

*Review*

## **A 50-year review on heavy metal pollution in the environment: Bivalves as bio-monitors**

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There has been a steady increase in the quantity and diversity of discharges that reach aquatic environment, both organic and inorganic, either synthesized or mobilized by man as a result of high population growth and development activities. This has led to the appearance of several types of pollution. Inland and coastal waters have been the most affected, since most major cities are located near water bodies. Monitoring programs and research for metals in the environment have become widely established because of concerns over accumulation and toxic effects, particularly in aquatic organisms. Quantifying metal concentrations in water and sediments may have limited value in this respect, particularly in circumstances in which it is difficult to define biologically relevant fractions. Therefore, the analysis of aquatic organisms have been used increasingly as a direct measure of the abundance and availability of metals and micro-pollutants in the environment and has led to the adoption of the bio-indicator (bio-monitor, sentinel organism) concept 50 years ago. All heavy metals (trace elements) are potentially toxic, even the essential ones if accumulated above (or below) levels needed by the organism. Temperature and salinity are the two major factors to be considered when considering abundance, availability, bioaccumulation and excretion of these metals in the aquatic environment. Almost all developed countries are found in the temperate region while the developing ones are located or/and found in the tropics where these factors are at the extreme. Hot spots for heavy metal pollution are found both in the developed and developing world.

**Key words:** Heavy metals, aquatic environment, bivalves, bio-indicator/monitors, seasonal variation, accumulation.

### **INTRODUCTION**

High population growth accompanied by intensive urbanization, increased industrial activity and higher exploitation of natural resources; as a result, there has been a steady increase in the quantity and diversity of

discharges that reach aquatic environments, both organic and inorganic, either synthesized or mobilized by man. This has led to the appearance of several types of pollution. Inland and coastal waters have been the most

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affected, since most major cities are located near them. Three main sources of pollution are recognized: urban development, industrial waste and agricultural use of pesticides. Urban pollution is mainly organic in origin and stems from domestic wastes, particularly sewage and garbage. Such wastes mainly originate from residential, commercial and recreational areas, offices and institutions. Most urban areas in developing countries still lack adequate waste collection; hence treatment and disposal facilities and wastes are therefore discharged untreated into natural water bodies. Industrial wastes are very diverse in nature and include chemicals, mine wastes, ferrous and non-ferrous metals and dust particles. Discharges may be either gas, liquid or solid that may finally reach the aquatic environment. These may include wastes from breweries, food processing plants, tanneries, textiles, paper pulp mills, manufacturing and use of paints, pesticides and fertilizers, mining operations, refining and burning of fossil fuels. The magnitude of waste varies with different industries. Extensive use of pesticides is considered necessary to increase production and improve food storage as well as vector control campaigns to curb endemic diseases such as malaria, onchocerciasis, schistosomiasis and trypanosomiasis. One of the strategies to increase crop production is effective pest management, because more than 30% of the world annual food production is lost through pest infestation (Pimentel, 1992; Kibuthu et al., 2016; Li and Jennings, 2018). In the tropical countries, crop loss is even more severe, because the prevailing high temperature and humidity are highly conducive to rapid multiplication of pests. Thus, the application of a wide variety of pesticides on crop plants is necessary to combat pests and diseases. The continuous production and use of these chemicals load the environment with xenobiotic substances and create complex ecological problems in the environment (Jennings and Li, 2014; Kibuthu et al., 2016).

