



Cerium Inhibits the Cadmium Uptake of Pacific Oyster *Crassostrea gigas*

Huang Guo Qing^{1,2}, Wang Dong Feng^{2,*}

¹ Qingdao Agricultural University, College of Food Science and Engineering, Qingdao 266109, China.

² Ocean University of China, College of Food Science and Engineering, Qingdao 266103, China.

* Corresponding Author: Tel.: +86.532 82031575; Fax: +86.532 82031575;
E-mail: wangdfouc@126.com

Received 10 January 2016
Accepted 20 April 2016

Abstract

Ce³⁺ is a potential antagonist of Cd²⁺ due to their similar ionic radius. Its effect on the bioaccumulation of Cd²⁺ in Pacific oyster *Crassostrea gigas* was explored through laboratory experiments. The oysters were exposed to 5, 10 and 25 µmol/L Ce(NO₃)₃ in the presence of 25 µg/L Cd²⁺ (0.22 µmol/L) for 12 days and the concentrations of Cd, Ca, Zn as well as Cu were indicated in the whole soft tissues and separate organs. Ce³⁺ in all the three doses significantly inhibited Cd²⁺ bioaccumulation in the oysters on day 12 and the inhibition ratios reached 30%, 31% and 41% respectively. Organ-level analysis revealed that Ce³⁺ possibly exerted its inhibitory action by delaying the saturated absorption of Cd²⁺ in the gill. Ce³⁺ in all the three doses antagonized the negative effects of Cd²⁺ on Ca concentration and restored its level to the normal range on day 12, but 25 µmol/L Ce³⁺ decreased the Zn and Cu concentrations by up to 27% and 88% respectively. It was concluded that Ce³⁺ could markedly inhibit the Cd²⁺ bioaccumulation of *Crassostrea gigas* and the presence of Ce³⁺ must be considered in marine pollution assessment by using oysters as the bioindicator.

Keywords: Cerium, cadmium, bioaccumulation, marine bivalve.

Introduction

Among various heavy metals, rare earth elements (REEs) are a special class that has shown interesting biological effects on organisms. In China, REE-based microfertilizers have been widely applied for over 30 years to enhance the yield and quality of crops, vegetables (Hu *et al.*, 2004) and animals (He *et al.*, 2003). Due to the outstanding positive biological effects, lanthanum (La) and cerium (Ce), the two most abundant REEs on the earth (Palasz and Czekaj, 2000), have been listed in the *Approved Feed Additives* by the Ministry of Agriculture of the People's Republic of China in 2008.

Marine bivalves are well known to have the ability to concentrate heavy metals in their soft tissues from water and sediments due to their poor mobility (Engel, 1999) and some marine bivalves have been selected as bioindicators for marine pollution assessment (Regoli and Orlando, 1993; Munoz-Barbosa *et al.*, 2000; Olivier *et al.*, 2002). Cadmium (Cd) is one of the most intensively studied heavy metals and its concentration in bivalves could indicate the Cd pollution level of the environment. However, the presence of other metals possibly affects the bioaccumulation of Cd²⁺. For example, exposure to

Zn²⁺ increased the tissue concentration of Cd in oysters (Liu and Wang, 2013), while elevated seawater Ca²⁺ concentration decreased the Cd²⁺ uptake of Asiatic clams (Qiu *et al.*, 2005). Hence, the existence of such competitive metals must be considered in the heavy metal pollution assessment of the marine environment.

The radii of REEs range from 9.6-11.5 nm and are very close to that of Ca²⁺ (9.9 nm). REEs have been nicknamed super-calcium (Brown *et al.*, 1990) and are theoretically able to interfere in the metabolism of Ca²⁺ and other metal ions with similar radius, such as Cd²⁺ (10.9 nm). Among the REEs, La³⁺ has been widely recognized as an antagonist of Ca²⁺ (Weiss, 1974) and a non-specific inhibitor of the Ca²⁺ channel (Verboost *et al.*, 1989). The effects of La³⁺ on the metabolism of Ca²⁺ and/or Cd²⁺ in marine bivalves (Wang and Fisher, 1999) and many plants (He *et al.*, 2005; Wang *et al.*, 2012) have been reported. Nevertheless, such information for Ce³⁺ is still nearly unavailable, except the report that Ce³⁺ significantly inhibited the absorption of Cd²⁺ by a plant *Azadirachta indica* (Wang *et al.*, 2008).

