

Accumulation of Copper and Zinc by *Balanus amphitrite* in a Tropical Estuary

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Balanus amphitrite (Cirripedia: Thoracica) a dominant fouling organism was analysed to assess the accumulation potentialities of copper and zinc. It was observed that it could accumulate Cu to 864.77 $\mu\text{g g}^{-1}$ dry wt. and Zn to 1937.50 $\mu\text{g g}^{-1}$ dry wt. The concentration of these two metals in the surrounding water was 0.001-0.011 mg l^{-1} for copper and 0.013-0.046 mg l^{-1} for zinc. The concentration factor of these two metals varied during different periods of the year and also with the size range of the animal.

Some organisms can accumulate high concentrations of trace metals. They may also represent moving time-averaged values for the relative biological availability of metals at each site (Phillips, 1977). Barnacles, an important group of fouling organism, have been studied recently for their potentialities as an indicator organism (Ireland, 1973, 1974; Walker *et al.*, 1975; Barbaro *et al.*, 1978; Barber & Trefry, 1981; Udayabhaskar *et al.*, 1983). However, the information available on this group of animals is very scanty and studies to date have not fully developed the indicator capabilities of barnacle species (Phillips, 1980). Keeping these aspects in view, investigations have been carried out on the bioaccumulation capabilities of barnacle *Balanus amphitrite* (Cirripedia: Thoracica) with respect to Cu and Zn.

Material and Methods

Balanus amphitrite samples were collected from the panels used for studying the biofouling community structure at the Shipyard and Harbour stations of the Zuari estuary (Anil, 1986). This estuary is Lat. 15° 25' N and Long. 73° 25' E on the west coast of India. The barnacles were grouped in 0-1 and 1-2 cm size groups based on the rostro-carinal diameter of the basis. On removal of the external detritus from the shell, they were rinsed with distilled water, the soft parts were

removed and dried to constant weight in an oven. The dried samples were digested with concentrated nitric acid and phosphoric acid for the analysis of the copper and zinc concentration by Atomic Absorption Spectrophotometry (Perkin-Elmer Model 5000). The calculated metal content is represented as $\mu\text{g g}^{-1}$ dry wt. of the whole soft part. The concentration of copper and zinc in water was also estimated by AAS. Metal content in the dissolved phase was preconcentrated with APDC/MIBK extraction method. The 0.45 μm millipore filter containing particulate matter was digested with 5 ml of 6N hydrochloric acid and 15 ml of acetone was added to dissolve the filter paper and form a single phase for further analysis with the AAS. The metal content in the dissolved and the particulate phase were added to get the total metal concentration and is expressed in terms of mg l^{-1} . The concentration factor for each metal has been calculated as the ratio of mean concentration ($\mu\text{g g}^{-1}$) in dry tissue to mean total concentration (mg l^{-1}) in water during the period considered. The seasonal averages of water concentration were taken into consideration for calculating the concentration factor of the 1-2 cm size group since they could have been in the environment for more than one month. For 0-1 cm barnacles, the concentration of metal in water for that particular month was taken into consideration, although the concentration did not vary much during each season.

Results

Copper

The total copper concentration (dissolved + particulate) in the surrounding water varied between 0.001 and 0.011 mg l^{-1} . The monthly averages indicated that the maximum monthly average concentration was 0.011 mg l^{-1} in August at the shipyard station and 0.009 mg l^{-1} at the harbour in July 1983 (Table 1). The minimum total concentration was 0.002 mg l^{-1} at the shipyard in May 1983 and 0.001 mg l^{-1} at the harbour in May 1984).

The maximum concentration of this metal in *Balanus* in the 0–1 cm group was $864.77 \mu\text{g g}^{-1}$ (dry wt.) whereas the maximum in the 1–2 cm group was $625.67 \mu\text{g g}^{-1}$. Both of the above samples were collected from harbour station in May 1984. In May 1984, the maximum accumulation of copper at the shipyard was $602.85 \mu\text{g g}^{-1}$ for the 0–1 cm group and $484.37 \mu\text{g g}^{-1}$ for the 1–2 cm group collected also in May 1984. The samples (0–1 cm) collected during September 1983 had the lowest concentration of this metal ($47.28 \mu\text{g g}^{-1}$ at the shipyard and $48.38 \mu\text{g g}^{-1}$ at the harbour); for the 1–2 cm group the lowest values were $39.70 \mu\text{g g}^{-1}$ for a sample collected at the shipyard in April 1983 and $43.40 \mu\text{g g}^{-1}$ for the sample collected at the harbour in August 1983. The 0–1 cm size group animals exhibited higher concentration of this metal than 1–2 cm size group animals, except for the observations made during March 1984.

