

Seasonal Variations in Trace Metal Content in Bivalve Molluscs, *Villorita cyprinoides* var. *cochinensis* (Hanley), *Meretrix casta* (Chemnitz) & *Perna viridis* (Linnaeus)

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Concentrations of Fe, Cu, Zn and Pb in the molluscs studied were influenced by season. Highest concentrations of these metals were found during low salinity and low pH of the habitat water (monsoon periods). Metal concentrations decreased in these species in summer months—the period of high salinity and pH. Fe was the most abundant trace metal in all the 3 species. Significant negative correlations were observed between trace metal content in the tissues of the molluscs and habitat water salinity. Nearly all the 4 metals were positively correlated in the clams, *V. cyprinoides* and *M. casta*. In *P. viridis* only a few combinations (Cu/Zn, Cu/Pb and Zn/Pb) were significantly correlated. The levels of various metals studied, in these bivalves were well below the permitted limits recommended for these metals in marine products.

In recent years, monitoring the concentration of heavy metals in marine and fresh water organisms has received much attention. Levels of these metals in a large variety of fish/shell fish would provide base line data for some of the toxic metals. Brookes and Rumsby¹ made a quantitative study of some trace metals in some bivalve molluscs of Newzealand. Segar *et al.*² gave the distribution of 6 major and 13 minor elements in the shells and entire soft parts of 11 species of molluscs from the Irish sea.

However, reports of investigations carried out in India are scant³⁻⁶. In this paper occurrence, distribution and seasonal variation of some trace metals in the bivalve molluscs, *Villorita cyprinoides* var. *cochinensis*, *Meretrix casta* and *Perna viridis* of the Cochin backwaters are reported.

Materials and Methods

Monthly collections of the clams were made from Cochin backwaters (lat. 9°55' & 10°05'N; long. 76°15' & 76°20' E) between Sept. 1976 and Aug. 1978. *M. casta* was collected using a dredge from a place 2 km southeast of Cochin barmouth and *V. cyprinoides* from a place 8 km northeast of barmouth. No data could be collected from Sept. to Nov. 1977. *P. viridis* was collected from a site close to barmouth, during Feb. to July 1977 and Feb. to Aug. 1978. This species was not available during the rest of the months due to the influx of fresh water and the resulting low salinity. Respective habitat water samples were collected and pH and salinity measured. Animals of nearly the same size

were collected to avoid any possible error due to size difference. The average shell lengths (mm) of *V. cyprinoides*, *M. casta* and *P. viridis* were 35.85, 32.02 and 61.67 respectively.

In the laboratory the animals were kept in seawater of respective habitat salinity for a day to remove the pseudofaecal materials. Whole soft portions from 10-15 animals were used each time for the determination of trace metal content. Dried (105°C overnight) and finely powdered material was digested in a mixture of conc. HNO₃ and conc. H₂SO₄ (2:1 v/v) and made up to a known volume using glass redistilled water⁷. The material used for the determination of lead was digested according to the AOAC⁷ procedure.

Trace metals, Fe, Cu, Zn and Pb, were estimated by atomic absorption spectrophotometric method using a Varian Techtron AAS, Model 1100. Standard addition technique was employed in a number of samples at random. Good agreement in values from the 2 different methods was observed.

Results and Discussion

The results of various analyses are presented in Table 1. Among the metals studied, concentration of Pb was lowest in all the 3 species and highest values were observed for Fe. Levels of Zn and Cu were comparable in magnitude in the 3 species.

Concentrations of Cu, Zn, Fe and Pb varied markedly, showing a definite pattern, with season in all the 3 molluscs. In general, high concentrations were confined to low salinity periods (June to August; Table 1) during southwest monsoon. In *V. cyprinoides*, yet another period of high metal content was found during Sept. to Dec. 1977 and in Jan. 1978—again a period of low salinity and pH. On the other hand the metal

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Table I - Seasonal Variation of Trace Metal Content in *V. cyprinoides* var. *cochinensis*, *M. casta* and *P. viridis*. ($\mu\text{g. g}^{-1}$ dry wt)