With mineral exploitation and increasing use as a strategic resource, REEs will inevitably enter the environment, especially aquatic environment (Cui *et*

al., 2012). The deposition of REEs in aquatic environment and marine organisms has been reported by several authors. For example, an investigation carried out in 2009 revealed that the bivalves collected from the Shenzhen coastal region of China could accumulate REEs, including Ce^{3+} , in their tissues, and oysters were suggested as a bioindicator for REEs level in marine environment due to their high potential of REEs enrichment (Zhang *et al.*, 2009); the sediments passing from the Volga delta could enrich various REEs except europium (Eu) (Sval'nov *et al.*, 2011); and the freshwater bivalve *Corbicula fluminea* sampled from the Rhine River of Europe had incorporated La and samarium (Sm) in their shells (Merschel and Bau, 2015).

Since Ce^{3+} has a radius similar to that of Ca^{2+} and Ca^{2+} shares the same transport channel with Cd^{2+} in marine bivalves (Verbost *et al.*, 1989), Ce^{3+} should impose certain effects on the bioaccumulation of Cd^{2+} and other related metals in aquatic organisms as La^{3+} . Hence, the purpose of the current work is to explore whether Ce^{3+} could influence the accumulation of Cd^{2+} as well as the concentrations of other related metals including Ca, Zn and Cu in pacific oyster *Crassostrea gigas* through laboratory experiments. This work is expected to provide useful information for better clarifying the importance of Cd concentration as an indicator of environmental pollution.

Materials and Methods

Oyster Collection and Maintenance

The individuals of pacific oyster *Crassostrea gigas* were collected from a breed farm in Qingdao, China. Animals of a narrow size range (mean shell length 6.0~7.0 cm) were selected to minimize the possible effects of size on metal metabolism. The bivalves were rinsed with seawater to remove debris from the shell surface and then acclimated to plastic tanks filled with 40 L natural seawater (containing a background Cd concentration of 0.027 $\mu\text{g/L}$ (0.24 nmol/L) and sand filtered before use) for 15 days. The oysters were not fed during acclimation and subsequent experiments. The seawater temperature was maintained at 20 ± 1 °C, salinity at 31 ± 0.1 ‰, alkalinity at 2.74 mmol/L and pH at 8.2 ± 0.3 with additional aeration supplied throughout the experiment. The seawater in each tank was exchanged every 24 h during acclimation, and

excreta and dead oysters were removed in the meantime.

Experimental Design

The Ce^{3+} doses 5, 10 and 25 $\mu\text{mol/L}$ together with a sublethal Cd^{2+} concentration 25 $\mu\text{g/L}$ (0.22 $\mu\text{mol/L}$) were selected in this work according to preliminary experiments. A total of 250 acclimated oysters were randomly divided into five groups, including the negative control group, the positive control group, the low Ce^{3+} group, the medium Ce^{3+} group and the high Ce^{3+} group, with 50 oysters in each group. The oysters in the negative control group were reared in natural seawater and those in the positive control group were placed in natural seawater supplemented with 25 $\mu\text{g/L}$ Cd^{2+} (added as $CdCl_2$). In the low, medium and high Ce^{3+} groups, the seawater contained 5, 10 and 25 $\mu\text{mol/L}$ Ce^{3+} (in $Ce(NO_3)_3$) respectively in addition to 25 $\mu\text{g/L}$ Cd^{2+} . The experimental design was summarized in Table 1. The bivalves were maintained in the conditioned seawater for 12 days and the seawater in the tanks was exchanged every day. On day 4, 8 and 12, 16 oysters were sampled from each tank for metal analysis.

Soft Tissue Preparation and Metal Measurement

The sampled oysters were immediately steamed in a pot until their shells were opened. Then, the soft tissues were taken out, rinsed thoroughly with deionized water and cooled to room temperature. Among the 16 oysters collected from each group each time, 8 individuals were used for whole-soft tissue Cd, Ca, Zn and Cu concentration analysis and the others were further dissected into gill, digestive gland and muscle for organ-level Cd concentration determination. All the soft tissues and organs were dried in 105 °C to constant weight, weighed and then digested in the mixture of 5 mL concentrated HNO_3 and 3 mL $HClO_4$ in 180 °C by using a digester (ED36 DigiBlock digester, LabTech, USA) until the solution became transparent.