The concentration factor calculated ranged from 7.06×10^3 to 384.32×10^3 at these two stations. The concentration factor in the animals belonging to 1–2 cm size group was lower than those belonging to the 0–1 cm size group, except in the samples obtained during March 1984, when the 1–2 cm size group exhibited higher concentration capabilities.

Zinc

The concentration in water was $0.013\text{--}0.046 \text{ mg l}^{-1}$. It varied $0.015\text{--}0.04 \text{ mg l}^{-1}$ at the shipyard and $0.013\text{--}0.046 \text{ mg l}^{-1}$ at the harbour. The maximum total concentration of this metal was observed during July and the minimum during February 1984. The seasonal averages indicated that the total concentration of zinc was almost uniform in all the seasons and ranged $0.018\text{--}0.027 \text{ mg l}^{-1}$.

The accumulation of zinc by *Balanus* ranged between 203.83 and $1469.57 \mu\text{g g}^{-1}$ at the shipyard and 203.64 to $1937.50 \mu\text{g g}^{-1}$ at the harbour for the 0–1 cm group. The 1–2 cm group exhibited a range of $204.26\text{--}910 \mu\text{g g}^{-1}$ at the shipyard and $402\text{--}1588.68 \mu\text{g g}^{-1}$ at the harbour. During January and May, 1984 the mean concentration was higher for the 0–1 cm group. During September 1983, March and April 1984 the 1–2 cm group had higher concentrations. During August 1983 both the groups had almost similar range of zinc concentrations (Table 2).

The concentration factor of zinc ranged between 10.66×10^3 and 84.60×10^3 for 0–1 cm group and 19.50×10^3 to 49.87×10^3 for 1–2 cm group. The concentration factor was found to be higher in the 0–1 cm

TABLE 1
Concentration and concentration factor of copper in *B. amphitrite*.

Month	Station	Size group (0–1 cm) conc. $\mu\text{g g}^{-1}$	Total water conc. mg l^{-1}	Conc. factor ($\times 10^3$)	Size group (1–2 cm) conc. $\mu\text{g g}^{-1}$	Total water conc. mg l^{-1}	Conc. factor ($\times 10^3$)
March 83	Shipyard	151.02 (136–165.50)	0.003	60.41	118.05	0.003	39.60
April 83	Harbour	106.89	0.003	40.18	–	–	–
	Shipyard	–	–	–	52.18 (39.70–82.07)	0.003	17.39
May 83	Harbour	–	–	–	–	–	–
	Shipyard	–	–	–	86.71 (62.37–147.40)	0.003	28.90
July 83	Harbour	–	–	–	–	–	–
	Shipyard	–	–	–	–	–	–
August 83	Harbour	66.97 (52.08–72.85)	0.009	7.12	–	–	–
	Shipyard	154.17 (140.64–167.71)	0.011	13.77	–	–	–
September 83	Harbour	68 (82.61–90.71)	0.008	11.55	65.03 (43.40–97.68)	0.006	10.16
	Shipyard	47.28	0.007	7.06	–	–	–
December 83	Harbour	68 (48.38–96.62)	0.005	13.60	58.83 (43.40–97.68)	0.006	9.19
	Shipyard	134.90 (77.97–158.72)	0.003	39.68	–	–	–
January 84	Harbour	113.03 (75.75–116.66)	0.004	28.98	75.48 (69.44–78.36)	0.006	13.72
	Shipyard	512.11	–	131.30	257.15 (125.92–432.85)	0.005	48.52
February 84	Harbour	389.57	0.004	99.89	–	–	–
	Shipyard	–	–	–	189.46 (144.14–213.83)	0.003	63.15
March 84	Harbour	–	–	–	198.46	0.003	70.88
	Shipyard	138.86 (138.13–139.56)	0.003	43.39	213.83	0.003	71.28
April 84	Harbour	–	–	–	216.96	0.003	77.49
	Shipyard	221.10	0.002	96.13	147.00 (111.23–192.58)	0.003	49.00
May 84	Harbour	178.70 (119.11–276.74)	0.003	63.82	159.27 (101.55–251.09)	0.003	56.88
	Shipyard	601.85	0.002	261.67	323.38 (201.06–484.37)	0.003	107.79
	Harbour	499.61 (217.56–864.77)	0.001	384.31	309.95 (192.74–625.67)	0.003	110.70

size group, except for the samples collected in March and April 1984.