The term 'heavy metals' is used in this text synonymously with 'trace metals' and includes both the essential (e.g. Fe, Cu, and Zn) and non-essential (Cd, Pb, and Hg) ones. All of these have the potential to be toxic to living organisms if present and available above (or below; for essential metals) a certain threshold which varies between taxa (Bleeker et al., 1992; Mouneyrac et al., 1998). Essential metals are required in small amounts (e.g. Zn: 0.75-2 mg/l and Cu: 0.06-0.16 mg/l for bivalves) (Watling, 1981) by organisms as component of proteins and enzymes (Underwood, 1974; Simkiss et al., 1982; Bleeker et al., 1992). Most animals can regulate the body concentration of these metals up to a certain ambient water concentration, above which accumulation starts and toxic effects may occur. Non-essential metals are considered not to play any biological role and cannot be regulated. These metals can be toxic at low concentration (that is, close to the ppt levels or ng/l) (Amiard et al., 1987; Fowler, 1990; Rainbow, 1995; Laporte et al., 1997; Otchere et al., 2003).

## TRACE ELEMENTS

At present, 14 elements are known to be essential to animal life and regulated at trace concentrations: Fe, Zn, Cu, Mn, Co, I, Mo, Se, Cr, Sn Ni, F, Si, and V. The non-essential ones including: Hg, Pb, Cd, As, Au, Ag, etc., are considered not to play any biological role and cannot be regulated. A further trace element, boron, is essential for plant life. In addition, 20 to 30 other elements occur, more or less constantly in low but variable concentrations in living tissues. The presence of these elements is believed to reflect the contact of the organism with its environment. Indeed Sn, F, Si, Ni and V, often referred to as the 'newer trace elements', only emerged as essential recently as an outcome of the use of purified diet and plastic isolator techniques. A few trace elements are referred to as toxic because they are toxic at relatively low concentrations (close to the ppt level or ng/l); however, this statement can be misleading because all the elements are toxic if ingested at sufficiently high levels and for long enough periods. With some, notably fluorine in man and Cu in sheep, the margins between beneficial and toxic intakes, that is, between needs and tolerance are small. With others such as Zn and Mn, tolerance is high and wide margins exist between minimum essential intake and those resulting in toxic effects (Underwood, 1974; Fowler, 1990; Bleeker et al., 1992; Laporte et al., 1997; Otchere, 2003; Obirikorang et al., 2011). Concentrations of heavy metals in aquatic environments have two phases: dissolved and particulate. The comparison of metal levels between different years in the same environment or between different environments should be considered very carefully, taking into accounts the seasonal variability which can be very important in estuaries. In these environments, concentrations may depend very much on salinity, apart from the seasonal variability (Baeyens, 1998; Baeyens et al., 1998a).

## BIO-MONITORS

Monitoring programs and research for trace metals in environmental samples have become widely established because of concerns over accumulation and toxic effects, particularly in aquatic organisms and to humans consuming these organisms. The criteria by which organisms are accepted as biological indicators for the assessment of contamination were proposed about fifty (50) years ago since 1970 and remain unchanged (Butler et al., 1971; Boyden, 1974; Phillips, 1976; Fowler and Oregioni, 1976; Otchere, 2003). Bivalves are widely used as bio-indicators of heavy metals pollution in coastal areas because of their abundance and known to concentrate these elements, providing a time integrated indication of environmental contamination. Bivalves are chosen because they are ideal indicators of heavy metal pollution, if and when pollution is present, and they facilitate analysis by this natural process of metal pro-

concentration. This bioaccumulation also gives a good idea of the amount of metal that is available for uptake in the system as metal concentrations in seawater or sediment alone do not give a true indication of what is actually available for uptake (Obirikorang et al., 2011). Bivalves are generally consumed by waterfowl and fish and so are sources of heavy metals to higher trophic levels. In comparison to fish and crustaceans, bivalves have a very low level of activity of enzyme systems capable of metabolizing persistent organic pollutants (POPs), such as aromatic hydrocarbons and polychlorinated biphenyls (Otchere, 2005). Therefore, contaminants concentrations in the tissues of bivalves more accurately reflect the magnitude of environmental contamination (Phillips, 1977a, b, 1980, 1990). Factors known to influence metal concentrations and accumulation in these organisms include metal bioavailability, season of sampling, hydrodynamics of the environment, size, sex, changes in tissue composition and reproductive cycle (Boyden and Phillips, 1986). Seasonal variations have been related to a great extent to seasonal changes in flesh weight during the development of gonadic tissues (Joiris et al., 1998, 2000). Element concentrations in mollusks at the same location differ between different species and individuals due to species-specific ability/capacity to regulate or accumulate trace metals (Reinfelder et al., 1997; Otchere et al., 2003). Different animals in the same community at the same trophic level could accumulate pollutants differently due to differences in habitat/niche's physical and chemical properties (Otchere, 2003, 2005).