The concentrations of Cd, Zn, Ca and Cu were determined with a flame atomic absorption spectrometry (AA6800, Shimadzu, Japan). The detection limits of the four metals were 2, 1, 1 and 3 $\mu\text{g/L}$ respectively. The standard stock solutions of the metals were purchased from the National Analysis Center for Iron and Steel of China (Beijing, China) and diluted to appropriate solutions with deionized

Table 1. Experimental design for the influence of Ce^{3+} on Cd^{2+} accumulation in *Crassostrea gigas*

| Experimental group | Seawater condition | Number of animals |
|------------------------|---|-------------------|
| Negative control | Seawater only | 50 |
| Positive control | Seawater containing 25 $\mu\text{g/L}$ Cd^{2+} | 50 |
| Low Ce^{3+} group | Seawater containing 25 $\mu\text{g/L}$ Cd^{2+} and 5 $\mu\text{mol/L}$ Ce^{3+} | 50 |
| Medium Ce^{3+} group | Seawater containing 25 $\mu\text{g/L}$ Cd^{2+} and 10 $\mu\text{mol/L}$ Ce^{3+} | 50 |
| High Ce^{3+} group | Seawater containing 25 $\mu\text{g/L}$ Cd^{2+} and 25 $\mu\text{mol/L}$ Ce^{3+} | 50 |

water prior to standard curve preparation. The analysis methods were simultaneously validated for each sample series by the analysis of a standard biological reference material LUTS-1 (non-defatted lobster hepatopancreas) purchased from the National Research Council (Ottawa, Canada). The results obtained were in good agreement with the certified values ($P < 0.05$). All the metal concentrations were presented in dry weight basis.

Statistical Analysis

Data were analyzed by SPSS 16.0. Values were the averages of eight replicates and significant differences among different treatments were determined using a one-way ANOVA followed by the multiple comparison with Dunnett's test. Statistical difference was accepted when $P < 0.05$.

Results and Discussion

Effect of Ce^{3+} on Whole-Soft Tissue Cd Bioaccumulation

As can be seen in Figure 1a, the background Cd concentration in the oysters was only 16.3 mg/kg·dw and remained constant in the entire experimental period. After the supplementation of 25 $\mu\text{g/L}$ Cd^{2+} to seawater, the Cd concentrations in the bivalves increased significantly to 43, 52 and 80 mg/kg·dw on day 4, 8 and 12 respectively, implying that the oysters successfully assimilated Cd^{2+} from the conditioned seawater.

Ce^{3+} has been reported able to inhibit the uptake of Cd^{2+} in the plant horseradish (*Armoracia rusticana*) (Wang *et al.*, 2008). A similar result was found in this work. Ce^{3+} in 25 $\mu\text{mol/L}$ significantly inhibited the bioaccumulation of Cd^{2+} in the oysters on all the sampling days and the inhibition ratios reached up to 30%, 31% and 41% on day 4, 8 and 12 respectively, but Ce^{3+} in the low and medium levels failed to affect Cd^{2+} uptake on day 4 and day 8. Although Ce^{3+} in all the three selected doses decreased Cd^{2+} uptake on day 12, no dose-dependent effect was observed.

Wang and Fisher (1999) reported that co-exposure to 50 $\mu\text{mol/L}$ La^{3+} and 18 nmol/L Cd^{2+} of 1 h for the marine mussel *Mytilus edulis* and 4 h for the marine clam *Macoma balthica* showed no major effect on the uptake of Cd^{2+} . Such different result with the current work implied that marine bivalves possibly possessed different sensitivity to La^{3+} and Ce^{3+} , or the inhibitory action was closely dependent on the metal dose and exposure duration, since 5 and 10 $\mu\text{mol/L}$ Ce^{3+} failed to affect Cd^{2+} bioaccumulation on day 4 and day 8 in contrast to on day 12.

Effect of Ce^{3+} on Cd Concentration in the Gill

To investigate the action pattern of Ce^{3+} , the effects of this REE on the Cd concentrations in the

gill, digestive gland and muscle of the oysters were also determined.

As shown in Figure 1b, the Cd concentration in the gill of the negative control group was as low as 23 mg/kg·dw and was nearly not changed during the experimental period, but the addition of Cd^{2+} to seawater led to increased Cd accumulation in the tissue and its concentration reached up to 74, 117 and 110 mg/kg·dw on day 4, 8 and 12 respectively. These values were much higher than the corresponding whole-soft tissue level on the same sampling days (Figure 1a), implying that the gill was an important organ for Cd^{2+} bioaccumulation. It should be noted that the Cd concentrations of the gill in the positive control group were not significantly different on day 8 and 12, but were significantly higher than that of the negative control group. This result was consistent with the findings that gill was only a temporal organ for Cd storage (Wu *et al.*, 2007) and could be saturated by excessive Cd (Javanshir *et al.*, 2009).

