BRAZILIAN JOURNAL OF OCEANOGRAPHY, 55(1):51-56, 2007

# ARE METALS OF ANTIFOULING PAINTS TRANSFERRED TO MARINE BIOTA?[1]

*Wladimir C. Paradas & Gilberto M. Amado Filho*[*]

Instituto de Pesquisas Jardim Botânico do Rio de Janeiro
(Rua Pacheco Leão, 915, 22460-030 Rio de Janeiro, RJ, Brazil)
[*]gfilho@jbrj.gov.br.

## ABSTRACT

Because of its high toxicity, TBT (trybutiltin) was banned since 2003, which resulted in a greater re-use of Cu as based-biocide in antifouling paints (AFP). The aim of this work is to determine if metals form of AFP are transferred to benthic organisms from Guanabara Bay (GB) (Rio de Janeiro, Brazil). Metal concentrations were measured in two main fouling algae species *Ulva flexuosa* and *U. fasciata* and one isopod species, *Sphaeroma serratum*, in two GB marinas areas from sites with artificial substrate covered by AFP and natural substrate. In addition, control samples were collected in an adjacent open ocean area. Concentrations of Cd, Cr, Cu, Pb and Zn were determined by Atomic Absorption Spectrophotometry. Higher concentrations of Cu, Pb and Zn were detected in both algal species from GB in relation to control areas. Among samples of algae and isopod species from GB, populations collected over artificial surfaces covered by AFP presented significantly higher metal concentration than population of rocky natural substrate. Our data showed that the leaching of metals by antifouling paints present on decks and boats are being taken up by algae and isopods. These results indicate that antifouling coatings are the main source of heavy metal to biota of GB marina area.

## RESUMO

Devido sua alta toxicidade, o TBT está banido desde 2003, o que resultou na re-utilização de tintas a base de cobre. O objetivo deste trabalho é determinar se os metais provenientes das tintas anti-incrustantes (AFP) são transferidos para organismos bentônicos da Baía de Guanabara (BG) (Rio de janeiro, Brasil). Concentrações de metais foram analisadas em duas espécies de algas *Ulva flexuosa* e *U. fasciata* e no isópoda, *Sphaeroma serratum*, em duas áreas de marinas em locais de substrato artificial coberto com tintas AFP e em locais de substrato natural. Também foram coletadas amostras em uma área oceânica (controle). Concentrações de Cd, Cr, Cu, Pb e Zn foram determinadas por Espectrofotometria de Absorção Atômica. Concentrações mais elevadas de Cu, Pb e Zn foram detectadas na BG em ambas espécies de algas em relação a área controle. Dentre as espécies de algas e do isópoda da BG, as populações coletadas sobre as superfícies cobertas com AFP apresentaram concentrações significativamente mais elevadas do que as populações do substrato natural. Os resultados obtidos demonstram que a liberação de metais presentes nas AFP dos decks e embarcações, estão sendo acumulados pelas algas e isópodas. Esses resultados indicam que o revestimento com AFP é a principal fonte de metais para a biota de marinas em áreas da BG.

Descriptors: Antifouling paints, Cu, Metal accumulation, Benthic organisms, Guanabara Bay.
Descritores: Tintas anti-incrustantes, Cobre, Acumulação, Organismos bentônicos, Baía de Guanabara.

## INTRODUCTION

Antifouling paints are usually applied on boat hulls, ships and small vessels to prevent the growth of fouling organisms, including bacteria, macroalgae, mussels, barnacles and invertebrates. Most of antifouling coatings are based on self-polishment system that contains biocides, which are slowly released in the environment being toxic to fouling organisms (Boxall *et al.,* 2000; Callow & Callow, 2002). In the recent past, trybutyltin (TBT) was used as the main biocide in antifouling paints. Because of TBT-based antifouling paints present high toxicity, its use was banned since 2003 and regulations were implemented to phase out on all vessels by 2008 (IMO, 2001) resulting in a greater re-use of Cu-based antifouling paints (Warken *et al.,* 2004).