The smallest individuals of shellfish often show the highest concentrations of trace metals on a weight specific basis. In such situations it is very difficult to assess whether observed differences in tissue levels between populations reflect real differences in environmental trace element constitution, or are merely due to variations in body size. This problem can be avoided by determining element concentration over a range of body sizes and reference made to a specific size (or size range) for comparative purposes. However, environmental metal levels are not the only factor affecting the metal content of molluscs, as both the size and season markedly affect this parameter in order to avoid the problem of cases where it is difficult to assess whether observed differences in tissue level/burden between these bivalves reflect real differences in environmental heavy metal constitution, or are merely due to variations in body size. Boyden (1974: 77) proposed model relating metal content to body weight; metabolic power function:  $Y = aW^b$ ; where  $Y$  = burden/content,  $W$  = dry weight,  $a$  = intercept and  $b$  = the slope co-efficient. He showed that the regression coefficient of the relationship between metal content and body weight was generally constant for a given metal and species. Thus, the intercept can be used to compare environmental metal levels from different locations

(Otchere, 2003).

Once in the organism, metals are stored or eliminated in different ways. Cd, Cu, Hg, Ag, Au, Bi and Zn can be complexed to cystein-rich heat stable, low molecular weight proteins called metallothioneins (MT), in different tissues of the organism (Langston and Zhou, 1986; Bouquegneau and Joiris, 1988; Bebiano and Langston, 1991; Bordin et al., 1992, 1997). The metal binding affinity for the protein order is Zn - Cd - Cu - Hg, suggesting that toxic metals such as Cd and Hg are able to displace essential metals (Bouquegneau and Joiris, 1988). Metal-binding proteins similar to MT have been recently identified in different tissues of *Mytilus edulis* and other bivalves (*Macoma balthica*) exposed to Cd, Cu, Zn, Hg and Ag (Roesijadi, 1982; Bebiano and Langston, 1991; Bordin et al., 1992, 1997). The functions attributed to MT included detoxification, storage and regulation of heavy metals. Moreover, it has been shown that metals such as Cd, Zn and Cu can induce MT synthesis in aquatic animals. Thus, MT has been proposed as indicators of trace metal pollution (Bouquegneau and Joiris, 1988; George and Olsson, 1994; Roesijadi, 1992; Bordin et al., 1997).

## SEASONAL VARIATIONS

The influence of season on the concentration of trace metals in bivalves has also been investigated (Dare and Edwards, 1975; Cossa et al., 1980; Boyden and Phillips, 1986; Cossa and Rondeau, 1985; Bordin et al., 1992; Otchere, 2003). Bryan (1973) gave a more detailed seasonal profile for the concentration of Zn, Pb, Cu, Co, Fe, Mn and Ni in tissues of *Pecten maximus* and *Chlamys operacularis* from the English Channel. In general, the concentrations of metals in these scallops were greatest in autumn and winter and it was suggested that metal concentrations were inversely related to phytoplankton production. However, Fowler and Oregoni (1976) in their studies of the variation in the concentration of ten metals in *Mytilus galloprovincialis*, suggested that the seasonal maximum (Cd: 4.0 µg/g dw; Pb: 12 µg/g dw; these are 2 and 3-fold respectively more than that of summer concentrations) seen in the samples collected in March (spring) was due to the reproductive state of the animals and to the winter run-offs increasing the amount of available metals. Phillips (1976) reached a similar conclusion concerning the seasonal variations of Zn, Cd, Pb and Cu in *M. edulis*. The seasonal fluctuations of trace metal concentrations were inversely linked to the seasonal changes in tissue weights of individual animals. Weight changes were related to the sexual cycle, with a minimum in late winter or early spring. Bordin et al. (1992) found similar seasonal fluctuations (in *M. balthica*, from Netherland and Belgian coasts) for four metals with higher concentrations in winter and lower in summer, ranging from 17 to 32. µg/g for Cu, 0.2 to 1.1 µg/g for Cd;