The presence of Ce^{3+} could reduce the accumulation of Cd^{2+} in the gill. As can be seen in Figure 2b, 5 and 10 $\mu\text{mol/L}$ Ce^{3+} failed to influence the Cd concentration in the gill on all the three sampling days, but 25 $\mu\text{mol/L}$ Ce^{3+} significantly reduced the Cd concentration by up to 49% and 58% on day 4 and day 8 respectively, implying that Ce^{3+} in high concentrations could delay the saturated absorption of Cd^{2+} in the gill. Significant differences were not observed between the three Ce^{3+} doses and the positive control group on day 12, indicating that saturated Cd^{2+} absorption occurred to the gill after exposure for 12 days (Javanshir *et al.*, 2009).

Effect of Ce^{3+} on Cd Concentration in the Digestive Gland

Digestive gland is another important tissue for Cd^{2+} accumulation in addition to gill (Roméo and Gnassia-Barelli, 1995). As can be seen in Figure 1c, the Cd concentration in the digestive gland of the positive control group increased significantly as the experiment proceeded and its levels amounted to 54, 65 and 80 mg/kg·dw on day 4, 8 and 12 respectively, which were 1.6, 2.0 and 2.9 folds of the negative control group.

Ce^{3+} in the three doses significantly inhibited Cd^{2+} accumulation in the organ on all the three sampling days, except the low concentration group on day 4. Besides, the high concentration group restored the Cd concentration in the digestive gland to the background level of the negative control group and dose-dependent pattern was observed on all the three sampling days.

On day 12, the Cd accumulation inhibition ratios reached 25%, 57% and 58% for the low, medium and high Ce^{3+} groups respectively. Previous researches have indicated that the gill was a temporal target organ of Cd^{2+} accumulation, and then Cd^{2+} was

transferred to digestive organs such as the digestive gland (Satish Nair and Robinson, 2001). The same trend was observed in oyster *Crassostreagigas* as evidenced by the continuously increasing Cd concentration in the digestive gland. Hence, it was proposed that Ce^{3+} hindered Cd^{2+} accumulation in *Crassostreagigas* by reducing the speed of saturated Cd^{2+} absorption in the gill.

Effect of Ce^{3+} on Cd Concentration in the Muscle

The variations of Cd concentrations in the muscle were illustrated in Figure 1d. It could be seen that the Cd concentration in the muscle kept increasing during the experiment, but since muscle was not a major tissue for Cd storage (Arockia Vasanthi *et al.*, 2012), its concentration was far less than that in the gill and digestive gland.

The presence of Ce^{3+} in the three levels exhibited no influence on the Cd concentration in the muscle on day 4 and day 12, but the ion in 10 and 25 $\mu\text{mol/L}$ significantly reduced the Cd concentration on day 8 by 37.5% and 45.1% respectively.

Effect of Ce^{3+} on the Whole-Soft Tissue Ca Concentration

Since Ce^{3+} has a similar ionic radius to that of Ca^{2+} , whether this REE could interfere in the Ca concentration of oysters was also concerned. As can be seen in Figure 2a, the addition of Cd^{2+} to seawater significantly decreased the whole-soft tissue Ca concentration of the positive control group by 14% and 23% on day 8 and day 12 respectively compared with the negative control group. This result was

consistent with the finding of Ngo *et al.* (2011), which found that the uptake of Cd^{2+} could decrease the Ca concentrations in various tissues of the bivalve *Anodontaanatina* (Linnaeus 1758).

Compared with the positive control group, the addition of Ce^{3+} in the three levels unexpectedly did not further reduce the Ca concentration in the oysters. In contrast, significant increase of Ca concentration was observed on all the three sampling days, though some differences were not significant. This result was inconsistent with the action of La^{3+} , which markedly reduced the Ca^{2+} concentrations in freshwater fish and crabs (Verbost *et al.*, 1989; Pedersen and Bjerregaard, 1995). To present, the effect of Ce^{3+} on the Ca metabolism has been reported only in horseradish, which found that Ce^{3+} decreased the Ca concentration in a low dose (20 mg/L), but increased the parameter in higher doses (100 and 300 mg/L) (Wang *et al.*, 2008). It was proposed that Ce^{3+} possibly also possessed the dose-dependent action pattern on the Ca concentration in marine bivalves as well and Ce^{3+} in the three doses selected in this work was beneficial for the Ca^{2+} absorption of pacific oyster.