Trace metals have been identified as important constituents contaminating sediments in marinas and harbors. Where dissolved metals in water column are measured in marinas, they frequently

---

(1) Paper presented at the 1st Brazilian Congress of Marine Biology, on 15-19 May, 2006. Rio de Janeiro, Brazil.

exceed levels of concern (Valkirs *et al.,* 2003). Marina and harbor areas are inherently protected from strong water movement, thus providing calm water for navigation, but at the same time restrict water circulation. Copper is known to be high in sediments and waters of ports (Valkirs *et al.,* 2003; Schiff *et al.,* 2004) and marinas (e.g. Turner *et al.,* 1997; Guerra-García & García-Gómez, 2005).

A lot of studies have been developed in order to quantify and predict the metal fraction that is being leached by antifouling paints from boats to the environment. It is well documented, that higher Cu concentrations are found in locked marinas rather than in open water areas, and that Cu oxide is the most used biocide in the composition of these paints (Boxall *et al.,* 2000; Schiff *et al.,* 2004; Guerra-García & García-Gómez, 2005). An increase in Cu contents in oyster populations at Arcachon Bay, were determined by Claisse & Alzieu (1993), since the TBT use was phased-out in France. Although Cu is important as a micronutrient for algal growth, it is highly toxic at elevated concentrations in the ionic forms (Gledhill *et al.,* 1997).

As result of several characteristics, *Ulva* species (Ulvophyceae) are widely used as biomonitors to assess metal concentration in aquatic environments. These include its sedentary and cosmopolitan nature, its abundant populations, and its high capability to accumulate Cu concentrations (Ho *et al.,* 1990; Riquelme *et al.,* 1997; Correa *et al.,* 1999; Haritonidis & Malea, 1999; Andrade *et al.,* 2004). The genus *Ulva* can tolerate lower and medium Cu concentrations, showing high capacity to metal accumulation, due to mechanisms of cellular immobilization (Andrade *et al.,* 2004).

Invertebrates are generally more sensitive to pollutants than either fish or algae. Among them, amphipods, isopods and decapods are important components of the marine intertidal and subtidal fauna (Levent *et al.,* 1999). Isopods live among algal turfs, crevices or under the stones, are omnivorous, feeding on bacterial biofilms, algae or/and organic detritus (Levent *et al.,* 1999). The isopod *S. serratum* that lives among *Ulva* turfs, present a large range of geographical distribution in the Atlantic Ocean and is an abundant species in marinas and harbor areas.

The aim of this work is to assess the concentrations of metals in two main fouling algal species, *Ulva flexuosa* and *Ulva fasciata*, and one isopod species *Sphaeroma serratum* (Sphaeromatidae) in marina areas at Guanabara Bay (Rio de Janeiro, Brazil) in order to determine the contribution of antifouling paints to metal accumulation by these organisms. In this way, samples were collected in three sites, two into the Bay and one in an open water rocky shore.

## MATERIAL AND METHODS

Samples, including the two seaweeds (*U. flexuosa* and *U. fasciata*) were collected at the intertidal zone in two sites at Guanabara Bay (GB), Marina da Gloria (MG) and Ribeira (RI) in winter (August 2004) and summer (February 2005). For comparative purpose, samples of the seaweed species were collected at Arpoador Beach (AB) (City of Rio de Janeiro, Brazil), an open ocean area. Populations of the epi-faunal isopod *S. serratum* were found only at the MG site and the individuals were hand collected. Samples were obtained in both natural (RS) and artificial substrate (DECK). Algal samples were collected using a stainless scissor in order to remove the basal portions (a mean distance of 5 mm from substrate surface). Samples were transported in local seawater to laboratory.

In the laboratory samples were washed in seawater, therefore washed in distilled water and dried at (60ºC, 48h). Samples were weighed and approximately 250 mg (triplicates) of each sample were ashed (450ºC, 48h) and digested in 10 ml $HNO_3$ (Merck PA 65%). After evaporation the residue was re-dissolved in 0.1N HCl (15 ml) and stored in polyethylene bottles until analyses. Five trace metals were measured Cd, Cu, Cr, Pb and Zn by flame atomic absorption spectrophotometry (AAS-Varian AA-147). The results were expressed in $\mu g.g^{-1}$ of dry weight. One common Brazilian antifouling paint (AFP) (RENNER AF 10) used on commercial and military vessels was submitted to analyses (for Cd, Cr, Cu, Pb and Zn), in this case, samples weighed approximately 100 mg (triplicates). Analytical procedures were tested by comparative analyses of International Atomic Energy Agency (IAEA) certified reference material IAEA-140 (sea plant homogenate, *Fucus*). One-Way Analysis of Variance (ANOVA) was used to compare metal concentrations among sampling sites and natural and artificial substrate. Differences were considered significant when $p < 0.05$ (STATISCA 4.2).