Discussion

The concentration of copper in coastal waters ranges from 0.4 to 107 $\mu\text{g l}^{-1}$ (Phillips, 1977). In the present investigation it was found that the concentration of copper was 0.001–0.011 mg l^{-1} . The earlier observations carried out in the months of April and May 1985 for estuarine waters around Goa by Zingde *et al.* (1976) indicated concentration values of 0.003–0.005 mg l^{-1} for copper. The concentration of zinc in the present investigations ranged between 0.013 and 0.046 mg l^{-1} . It has been observed earlier that the concentration of zinc may reach 3560 $\mu\text{g l}^{-1}$ (Stenner & Nickless, 1974).

Total concentration of the metal in water (dissolved + total particulate) has been taken into consideration as the available source of metal for the animal in the present investigation since it is a filter feeder. It has been observed earlier that bivalves which are also filter feeders have capabilities of obtaining trace metals not only from solution but also from the ingestion of inorganic particulate material (Moore, 1971). Barbaro *et al.* (1978) also noted that suspended

particles may also be an important source of copper to barnacles.

Comparative data on accumulation of Cu and Zn by the barnacle is given in Table 3. The accumulation of copper by *B. amphitrite* in the present investigation ranged between 47.28–864.77 $\mu\text{g g}^{-1}$. Earlier it was observed that *Balanus balanoides* could accumulate up to 3750 $\mu\text{g g}^{-1}$ (Walker, 1977) however for *Balanus amphitrite* the maximum accumulation of this metal was found to be 600 $\mu\text{g g}^{-1}$ (Stenner & Nickless, 1975). In case of zinc, the maximum accumulation was found up to 23 000 $\mu\text{g g}^{-1}$ by Ireland (1973) for *Balanus balanoides* and 3300 $\mu\text{g g}^{-1}$ for *Balanus amphitrite* (Stenner & Nickless, 1975).

In the present investigation the accumulation of zinc was found to a maximum of 1937.50 $\mu\text{g g}^{-1}$. The concentration factor of copper during the present studies ranged from 7.06×10^3 to 38.43×10^4 . This is substantially higher than the concentration factors observed earlier for *Balanus amphitrite* at Venice and Grado 1.5×10^3 , and 5×10^3 at Rio Tinto (Stenner & Nickless, 1975). Under laboratory conditions *Balanus eburneus* accumulates copper to a concentration of 1090 $\mu\text{g g}^{-1}$ (dry wt.) in 15 days when exposed to 140 $\mu\text{g l}^{-1}$ cupric citrate i.e., a concentration factor of 7.78×10^3 (Clark,

TABLE 2
Concentration and concentration factor of zinc in *B. amphitrite*.

Month	Station	Size group (0–1 cm) conc. $\mu\text{g g}^{-1}$	Total water conc. mg l^{-1}	Conc. factor ($\times 10^3$)	Size group (1–2 cm) conc. $\mu\text{g g}^{-1}$	Total water conc. mg l^{-1}	Conc. factor ($\times 10^3$)
March 83	Shipyards	325.56 (244.93–406.18)	0.024	13.40	–	–	–
	Harbour	935.56 (876.26–994.80)	0.396	23.64	–	–	–
April 83	Shipyards	–	–	–	347.14 (204.26–589.62)	0.018	19.50
	Harbour	–	–	–	–	–	–
May 83	Shipyards	–	–	–	433.60 (275.07–541.11)	0.018	24.36
	Harbour	–	–	–	–	–	–
July 83	Shipyards	–	–	–	–	–	–
	Harbour	933.23 (767.46–1035.62)	0.046	20.38	–	–	–
August 83	Shipyards	–	–	–	–	–	–
	Harbour	967.35 (830.44–1081.44)	0.017	58.27	967.84 (763.13–1177.08)	0.025	39.39
September 83	Shipyards	–	–	–	–	–	–
	Harbour	706.84 (381.64–915.46)	0.019	37.75	835.15 (493.33–1151.53)	0.025	33.95
December 83	Shipyards	1226.76 (867.19–1469.57)	0.015	84.60	–	–	–
	Harbour	1062.77 (819.83–1310.16)	0.016	65.20	1117.08 (814.65–1588.68)	0.022	49.87
January 84	Shipyards	669.04	–	29.60	533.70 (125.92–432.85)	0.022	23.93
	Harbour	1750	0.023	77.43	–	–	–
February 84	Shipyards	309.56 (203.88–415.24)	0.016	19.84	–	–	–
	Harbour	347.75	0.013	26.15	–	–	–
March 84	Shipyards	290.03 (244.41–335.45)	0.027	10.66	574.17	0.022	25.98
	Harbour	–	–	–	842.08	0.023	36.77
April 84	Shipyards	507.51	0.028	18.45	669.07 (409.57–910.00)	0.022	30.27
	Harbour	474.39 (203.66–784.38)	0.023	20.45	–	–	–
May 84	Shipyards	–	–	–	458.73 (385.54–561.49)	0.022	20.76
	Harbour	1647.18 (1468.23–1937.50)	0.020	81.95	662.91 (402.00–1189.49)	0.023	28.95

TABLE 3
Comparative data on accumulation of copper and zinc in barnacle.