377 to 690  $\mu\text{g/g}$  for Zn and 500 to 2000  $\mu\text{g/g}$  for Fe (all in dw). Joiris et al. (1998) working with the cockle *Anadara (Senilia) senilis* from West Africa found a seasonal maximum of Hg in the dry season (0.4  $\mu\text{g/g}$ ) and a minimum in the wet season (0.1  $\mu\text{g/g}$  dw), partly attributed this to the seasonal fluctuation of phytoplankton production, seasonal abiotic factors (such as temperature and salinity) and the spawning/physiology of the cockles.

## SOURCES OF METAL POLLUTION

Generally, the sources for trace metals in aquatic systems are through natural and anthropogenic. In the marine ecosystem, the natural source has been categorized into:

- i) Coastal supply, including inputs from rivers and from erosion produced by wave action and glaciers.
- ii) Deep sea supply, including metals released from deep sea volcanism and those removed from particles or sediments by chemical processes.
- iii) Supply which bypasses the near-shore environment; in particular, metals transportation through the atmosphere as dust particles or as aerosols and also material produced by glacial erosion in polar regions which is then transported elsewhere by floating ice.

The two main routes of anthropogenic inputs into the sea are the atmosphere and rivers. Metal particles released into the air at ground level are mixed vertically and consequently, contaminants may be transported many thousands of kilometers from where they were first released. Differences in global climate also results in uneven deposition of trace metals. For example, the net accumulation of Hg, Cd, V and Mn in Arctic biota and ice has been attributed to the relative absence of precipitation, scavenging and strong atmospheric inversions. The emission source of these metals is thought to have come from industrialized temperate zones and thus considerable directional transport has occurred. In reviewing the aquatic transport of chemical, Goldberg (1989) concluded that organic compounds in the sea play a key role in trace metal transport. Comparisons of atmospheric and riverine input of trace metals in the sea indicates that only 2% of the Pb, for instance, which eventually dissolves in seawater enters the global ocean via rivers. The primary source of most of the dissolved Cd, Cu, Fe and Zn is also the atmosphere. At a regional level, it is thought that for the trace metals Cd, Hg, Cu, Pb and Zn, 40 to 60% of the input into the North Sea is via atmospheric deposition. Domestic effluent and urban storm water runoff have also been identified as significant sources of trace metal input into coastal waters. Concentrations in the milligram per liter range can be found in domestic effluent; metabolic waste, corrosion of water pipes (Cu, Pb, Zn and Cd) and

consumer products (e.g. Detergents formulations containing Fe, Mn, Cr, Ni, Co, Zn, B and As) all contribute appreciable amounts. Finally, it should be remembered that metals are indestructible (unlike organic compounds); they are retained and accumulated within ecosystem because reactive forms bind to sediments and non-reactive forms occur as insoluble oxides and salts. Thus, ecotoxicological effects of metals can persist for decades after pollution incidents and also when sediments and mine waste are disturbed (Connell and Miller, 1984).