## RESULTS

Among the five analyzed metals, concentrations of Cu, Pb and Zn were significantly higher in algae from the GB sites than AB ones ($n = 60$, $p < 0.05$). The Cu concentration in GB seaweed samples varied between 16.95 ± 7.78 $\mu g.g^{-1}$ and 152.32 ± 16.32 $\mu g.g^{-1}$; the highest value (152.32 ± 16.07 $\mu g.g^{-1}$) was observed in *U. flexuosa* samples of MG DECK (twenty-seven times higher than the value of AB, 5.34 ± 1.74 $\mu g.g^{-1}$) and MG DECK concentration is still significantly higher ($n = 9$, $p < 0.05$) than concentration observed in *U. flexuosa* populations from the two other GB sampled sites (MG

RS samples = 19.41 ± 1.47 μg.g$^{-1}$ and RI RS samples = 18.30 ± 3.40 μg.g$^{-1}$).

Levels of Pb in seaweeds from GB samples varied between 8.04 ± 1.75 μg.g$^{-1}$ and 27.68 ± 8.05 μg.g$^{-1}$; the highest value was found in *U. flexuosa* samples from MG DECK (six times higher than the value of AB, 4.60 ± 0.50 μg.g$^{-1}$). In the same way of the Cu results, *U. flexuosa* MG DECK Pb concentration was significantly higher ($n = 9$ $p < 0.05$) than that of samples from the two other GB sites (MG RS, 16.60 ± 3.3 and RI RS, 15.40 ± 3.25 μg.g$^{-1}$ ). At MG RS, *U. flexuosa* samples showed significantly ($n = 6$, $p < 0.05$) higher Pb concentration than *U. fasciata* (Table 1).

Levels of Zn varied between 27.44 ± 1.67 μg.g$^{-1}$ to 174.37 ± 32.79 μg.g$^{-1}$ in GB samples; the highest concentration was observed in *U. flexuosa* (MG DECK) and it was seven times higher than in AB samples (24.76 ± 4.79 μg.g$^{-1}$) and yet, it was significantly higher ($n = 9$ $p < 0.05$) than the concentration in the two other GB populations (MG RS, 68.88 ± 0.01 μg.g$^{-1}$and RI RS, 26.47 ± 1.42 μg.g$^{1}$).

Concentration of Cd and Cr in seaweeds samples from GB are in the same range of that observed in samples from the open water collecting site (Table 2). No difference in metal concentration was found between organisms collected in the two distinct seasons (Table 1). Comparing the mean concentration of Cu, Pb and Zn in algal population from MG DECK it was observed that *U. flexuosa* presented significantly higher Cu and Zn concentration ($n = 18$, $p < 0.05$) than *U. fasciata* samples (Fig. 1).

Table 1. Copper, lead and zinc concentrations (μg.g$^{-1}$ dry weight) in the three species (*Ulva flexuosa*, *U. fasciata* and *Sphaeroma serratum*) at the sampled sites of Guanabara Bay (MG DECK= Marina da Glória deck; MG RS= Marina da Glória rocky shore; RI RS= Ribeira rocky shore) and at the control area (Arpoador beach). Mean values and standard deviation (in italic).

