.132]	Noël et al. 2011
	Heliocidaris tuberculata	0.040 [0.040 ; 0.040]	Noël et al. 2011
	Littorina littorea	0.063 [0.040 ; 0.140]	Noël et al. 2011
	Littorina littorea	10	Bryan et al. 1983
	Littorina obtusata	7.8	Bryan et al. 1983
	Littorina saxatilis	13	Bryan et al. 1983
	Littorina saxatilis	2.3-16.6	Daka et al. 2004
	Littorina saxatilis	0.468	Daka 2005
	Monodonta mutabilis	0.12-0.15	Cubadda et al. 2001
	Monodonta turbinata	0.13-0.47	Cubadda et al. 2001
	Monodonta turbinata	0.69 [0.39 ; 1.06]	Conti et al. 2007
	Murex brandaris	0.078 [0.040 ; 0.157]	Noël et al. 2011
	Mytilus edulis	2.53-5.97	Noël et al. 2011
	Mytilus edulis	16-309	Daka et al. 2004
	Nacella magellanica	3.09-5.91	Comoglio et al. 2011
	Nacella magellanica	0.13 ± 0.16	Conti et al. 2012
	Patella caerulea	1.02 [0.85; 1.30]	Conti et al. 2007
	Patella caerulea	1.39 [0.81; 1.97]	Conti et al. 2017
	Patella caerulea	0.10-1.42	Cubadda et al. 2001
	Patella lusitanica	0.14-0.71	Cubadda et al. 2001
	Patella sp	0.93-1.34	Connan et Tack 2008
Porifera	*Spheciospongia vagabunda*	0.26-2.55	Padovan et al. 2012

Table 4. Lead EC_{50} values (µg/L) in various marine invertebrate species

Note: numbers indicated are average and range (in square brackets)

Phylum	Species	Test endpoint	Test duration	EC_{50} (µg/L)	Reference
Cnidaria	*Aiptasia pulchella*	Retractation of tentacles	12h	1 740 [1 310 ;3 850]	Howe et al. 2014
Echinodermata	*Paracentrotus lividus*	Growth of larvae	48h	509	Fernández and Beiras 2001
	Arbacia punctulata	Growth of embryos	4h	32 500	Nacci et al. 1986
	Strongylocentrotus purpuratus	Development of larvae	72h	74 [50-101]	Nadella et al. 2013
	Evechinus chloroticus	Development of larvae	72h	52.2 [39.6-73]	Rouchon and Phillips 2016
Mollusca	*Crassostrea gigas*	Development of larvae	48h	380-550	Chapman and McPherson 1993
	Mytilus edulis	Development of embryos	48h	476	Martin et al. 1981
	Mytilus galloprovincialis	Development of larvae	48h	63 [36-94]	Nadella et al. 2013
	Mytilus trossolus	Development of larvae	48h	45 [22-72]	Nadella et al. 2013
	Mytilus trossolus	Development of larvae	48h	67 [37-100]	Nadella et al. 2013
	Crassostrea gigas	Development of larvae		660.3 [453.5-1062.4]	Xie et al. 2017
	Ruditapes decussatus	Embryogenesis inhibition	48h	156-312	Beiras and Albentosa 2004
	Mytilus galloprovincialis	Embryogenesis inhibition	48h	221 [58.9-346.3]	Beiras and Albentosa 2004
	Metrix metrix	Embryogenesis inhibition	24h	296 [246-501]	Wang et al. 2009
	Metrix metrix	Development of larvae	24h	199 [85-4 175]	Wang et al. 2009
	Ruditapes decussatus	Embryogenesis inhibition	24h	256.5 [145.4-385.7]	Fatthallah et al. 2013

Table 5. Lead LC$_{50}$ value (µg/L) in various marine invertebrate species.

Notes: numbers indicated are average and range (in square brackets).

Phylum	**Species**	**Lead form**	**Test duration**	**LC$_{50}$ (µg/L)**	**Reference**
Annelida	*Hydroides elegans*	PbCl$_2$	96h	946.05 [796.29; 1150.41]	Howe et al. 2014
Arthropoda	*Crangon crangon*	Tetramethyllead	96h	270 [330.2 ; 597.1]	Maddock and Taylor 1980
		Tetraethyllead	96h	100	
	Palaemon adspersus	Pb(NO$_3$)$_2$	96h	68 000 [55 000 ; 74 000]	Bat et al. 2001
Cnidaria	*Aiptasia pulchella*	PbCO$_3$	96h	8 050 [0 ; 11 700]	Howe et al. 2014
Mollusca	*Babylonia aerolata*	Pb(NO$_3$)$_2$	24h	22 210	Supanopas et al. 2005
			48h	14 860 [13 950 ; 15 760]	
			72h	12 440 [11 520 ; 13 250]	
			96h	10 500 [9 560 ; 11 170]	
	Mytilus edulis	Tetramethyllead	96h	110	Maddock et Taylor. 1980
		Tetraethyllead	96h	20	