Effect of Ce^{3+} on the Whole-Soft Tissue Zn Concentration

In marine bivalves, Zn^{2+} was also absorbed through the Ca^{2+} channel (Wang and Fisher, 1999). Hence, the variation of Zn concentration during Ce^{3+} exposure was also measured. As can be seen in Figure 2b, exposure to Cd^{2+} did not influence the Zn concentration in the oysters. This result was consistent with many reports, which indicated no interaction between Cd and Zn in marine

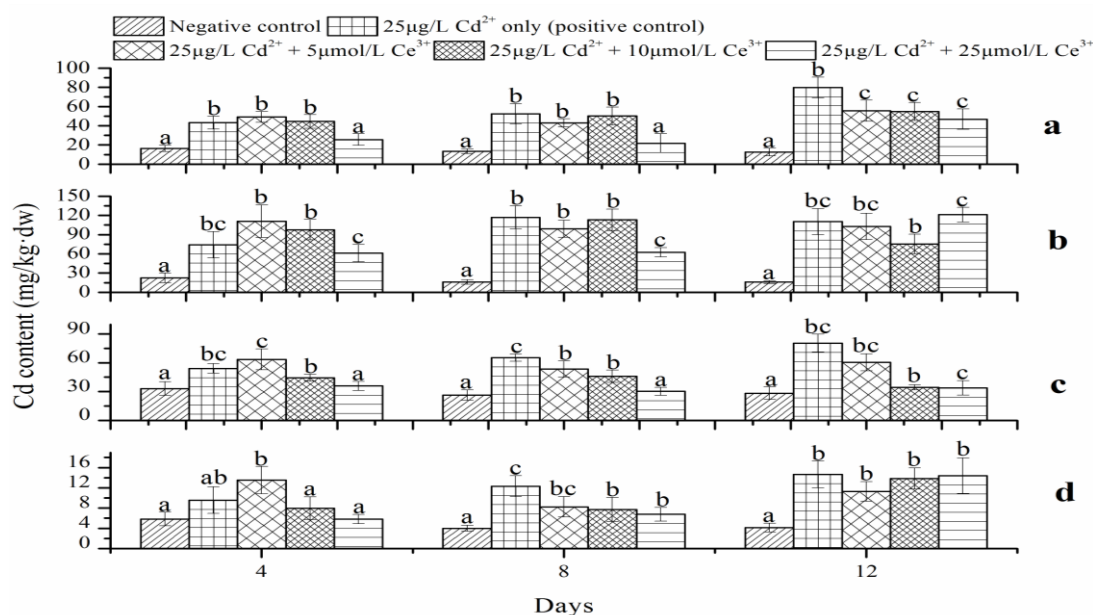


Figure 1. Effect of Ce^{3+} on the Cd concentrations in the whole-soft tissue (a), gill (b), digestive gland (c) and muscle (d) of *Crassostreagigas*. Multiple comparisons were performed between values of the same days. Bars with same letter(s) do not differ significantly at $P < 0.05$.

mussels(Wang and Fisher, 1996, 1999).

Nevertheless, the presence of Ce^{3+} greatly affected the Zn concentration of the oysters. According to Figure 2b, Ce^{3+} in 25 $\mu\text{mol/L}$ significantly reduced the Zn concentration on all the three sampling days, with the inhibition ratios reaching up to 33%, 48% and 27% on day 4, 8 and 12 respectively. The effects of Ce^{3+} on the Zn metabolism of organisms have not been reported to the present, but such effects have been observed for La^{3+} in plants (Xiong *et al.*, 2006; Wang *et al.*, 2012). Hence, Ce^{3+} possibly possessed similar impacts on Zn concentration as La^{3+} . Oyster is recognized as one of the richest sources for Zn^{2+} . Therefore, the presence of Ce^{3+} in the environment could possibly reduce the nutrition value of oysters in term of Zn concentration.

Effect of Ce^{3+} on the Whole-Soft Tissue Cu Concentration

Luxamaet *al.* (2011) found that diltiazem, which is a well-known Ca^{2+} channel blocker, could reduce Cu^{2+} accumulation in the gill of *Crassostrea virginica*, while La^{3+} did not. Hence, the effect of exposure to Ce^{3+} on the Cu concentrations of the oysters was explored as well and the result was presented in Figure 2c. It could be seen that the addition of Cd^{2+} to seawater did not affect the Cu concentration of the bivalve, but the presence of Ce^{3+} tremendously reduced the index. On day 12, Ce^{3+} in the three doses decreased the Cu concentration by up to 37%, 58% and 88% respectively. Such effect differed from that of La^{3+} (Luxama *et al.*, 2011), indicating that the two REEs exerted different effects on the Cu metabolism of marine bivalves.