## FINDINGS

Mercury levels from literature around the world generally, are the same order of magnitude to those reported in Table 1. Biney (1991) found mean concentrations in oysters: 0.06  $\mu\text{g/g}$  fw, cockles: 0.055  $\mu\text{g/g}$  fw and mussels: 0.06  $\mu\text{g/g}$  fw (fw = fresh weight). These were equivalent to 0.48, 0.37 and 0.46  $\mu\text{g/g}$  dw, respectively similar to concentrations reported in Table 1. Average Hg concentration from Ivory Coast using the same species of oysters was 0.125  $\mu\text{g/g}$  fw; this high value may be due to higher urbanization at Abidjan (Metongo, 1991), and while Mbome (1988) recorded a mean value of 0.083  $\mu\text{g/g}$  fw in Cameroon waters with *Crassostrea tulipa*. These were within the range of 0.034-0.13  $\mu\text{g/g}$  fw for western and central Africa sub regions reported by Biney et al. (1994).

Metal concentrations in bivalves from West Africa and elsewhere in Africa were not exceptional when compared with those reported for other coastal areas throughout the world. For example, Cu, Fe and Mn levels in the oysters, cockles and mussels have the same order of magnitude as in literature (Table 2); while Cd levels recorded in Africa were similar or lower than concentrations in other studies. In the case of oysters, the Zn level reported in Ghana (175  $\mu\text{g/g}$  fw) was much lower than the levels from other studies in West Africa (e.g. Côte d'Ivoire -1200 and Nigeria - 630  $\mu\text{g/g}$  fw). Metal concentrations in mussels from Oman and India compared to those from African and European coastal waters are similar.

## The comparison

Differences in Zn, Fe and Cu, essential elements for cockles, mussels and oysters, could be due to specific internal regulatory processes. Since these three elements are essential to life, we may expect a stronger influence of specific species than the variations in Mn. Higher wet season concentrations in Zn, Fe and Mn in the bivalves were similar to levels recorded in the mussel *Perna viridis* from India by Rivonker and Parulekar (1998); they attributed this observation to higher content of organic matter brought in by monsoon rains. Joiris and Azokwu (1999) reported higher concentrations (Cu, Fe and Zn) in

**Table 1.** Total and organic Hg ( $\mu\text{g/g dw}$ ) and percent MeHg reported in bivalves by different authors: median and range where appropriate.

Species	Place	$\Sigma\text{Hg}$	MeHg	%MeHg	References
<i>Anadara (S) senilis</i>	Ghana	0.19	0.10	46	Otchere et al. (2003)
<i>Anadara tuberculosa</i>	Costa Rica	0.16	0.08	51	De la Cruz (1994)
<i>Anadara granosa</i>	Malaysian Coast	0.03 - 0.6	-	-	Jothy et al. (1983)
<i>Anadara (S) senilis</i>	Nigeria	0.18	0.06	32	Joiris et al. (1998)
<i>Cerastoderma glaucum</i>	France	0.55	-	-	Szefer et al. (1999)
<i>Venerupis galactites</i>	Australia	0.71	0.13	18	Francesconi and Lenenton (1992)
<i>Crassostrea tulipa</i>	Ghana	0.17	0.08	52	Otchere et al (2003)
<i>Crassostrea virginica</i>	Georgia USA	1.60	0.27	17	Gardner et al. (1978)
<i>Crassostrea virginica</i>	Indian coast	0.10	-	-	Sanzgiri et al. (1988)
<i>Saccostrea echinata</i>	N. Australia	0.27	-	-	Peerzada et al. (1993)
<i>Macoma phenax</i>	Georgia USA	1.7	0.03	2	Gardner et al. (1978)
<i>Katelysia scalarina</i>	W. Australia	0.49-2.7	0.19-0.45	30	Jackson et al. (1986)
<i>Perna perna</i>	Ghana	0.26	0.10	37	Otchere et al. (2003)
<i>Perna viridis</i>	Indian coast	0.46	-	-	Sanzgiri et al. (1988)
<i>Mytilus galloprovincialis</i>	N. Adriatic Sea	2.0	0.5	25	Mikac et al. (1985)
<i>Mytilus edulis</i>	Belgian coast	0.17 - 0.23	0.02 - 0.05	-	Gurney (1992)
<i>Mytilus edulis</i>	Canada	0.07-0.38	-	-	Cossa and Rondeau (1985)
<i>Mytilus edulis</i>	W. Danish coast	7.5	0.3	4	Riisgard et al. (1985)
<i>Mercenaria mercenaria</i>	Georgia USA	0.80	0.28	35	Gardner et al. (1978)

dw: Dry weight.