Species	Locality	Metal	Metal concentration	Reference
<i>Balanus balanoides</i>	Aberystwyth (Wales)	Zn	4.5–23.1 mg g ⁻¹ dry wt.	Ireland, 1973
<i>Balanus balanoides</i>	Cardigan Bay (Wales)	Cu	0.1–0.7 mg g ⁻¹ dry wt.	Ireland, 1974
		Zn	3.0–28.0 mg g ⁻¹ dry wt.	
<i>Balanus balanoides</i>	Menai Strait (Wales)	Zn	1.03 mg g ⁻¹ wet wt.	Walker <i>et al.</i> , 1975
<i>Balanus balanoides</i>	Allwen	Zn	1.77 mg g ⁻¹ wet wt.	
	Castle Point	Zn	2.10 mg g ⁻¹ wet wt.	
	R–Yetwyth	Zn	3.44 mg g ⁻¹ wet wt.	
<i>Balanus balanoides</i>	Dulas Bay (Anglesey, Wales)	Cu	3.75 mg g ⁻¹ dry wt.	Walker, 1977
		Zn	50.28 mg g ⁻¹ dry wt.	
<i>Chthamalus stellatus</i>	Coast of S.W. Spain and Portugal	Cu	6.3–11.3 µg g ⁻¹ dry wt.	Stenner & Nickless, 1975
		Zn	100–237 µg g ⁻¹ dry wt.	
<i>Balanus perforatus</i>	Coast of S.W. Spain and Portugal	Cu	8–8.5 µg g ⁻¹ dry wt.	
		Zn	100–237 µg g ⁻¹ dry wt.	
<i>Balanus eburneus</i>	Indian River Lagoon (Florida coast)	Cu	11–64 µg g ⁻¹ dry wt.	Barber & Trefry, 1981
		Zn	360–12 000 µg g ⁻¹ dry wt.	
<i>Chelionibia testudinaria</i>	Manta (Ecuador)	Cu	18 µg g ⁻¹ ash wt.	Witkowski & Frazier, 1982
		Zn	46 µg g ⁻¹ ash wt.	
<i>Balanus amphitrite</i>	Coast of S.W. Spain and Portugal	Cu	500 µg g ⁻¹ ash wt.	Stenner & Nickless, 1975
		Zn	1780–3300 µg g ⁻¹ ash wt.	
<i>Balanus amphitrite</i>	North Adriatic Lagoon	Cu	41–145 µg g ⁻¹ dry wt.	Barbaro <i>et al.</i> , 1978
<i>Balanus amphitrite</i>	Zuari estuary, Goa	Cu	47.28–864.77 µg g ⁻¹ dry wt.	Present studies
		Zn	203.66–1937.50 µg g ⁻¹ dry wt.	

1947). The higher concentration factor observed in the present studies reveals a high affinity for this metal by this species. The same opinion has been expressed by Barbaro *et al.* (1978) while investigating the potentialities of this species for copper accumulation. It may be due to the capability of this group of animals to store the excessive copper in the form of relatively insoluble granules within the parenchyma cells of the prosoma (Walker, 1977).

The concentration factor for zinc varied between 10.66×10^3 and 84.60×10^3 which is considerably lower than 2.6×10^5 for *Balanus balanoides* at Menai Strait (Walker *et al.*, 1975). It was calculated by Pequegnat *et al.* (1969) that 2.7 ppm of zinc in live marine organisms represented the theoretical maximum biochemical requirement. In the present studies it has been found that the lowest concentration observed was $203.66 \mu\text{g g}^{-1}$ which is about 75 times more than the calculated requirement. The concentration of zinc in the present test species is less than the concentration of zinc observed in *B. balanoides* by Walker *et al.* (1975). They also found that the greater part of zinc was confined to the tissues surrounding the gut in the form of special granules.

It was also observed on several occasions, that the concentration of the metal in the 1–2 cm group was not proportionally higher than the concentration in the 0–1 cm group sampled during the same period. This may be due to ecdysis, which might play a vital role in the loss of accumulated metal through the moult (Eisler, 1981) or due to greater intensity of feeding in the younger forms.

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