The antagonistic effect of Ce^{3+} against Cu concentration could be possibly related to the presence forms of these metals. Cu^{2+} is well-known to bind to a sulphur-rich protein called metallothionein (MT) in marine bivalves for reduced toxicity (Wang *et al.*, 2011). Besides, REEs can also induce MT synthesis in mice (Kawagoe *et al.*, 2005) and *Ruditapes philippinarum* (Fu *et al.*, 2011). Although Ce^{3+} can induce MT synthesis in marine bivalves (Fu *et al.*, 2011), the newly produced MT was insufficient for the assimilated REE under acute exposure conditions and consequently competition for MT molecules occurred. Ce^{3+} possibly had a higher affinity to MT and Cu^{2+} was replaced by Ce^{3+} , leading to Cu^{2+} leakage from the oysters as a result.

Conclusion

Ce^{3+} could inhibit the bioaccumulation of Cd^{2+} in pacific oyster *Crassostrea gigas* by delaying the saturated absorption of Cd^{2+} in the gill. Besides, this metal also greatly affected the concentrations of other related metals, especially Zn and Cu, in the bivalves. Hence, the presence of this REE must be considered in the pollution assessment of the marine environment by using marine bivalves as the bioindicator.

Acknowledgements

We acknowledge the financial support from the Natural Science Foundation of China (30972289).

References

Arockia Vasanthi, L., Revathi, P., Arulvasu, C. and

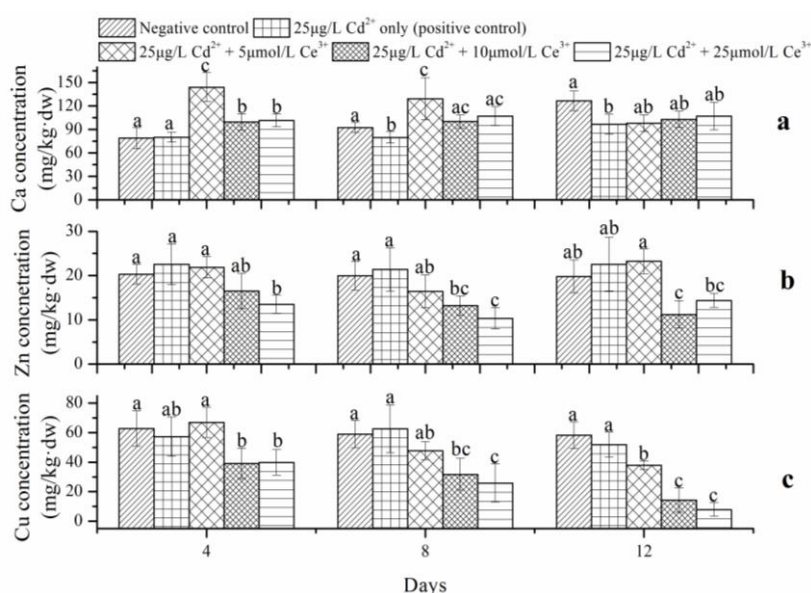


Figure 2. Effect of Ce^{3+} on the whole-soft tissue Ca (a), Zn (b) and Cu (c) concentrations of *Crassostrea gigas*. Multiple comparisons were performed between values of the same days. Bars with same letter(s) do not differ significantly at $P < 0.05$.