**Table 2.** Average metal concentrations ( $\mu\text{g/g fw}$ ) reported in bivalves by different authors for other coastal areas throughout the world.

Parameter	Cu	Zn	Fe	Mn	Cd	Reference
<b>Oysters</b>						
Ghana	4.4	175	56	1.84	0.06	Otchere (2003)
Ghana	3.1	460	76	2.95	-	Biney (1991)
Nigeria	5.8	630	-	-	0.17	Okoye (1991a)
Cameroon	8.5	410	-	-	0.25	Mbome (1988)
Côte d'Ivoire	24.5	1200	-	-	0.65	Metongo (1991)
Venezuela	3.9	0.14	-	-	0.09	Jaffé et al. (1998)
Oman	27.6	140	48	0.64	1.96	Fowler et al. (1993)
<b>Cockles</b>						
Ghana	0.90	7.3	110	1.98	0.05	Otchere (2003)
Ghana	1.01	13	11	1.59	-	Biney (1991)
Nigeria	1.0	15	62	-	0.03	Joiris and Azokwu (1999)
Belgium	3.6	74	198	-	0.13	Bordin et al. (1992)
Costa Rica	0.83	10	90	-	0.63	De la Cruz (1994)
France	1.6	11	130	1.2	0.19	Szefer et al. (1999)
Saudi Arabia	1.2	15	71	9.8	0.12	Fowler et al. (1993)
<b>Mussels</b>						
Ghana	1.58	5.2	130	1.74	0.13	Otchere (2003)
Ghana	1.96	18	65	-	-	Biney (1991)
Oman	0.91	5.2	10	0.53	3.6	Fowler et al. (1993)
India	3.1	64	340	17.7	-	Rivonker and Parulekar (1998)
WHO limits	30.0	1000	-	-	2.0	Moraes et al. (1997)

fw: Fresh weight.

wet season in the cockle (*Anadara senilis*) from Nigeria and they ascribed this variation to increased run-off water with possible increase in pollutants load. Likewise, Joseph and Srivastava (1993), Mitra and Choudhury (1993) and Pillai and Valsala (1995) observed increased concentrations of heavy metals during the monsoon season.

Elevated levels of Zn in both (wet and dry) seasons (2400 and 2800 µg/g dw for dry and wet seasons) in oysters from Ghana, Fe in cockles and mussels might be due to treated wood used in boat construction (e.g. marine paints, etc.) and anthropogenic flux of metallic contaminants (Weis et al., 1993; Ferreira and Vale, 1995; Szefer et al., 1999). These high levels might also reflect the presence of blood systems or transport medium with these metals as essential components; for example, hemoglobin in cockles (high Fe content) and haemocyte in oysters (high in Cu and Zn). Similarly, Rivonker and Parulekar (1998) reported high levels of Fe in mussels from India (ranged: 200 - 4000 µg/g dw) and attributed these levels to the high uptake capacity of mussels towards this particular metal. While Boyden and Phillips (1981) working on *Crassostrea gigas* found similar trends of high concentrations (Cu ranged: 100 - 7000 µg/g dw; Zn ranged: 2 - 16 mg/g dw), they concluded that inherent variability of elements in a bivalve population depended on the particular species-metal pair considered, and also on the degree of contamination involved. Other studies with these species-metal pairs are shown in Table 2.