| **Species/ Samples sites** | **Metals/Seasons** | | | | | |
|---|---|---|---|---|---|---|
| | **Cu** | | **Pb** | | **Zn** | |
| | **summer** | **winter** | **summer** | **winter** | **summer** | **winter** |
| *U. flexuosa* | | | | | | |
| **MG DECK** | 144.26 | 152.32 | 27.68 | 27.40 | 174.37 | 172.42 |
| | *15.21* | *16.07* | *8.05* | *7.5* | *32.79* | *31.47* |
| **MG RS** | 19.41 | 19.39 | 15.99 | 16.60 | 42.54 | 68.88 |
| | *1.47* | *0.86* | *2.38* | *3.3* | *5.08* | *0.01* |
| **RI RS** | 17.25 | 18.30 | 13.33 | 15.40 | 27.44 | 26.47 |
| | *2.8* | *3.4* | *3.36* | *3.25* | *1.67* | *1.42* |
| **AB** | 5.79 | 5.40 | 4.51 | 4.60 | 24.76 | 23. 40 |
| | *0.80* | *0.75* | *0.48* | *0.50* | *4.79* | *4.60* |
| *U. fasciata* | | | | | | |
| **MG DECK** | 27.83 | 26.75 | 27.01 | 27.30 | 45.62 | 44.35 |
| | *3.35* | *3.34* | *8.05* | *7.5* | *3.83* | *2.08* |
| **MG RS** | 21.09 | 13.41 | 9.02 | 9.82 | 35.5 | 34.5 |
| | *1.64* | *2.8* | *1.75* | *4.45* | *0.83* | *0.90* |
| **RI RS** | 16.95 | 21.05 | 8.04 | 10.05 | 34.21 | 32.45 |
| | *7.78* | *4.45* | *1.75* | *2.85* | *0,75* | *0.50* |
| **AB** | 5.40 | 5.34 | 2.81 | 2.70 | 14.97 | 15.14 |
| | *1.60* | *1.74* | *0.45* | *0.42* | *5.54* | *4.35* |
| *S.serratum* | | | | | | |
| **MG DECK** | 200.29 | 199.05 | 17.05 | 18.05 | 230.00 | 225.04 |
| | *4.48* | *3.3* | *0.48* | *0.55* | *11.06* | *12.45* |
| **MG RS** | 119.95 | 120.30 | 6.66 | 6.72 | 128.28 | 122.28 |
| | *7.65* | *6.8* | *0.33* | *0.35* | *13.01* | *12.01* |

Table 2. Cadmium and chromium mean concentration ($\mu g.g^{-1}$ dry weight ± standard deviation) in three species from GB (Guanabara Bay) and AB (Arpoador Beach - Control area) sample sites. ND means below the limit of detection.

| **Sites/species** | **Metals** | |
|---|---|---|
| | **Cd** | **Cr** |
| GB | | |
| *U. flexuosa* | 0.66 | 16.00 |
| | 0.07 | 2.85 |
| *U. fasciata* | 0.49 | 12.61 |
| | 0.13 | 0.93 |
| *S. serratum* | ND | ND |
| AB | | |
| *U. flexuosa* | 0.50 | 15.00 |
| | 0.05 | 2.4 |
| *U. fasciata* | 0.71 | 13.9 |
| | 0.11 | 3.6 |

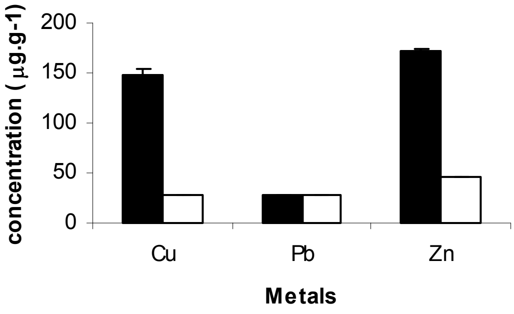

Fig. 1. Mean (± standard deviation) metal concentration in *U.flexuosa* (black columm) and *U.fasciata* (white columm) from MG DECK sample site.

In relation to the isopod *S. serratum*, significantly higher concentrations of Cu, Pb and Zn were observed in the population from MG DECK (Cu = 200.29 ± 4.48 $\mu g.g^{-1}$; Pb = 18 ± 0.55 $\mu g.g^{-1}$ and Zn = 230 ± 11.06 $\mu g.g^{-1}$) than RS (Cu = 120.30 ± 6.8 $\mu g.g^{-1}$; Pb = 6.72 ± 0.35 $\mu g.g^{-1}$ and Zn = 128.28 ± 13.01 $\mu g.g^{-1}$), that means differences in concentration of 1.6 (Cu), 2.6 (Pb) and 1.8 (Zn) times between MG DECK and RS. (Table 1). Concentrations of Cd and Cr in *S. serratum* were below the analytical limit of detection.