- Munuswamy, N. 2012. Biomarkers of metal toxicity and histology of *Perna viridis* from Ennore estuary, Chennai, south east coast of India. *Ecotoxicology and Environmental Safety*, 84: 92-98. doi:10.1016/j.ecoenv.2012.06.025
- Brown, P.H., Rathjen, A.H., Graham, R.D., Tribe, D.E., Karl A. Gschneidner, Jr. and LeRoy, E. 1990. Chapter 92 Rare earth elements in biological systems. In: Bünzli, J.C. and Pecharsky V.K. (Ed.), *Handbook on the Physics and Chemistry of Rare Earths*, Elsevier, Netherlands: 423-452. doi:10.1016/S0168-1273(05)80135-7
- Cui, J.A., Zhang, Z., Bai, W., Zhang, L., He, X., Ma, Y., Liu, Y. and Chai, Z. 2012. Effects of rare earth elements La and Yb on the morphological and functional development of zebrafish embryos. *Journal of Environmental Sciences*, 24: 209-213. doi:10.1016/S1001-0742(11)60755-9
- Engel, D.W. 1999. Accumulation and cytosolic partitioning of metals in the American oyster *Crassostrea virginica*. *Marine Environmental Research*, 47: 89-102. doi: 10.1016/S0141-1136(97)00100-1
- Fu, H.F., Meng, F.P., Gao, Y. and Zhao, S.S. 2011. Induction effect of metallothionein synthesis in *Ruditapes philippinarum* exposed to rare earth element Nd. *Research of Environmental Science*, 24: 1435-1440. doi: 10.13198/j.res.2011.12.107.fuhf.004
- He, M.L., Wang, Y.Z., Xu, Z.R., Chen, M.L. and Rambeck, W.A. 2003. Effect of dietary rare earth elements on growth performance and blood parameters of rats. *Journal of Animal Physiology and Animal Nutrition*, 87: 229-235. doi: 10.1046/j.1439-0396.2003.00432.x
- He, Z., Li, J., Zhang, H. and Ma, M. 2005. Different effects of calcium and lanthanum on the expression of phytochelatin synthase gene and cadmium absorption in *Lactuca sativa*. *Plant Science*, 168: 309-318. doi:10.1016/j.plantsci.2004.07.001
- Hu, Z., Richter, H., Sparovek, G. and Schnug, E. 2004. Physiological and biochemical effects of rare earth elements on plants and their agricultural significance: a review. *Journal of Plant Nutrition*, 27: 183-220. doi: 10.1081/PLN-120027555
- Javanshir, A., Shapoori, M., Azarbad, H., Mir-Vaghefi, A.R. and Daneh-Kar, A. 2009. Influence of calcium presence on the absorption of cadmium by the rock oyster *Saccostrea cucullata* from Persian Gulf (Ostreidae; Bivalvia) in laboratory conditions. *Journal of Ecology and The Natural Environment*, 1: 178-183.
- Kawagoe, M., Hirasawa, F., Wang, S.C., Liu, Y., Ueno, Y. and Sugiyama, T. 2005. Orally administered rare earth element cerium induces metallothionein synthesis and increases glutathione in the mouse liver. *Life Sciences*, 77: 922-937. doi:10.1016/j.lfs.2005.02.004
- Liu, F. and Wang, W.X. 2013. Facilitated bioaccumulation of cadmium and copper in the oyster *Crassostrea hongkongensis* solely exposed to zinc. *Environmental Science & Technology*, 47: 1670-1677. doi: 10.1021/es304198h.
- Luxama, J.D., Carroll, M.A. and Catapane, E.J. 2011. Effects of potential therapeutic agents on copper accumulations in gill of *Crassostrea virginica*. *In Vivo*, 31: 34-42.
- Merschel, G. and Bau, M. 2015. Rare earth elements in the aragonitic shell of freshwater mussel *Corbicula fluminea* and the bioavailability of anthropogenic lanthanum, samarium and gadolinium in river water. *Science of The Total Environment*, 533: 91-101. doi:10.1016/j.scitotenv.2015.06.042
- Munoz-Barbosa, A., Gutierrez-Galindo, E.A. and Flores-Munz, G. 2000. *Mytilus californianus* as an indicator of heavy metals on the northwest coast of Baja California, Mexico. *Marine Environmental Research*, 49: 123-144. doi:10.1016/S0141-1136(99)00052-5
- Ngo, H.T.T., Gerstmann, S. and Frank, H. 2011. Subchronic effects of environment-like cadmium levels on the bivalve *Anodonta anatina* (Linnaeus 1758): I. Bioaccumulation, distribution and effects on calcium metabolism. *Toxicological & Environmental Chemistry*, 9: 1788-1801. doi: 10.1080/02772240802386049
- Olivier, F., Ridd, M. and Klumpp, D. 2002. The use of transplanted cultured tropical oysters (*Saccostrea commercialis*) to monitor Cd levels in North Queensland coastal waters (Australia). *Marine Pollution Bulletin*, 44: 1051-1062. doi:10.1016/S0025-326X(02)00157-1
- Palasz, A. and Czekaj, P. 2000. Toxicological and cytophysiological aspects of lanthanides action. *Acta Biochimica Polonica*, 47: 1107-1114.
- Pedersen, T.V. and Bjerregaard, P. 1995. Calcium and cadmium fluxes across the gills of the shore crab, *Carcinus maenas*. *Marine Pollution Bulletin*, 31: 73-77. doi:10.1016/0025-326X(94)00260-G
- Qiu, J.W., Xie, Z.C. and Wang, W.X. 2005. Effects of calcium on the uptake and elimination of cadmium and zinc in Asiatic clams. *Archives of Environmental Contamination and Toxicology*, 48: 278-287. doi: 10.1007/s00244-004-0001-8
- Regoli, F. and Orlando, E. 1993. *Mytilus galloprovincialis* as a bioindicator of lead pollution: biological variables and cellular responses. *Science of The Total Environment*, 134(s2): 1283-1292. doi:10.1016/S0048-9697(05)80134-2
- Roméo, M. and Gnassia-Barelli, M. 1995. Metal distribution in different tissues and in subcellular fractions of the Mediterranean clam *Ruditapes decussatus* treated with cadmium, copper, or zinc. *Comparative Biochemistry and Physiology Part C: Comparative Pharmacology and Toxicology*, 111: 457-463. doi:10.1016/0742-8413(95)00060-7
- Satish Nair, P. and Robinson, W.E. 2001. Histidine-rich glycoprotein in the blood of the bivalve *Mytilus edulis*: role in cadmium speciation and cadmium transfer to the kidney. *Aquatic Toxicology*, 52: 133-142. doi:10.1016/S0166-445X(00)00138-7
- Sval'nov, V.N., Alekseeva, T.N. and Gazenko, A.O. 2011. Modern deposits of the Northern Caspian Sea and the Volga delta. *Oceanology*, 51: 118-130. doi: 10.1134/S0001437011010188
- Verboost, P.M., Van Rooij, J., Flik, G., Lock, R.A.C. and Bonga, S.E.W. 1989. The movement of cadmium through freshwater trout branchial epithelium and its interference with calcium transport. *Journal of Experimental Biology*, 145: 185-197.
- Wang, C., Luo, X., Tian, Y., Xie, Y., Wang, S., Li, Y., Tian, L. and Wang, X. 2012. Biphasic effects of lanthanum on *Vicia faba* L. seedlings under cadmium stress, implicating finite antioxidation and potential ecological risk. *Chemosphere*, 86: 530-537. doi:10.1016/j.chemosphere.2011.10.030