Manganese levels in cockles and mussels do not seem to follow any specific trend in variations; and may reflect which of the two bivalves could efficiently regulate this metal. On the other hand in oysters, concentrations were more variable. Frazier (1975, 1976) found high turnover of Mn in soft tissue of *Crassostrea virginica* during the period of shell growth and the amount of Mn incorporated into the shell in one day was twice the total Mn burden of the whole soft parts. Boyden and Phillips (1981) observed a similar phenomenon; they inferred that seasonal changes in the concentration and body burden of Mn in oysters depended primarily on factors other than the cyclic changes of tissue weight associated with gametogenesis and spawning. Levels reported in this review might reflect over-riding influence of shell deposition, even though the data are also suggestive of significant losses of Mn in gametes at spawning. No other plausible explanation for Mn variations in this review has been found. Results presented in this review showed that from a practical point of view when using bivalves as a quantitative indicator of metal pollution, influence of the size of molluscs must be taken into account even when the shell length has been classified. For example, Cossa and Rondeau (1985) found that the Hg load of mussels of 3 and 4 cm in length differ by 100% due to size. In addition, changes in weight during a season may cause a 300% difference in Hg content of the mussels.

Expressing results in terms of metal concentration in

the soft tissue circumvents the major bias due to size. Neglecting to take into account the influence of length and/or weight could have more drastic consequences, as noted above and by other authors (Joiris and Azokwu, 1999; Joiris et al., 1998, 2000; Otchere, 2003).

## CONCLUSION

Fluctuations in trace metal concentrations have been related to changes in metal bioavailability. Not only this but also several biological variables such as size, sex or changes in tissue composition and reproductive cycle as well as the season of sampling and the hydrodynamics of the environment have to be considered (Boyden and Phillips, 1981; Phillips and Segar, 1986; Joiris et al., 1998, 2000; Szefer et al., 1999). Many authors writing on seasonal variations have reported higher concentration in soft tissue during winter than in summer (Cossa et al., 1980; Cossa and Rondeau, 1985; Bordin et al., 1992; Regoli and Orlando, 1993, 1994; Soto et al., 1995; Regoli, 1998; Szefer et al., 1999). These seasonal variations have been related to a great extent to seasonal changes in flesh weight during development of gonadic tissues (Regoli and Orlando, 1994; Joiris et al., 1998; Szefer et al., 1999; Bordin et al., 1997). However, even if the effects of these parameters were eliminated by the use of specific sampling procedures, element concentrations will still differ between different species due to species specific ability/capacity to regulate and/or accumulate trace metals (Tanabe et al., 1987; Reinfelder et al., 1997; Otchere, 2003).

Concluding, the wet season/winter-spring maxima in Zn, Fe and Mn observed should reflect a higher metal availability during this season (through 'import'). This could not be reproducible (due to differential ability of some species to regulate more efficiently than others) and would allow one to infer that temporal variability in metal concentration from sites to sites are irregular; nevertheless, it could have also depended on the amount of rainfall/precipitation at each location during the season. This irregularity might also be due to varying gut content since bivalves were neither depurated nor drained prior to storage. The essential metals which were reviewed, were present in similar respective concentrations to those found around the world and exhibited similar seasonal pattern in terms of their concentrations although of different magnitudes. In order to remove the variability due to seasonally changing body weight, the use of metal content (absolute value) instead of metal concentration in tissue against weight or size has been proposed by several authors (Boyden, 1974; Cossa et al., 1980; Boyden and Phillips, 1981; Cossa and Rondeau, 1985). Considering metal load or content instead of metal concentration provides more information on metal behavior. While temporal variations in element concentration were mainly caused by changes in the

tissue weight of bivalves according to the sexual cycle, body burden of elements alter only a little throughout the year. Hence data expressed both as metal concentration and load/content could be integrated for a better assessment of differences in the bioavailability of these metals as a function of site and time of collection.

## CONFLICT OF INTERESTS

The author has not declared any conflict of interests.

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