Among the five metals analyzed in the antifouling paint sample, Cu and Zn presented the highest concentrations, respectively 324,785 ± 57,000 $\mu g.g^{-1}$ and 145,232 ± 12,401. The following metal concentration in AFP (RENNER AF 10) samples were in order to Cu > Zn > Pb > Cr > Cd. The results showed that Cu and Zn are the main metal elements present in this type of paint (Table 3).

Table 3. Mean metal concentration ($\mu g.g^{-1}$ dry weight ± standard deviation) in antifouling paint RENNER AF10.

| Metals | Concentration |
|---|---|
| Cd | 3.59 ± 0.40 |
| Cr | 16.84 ± 2.59 |
| Cu | 324,785 ± 57,000 |
| Pb | 247.63 ± 37.53 |
| Zn | 145,232 ± 12,401 |

## DISCUSSION

Copper loading from antifouling surfaces can be a substantial portion of the total loading of Cu into a harbor or estuary (Valkirs *et al.,* 2003). The rank order in terms of number of vessels treated with a particular biocide was Cu (I) oxide > diuron >Cu thiocyanate > Irgarol 1051 and Zn pyrythione (Boxall *et al.,* 2000). Our results indicated that the main biocide present in AFP paint analyzed was Cu. Dissolved Cu is the second most toxic element after mercury (Gledhill *et al.,* 1997), and presented a profound effect on the environment, principally on the initial-stage of growth in several organisms (Andrade *et al.,* 2004). Our results showed high amount of Cu and Zn in AFP (RENNER AF 10) formula and this observed pattern of Cu and Zn concentration is found in the majorority of AFP paints compositions (Yonerara *et al.,* 2001; Callow & Callow, 2002; Thouvenin *et al.,* 2002; Valkirs *et al.,* 2003; Löschau *et al.*, 2005).

In the present study, heavy metal concentrations in organisms were in the following order Zn > Cu > Pb > Cr > Cd. Cadmium and chromium in seaweeds presented the same metal levels found in non-contaminated waters (Amado Filho *et al.,* 1999). Our data showed higher Cu, Pb and Zn concentrations in algae populations from GB sites than AB. Comparisons among the present data with previous studies of metal concentration in GB far from marina areas (Carvalho *et al.,* 1992; Karez *et al.,* 1994) showed that samples collected in marina sites (present study) were higher for Cu, Pb and Zn. Marina areas are generally protected against strong water movement, which restricts the water circulation and contributing to the metal bioavailability (Valkirs *et al.,* 2003). Although differences between GB and control area were seen, Moore & Ramamurti (1987) considered that in general Zn levels in benthic algae from non-polluted areas do not exceed 100 $\mu g.g^{-1}$. Only in MG DECK Zn levels exceeded the background level of 100 $\mu g.g^{-1}$, suggesting that antifouling paints are the main source of Zn to marina and yacht club areas. Copper based antifouling paints regularly contain Zn (as ZnO) that is normally used as booster biocide for Cu, increasing the toxicity of Cu by 200 times (Waterman *et al.,* 2005). Algae populations from MG DECK (which were located just a few meters from RS site), presented higher Cu concentration than RS and RI populations, indicating that Cu present in antifouling paints are being transferred to these populations. Ambient concentrations of dissolved ($Cu^{++}$) in sea water are very low, averaging 4nM in waters with salinity over 35% (Gledhill *et al.,* 1997). Cuprous ion ($Cu^{+}$) released from Cu-based coatings is rapidly converted to cupric ion ($Cu^{++}$) and then more slowly forms inorganic and organic complexes (Valkirs *et al.,* 2003). In this way, the higher concentrations found in samples from MG DECK and the metal concentrations decrease along a short distance gradient between MG DECK and MG RS should be related to the proximity of the Cu source and availability in seawater. In the same way of Cu and Zn, Pb in seaweeds samples from MG DECK was higher than RS, showing the contribution of antifouling paints as a source of Pb.

Reed & Moffat (1983) shown that different ecotypes of *U. compressa* presented different Cu tolerance (ship fouling algae presented higher metal concentration than non-fouling populations). Similar results were observed in our data, where population from surface covered by antifouling paints revealed higher metal concentration than populations from natural substrates. The higher Cu and Zn concentrations in *U. flexuosa* than *U. fasciata* confirm the capability of *U. flexuosa* to accumulate and immobilize metals (Andrade *at al.,* 2004), which explain why this species is considered the major fouling organism commonly found adhering to boat hulls protected with antifouling paints (Callow & Callow, 2002).