- Wang, L., Huang, X. and Zhou, Q. 2008. Effects of rare earth elements on the distribution of mineral elements and heavy metals in horseradish. *Chemosphere*, 73: 314-319. doi:10.1016/j.chemosphere.2008.06.004
- Wang W.X. and Fisher, N.S. 1996. Assimilation of trace elements by the mussel *Mytilus edulis*: effects of diatom chemical composition. *Marine Biology*, 125: 715-724. doi: 10.1007/BF00349254
- Wang, W.X. and Fisher, N.S. 1999. Effects of calcium and metabolic inhibitors on trace element uptake in two marine bivalves. *Journal of Experimental Marine Biology and Ecology*, 236: 149-164. doi:10.1016/S0022-0981(98)00195-6
- Wang, W.X., Yang, Y.B., Guo, X.Y., He, M., Guo, F. and Ke, CH. 2011. Copper and zinc contamination in oysters: Subcellular distribution and detoxification. *Environmental Chemistry*, 30: 1767-1774. doi: 10.1002/etc.571
- Weiss, G.B. 1974. Cellular pharmacology of lanthanum. *Annual Review of Pharmacology*, 14: 343-354. doi: 10.1146/annurev.pa.14.040174.002015
- Wu, S.M., Shih, M.J. and Ho, Y.C. 2007. Toxicological stress response and cadmium distribution in hybrid tilapia (*Oreochromis sp.*) upon cadmium exposure. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, 145: 218-226. doi:10.1016/j.cbpc.2006.12.003
- Xiong, S.L., Xiong, Z.T., Chen, Y.C. and Huang, H. 2006. Interactive effects of lanthanum and cadmium on plant growth and mineral element uptake in crisped-leaf mustard under hydroponic conditions. *Journal of Plant Nutrition*, 29: 1889-1902. doi: 10.1080/01904160600899485
- Zhang, H., Deng, L., Yang, J., Jiang, J., Shen, Z. and Xie, J. 2009. Rare earth elements in marine organisms from Shenzhen coastal region. *Wei Sheng Yan Jiu*, 38: 543-545.