Like algal samples the isopod *S. serratum* presented higher Cu and Zn concentrations in DECK samples than in rocky shore ones, indicating that the same pattern of metal levels found in algae were seen. Populations that were located over antifouling paint surface into algal turfs showed higher metal concentration than natural substrate populations. Probably the saprophagous and omnivorous food habits of isopods species (Rupert *et al*., 2005) contributed to the high Cu, Zn and Pb levels found in marina area. In sites where the food contains high concentrations of cooper, the isopods assimilate metals in excess of their requirements, storing the excess of Cu within insoluble granules associated with sulphur and calcium, in the S cells of the hepatopancreas (Wieser, 1967; Donadey & Besse, 1972; Hopkin & Martin, 1982).

Finally, our data showed that the leaching of metals by antifouling paints present on decks and boats are being taken up by algae and isopods. These results indicate that antifouling coatings are the main source of heavy metal to marina areas. The utilization of Cu based formulations as a substitute for TBT did not avoid the negative effect of antifouling paints released to marine ecosystems, especially benthic communities of marina and harbor surrounding areas, claiming to new biomonitoring studies in this field, concomitant with efforts of research groups around the world on the development of antifouling paints free of toxic biocides.

## ACKNOWLEDGMENTS

The authors thank the director of Marina da Glória, Dr. Sérgio Ricardo M. Meirelles by the access and support for sampling into the establishment; the Captain of Material Division (Brazil Navy) Msc. Dauton L. F. Menezes for providing antifouling paints; Dr. Cristiana S. Serejo (Museu Nacional/UFRJ) and Dr. Elaine Albuquerque Figueiredo (Universidade Santa Úrsula) by the isopods identification and Ricardo Thomas for AAS measurements. This work was supported by CNPq (Edita Universal 2004, research grants to G.M. Amado Filho - Produtividade em Pesquisa/Oceanografia, and W. C. Paradas, Iniciação Científica, quota/ Oceanografia).

## REFERENCES

Amado Filho, G. M.; Andrade, L. R.; Karez, C. S.; Farina, M. & Pfeiffer, W. C. 1999. Brown algae species as biomonitors of Zn and Cd at Sepetiba Bay, Rio de Janeiro, Brazil. Mar. Environ. Res., 48:213-224.

Andrade, L. R.; Farina, M. & Amado Filho, G. M. 2004. Effects of copper on *Ulva flexuosa* (Chlorophyta) in vitro. Ecotoxicol. Environ. Saf., 58:117-125.

Boxall, A. B. A.; Comber, S. D.; Conrad, A. U.; Howcroft, J. & Zaman, N. 2000. Inputs, monitoring and fate modeling of antifouling biocides in UK Estuaries. Mar. Pollut. Bull., 40(11):898-905.

Callow, M. E. & Callow, J. A. 2002. Marine biofouling: a sticky problem. Biologist 49(1):1-5.

Claise, D. & Alsieu, C. L. 1993. Copper contaminations as a result of antifouling paints regulation? Mar. Pollut. Bull., 26(7):395-397.

Carvalho, C. E. & Lacerda, L. D. 1992. Heavy metals in the Guanabara Bay biota: why such low concentrations? Ci. Cult. J. Ass. Bras. Adv. Sci., 44:184-186.

Correa, J. A.; Gonzáles, P.; Sánchez, P.; Munõz, J. & Orellana, M. C. 1996. Copper-algae interactions inheritance or adaptation? Environ. Monit. Assessment., 40:41-54.

Donadey, C. & Besse, G. 1972. Etude histologique, ultrastructurale et experimentale dês caecums digestifs de *Porcellio dilatatus* et *Ligia oceânica* (Crustaceae, isopoda). Tethys, 4:145-62.

Gledhill, M.; Nimno, M.; Hill, S. J. & Brown, T. M. 1997. The toxicity of copper (II) species to marine algae, with particular reference to macroalgae. J.Phycol., 33: 2-11.

Guerra-García, J. M. & García-Gómez, J. C. 2005. Assessing pollution levels in sediments of a harbor with two opposing entrances. Environmental implications. J. Environ. Mgnt., 77:1-11.

Haritonidis, S. & Malea, P. 1999. Bioaccumulation of metals by the green alga *Ulva rigida* from Thermaikos Gulf, Greece. Environ. Pollut., 104:365-372.

Ho, Y. 1987. Metals in 19 intertidal macroalgae in Hong Kong waters. Mar. Pollut. Bull., 18:564-566.

Hopkin, S. P. & Martin, M. H. 1982. The distribution of zinc, cadmium, lead and copper within woodlouse *Oniscus asellus* (Crustaceae, Isopoda). Oecologia., 54: 227-32.

IMO - International Maritime Organization. 2001. International Conference on the control of Harmful Anti-fouling Systems on Ships. London: IMO Headquartes.

Karez, C. S.; Amado Filho, G. M.; Moll, D. M. & Pfeiffer, W. C. 1994. Concentrações de metais em algas marinhas bentônicas de três regiões do Estado do Rio de Janeiro. An. Acad. bras. Ci., 66 (2):205-211.

Levent, B. A. T.; Ayse, G.; Murat, S.; Mehmet, Ç.; Gamze, G. & Mehmet, A. 1999. Acute toxicity of zinc, copper and lead to three species of marine organisms from the Sinop Peninsula, Black sea. Turkish. J. Biol., 23:537-544.

Löschau, M. & Krätke, R. 2005. Efficacy and toxicity of self-polishing biocide-free antifouling paints. Environ. Pollut., 138(2):260-267.

Moore, J. V. & Ramamurti, S. 1987. Heavy Metals in near-bottom water. Moscow. 285p.

Reed, R. H. & Moffat, L. 1983. Copper toxicity and copper tolerance in *Ulva compressa* (L.) Grev. J. expl. mar. Biol. Ecol., 69: 85-103.

Riquelme, C.; Rojas, A.; Flores, V. & Correa, J. A. 1997. Epiphytic bacteria in a copper-enriched environment in Northern Chile. Mar. Pollut. Bull., 34:816-820.

Ruppert, E. E.; Fox, R. S. & Barnes, R. D. 2005. Zoologia dos vertebrados: uma abordagem funcional – evolutiva. 7. ed. São Paulo. Rocca, 1.145p.

Schiff, K.; Diehl, D. & Valkirs, A. O. 2004. Copper emissions from antifouling paint on recreational vessels. Mar. Pollut. Bull., 48: 371-377.

Turner, S. J.; Cummings, V. J.; Hewitt, J. E.; Wilkinson, M. R.; Williamson, R. B. & Lee, D. J. Changes in epifaunal assemblages in response to marina operations and boating activities. Mar. environ. Res., 43:181-199.

Thouvenin, M.; Peron, J. J.; Charrateurm, C.; Guerin, P.; Langlois, J. Y. & Vallee-Rehel, K. 2002. A study of the biocide release from antifouling paints. Progr. Org. Coat., 44:75-83.

Valkirs, A. O.; Seligman, P. F.; Haslbeck, E. & Caso, J. S. 2003. Measurement of copper release rates from antifouling paint under laboratory and in situ conditions: implications for loading estimation to marine water bodies. Mar. Pollut. Bull., 46:763-779.

Warken, J.; Dunn, R. J. K. & Teasdale, P. R. 2004. Investigation of recreational boats as a source of copper at anchorage sites using time-integrated diffusive gradients in thin film and sediments measurements. Mar. Pollut. Bull., 49:833-843.

Waterman, B. T.; Daehne, B.; Sievers, S.; Dannenberg, R.; Overbeke, J. C.; Klijnstra, J. W. & Heemken, O. 2005. Bioassays and selected chemical analysis of biocide-free antifouling coatings. Chem., 60:1530-1541.

Wieser, W. 1967. Conquering terra firma: The copper problem from the isopod´s point of view. Helgoländer. Wiss. Meeresunter., 15:282-93.

Yenehara, Y.; Yamashita, H.; Kawakamura, C. & Itoh, K. 2001. A new antifouling paint base don a zinc acrylate copolymerer. Prog. Org. Coat., 42:150-158.

*(Manuscript received 09 Juner 2005; revised 06 October 2006; accepted 18 October 2006)*