	Habitat sal. ($\times 10^{-3}$)	pH of water	Cu	Zn	Fe	Pb
<i>V. cyprinoides</i> var. <i>cochinensis</i>						
Sept. '76	0	6.3	39.02 \pm 1.15	105.58 \pm 5.79	459.43 \pm 14.16	9.23 \pm 0.35
Oct.	1.53	6.6	37.46 \pm 1.48	75.68 \pm 3.60	396.26 \pm 4.92	8.67 \pm 0.26
Nov.	1	6.4	41.26 \pm 1.65	82.80 \pm 4.47	377.12 \pm 9.89	7.60 \pm 0.31
Dec.	0.5	6.8	44.46 \pm 1.98	65.62 \pm 2.19	307.71 \pm 3.18	8.82 \pm 0.47
Jan. '77	15.21	7.8	23.93 \pm 1.58	80.55 \pm 3.14	466.96 \pm 4.48	8.52 \pm 0.34
Feb.	22.81	8	20.26 \pm 1.15	54.33 \pm 1.99	293.97 \pm 3.20	6.24 \pm 0.21
Apr.	20.3	7.6	24.93 \pm 1.37	53.07 \pm 1.54	250.51 \pm 2.73	6.59 \pm 0.34
May	8.35	7	32.62 \pm 1.41	88.84 \pm 3.46	376.77 \pm 14.57	8.32 \pm 0.30
June	4.34	6.8	37.70 \pm 1.07	90.92 \pm 3.92	488.52 \pm 7.48	N.D.
July	0	6.9	40.25 \pm 2.21	94.17 \pm 3.49	534.19 \pm 7.50	9.42 \pm 0.33
Aug.	0	6	34.91 \pm 1.36	89.81 \pm 3.80	584.60 \pm 9.08	N.D.
Dec.	1.93	6.2	40.56 \pm 1.38	91.99 \pm 4.56	401.20 \pm 8.74	9.58 \pm 0.35
Jan. '78	11.67	6.8	38.20 \pm 1.77	86.32 \pm 3.55	474.05 \pm 8.08	8.44 \pm 0.24
Feb.	14.52	7.05	18.13 \pm 0.94	77.62 \pm 3.83	241.19 \pm 6.22	8.68 \pm 0.29
Mar.	17.63	8.1	19.71 \pm 1.25	71.26 \pm 2.65	200.59 \pm 5.52	7.78 \pm 0.31
Apr.	16	7.25	24.18 \pm 1.18	58.00 \pm 2.83	359.15 \pm 4.23	6.70 \pm 0.30
May	11.13	8.65	26.61 \pm 1.64	74.19 \pm 3.32	367.93 \pm 6.88	9.00 \pm 0.38
June	0.54	6.2	33.97 \pm 2.19	84.97 \pm 3.73	439.50 \pm 7.89	8.61 \pm 0.34
July	0	6.2	37.65 \pm 1.55	81.45 \pm 3.14	665.41 \pm 7.78	10.61 \pm 0.49
Mean			34.41 \pm 8.15	79.32 \pm 13.70	404.47 \pm 116.05	8.37 \pm 1.04
<i>M. casta</i>						
Sept. '76	27.12	7.8	26.95 \pm 2.02	64.44 \pm 2.32	279.90 \pm 4.97	11.20 \pm 0.51
Oct.	31.73	8.15	23.90 \pm 1.54	52.03 \pm 2.31	239.95 \pm 4.81	10.08 \pm 0.49
Nov.	28.46	8	19.27 \pm 1.37	64.92 \pm 3.55	263.95 \pm 8.02	10.94 \pm 0.37
Dec.	30.7	8.1	20.63 \pm 1.61	59.09 \pm 3.53	238.91 \pm 5.81	6.83 \pm 0.31
Jan. '77	31.92	8.2	25.49 \pm 2.10	63.44 \pm 3.02	217.50 \pm 5.32	6.73 \pm 0.34
Feb.	32.4	8.1	17.35 \pm 0.97	49.82 \pm 2.55	250.97 \pm 4.75	N.D.
Apr.	24	7.8	31.31 \pm 1.96	60.01 \pm 2.95	262.15 \pm 5.67	10.67 \pm 0.45
May	24.16	8	26.43 \pm 1.85	68.79 \pm 2.89	222.48 \pm 5.38	11.05 \pm 0.47
June	17.25	7.8	28.74 \pm 1.54	66.53 \pm 3.20	247.40 \pm 5.92	11.70 \pm 0.51
July	10.5	7.2	34.25 \pm 1.36	70.01 \pm 3.41	252.00 \pm 6.62	16.49 \pm 0.89
Aug.	5.72	7	38.89 \pm 1.59	72.34 \pm 3.41	248.35 \pm 6.08	17.36 \pm 0.95
Dec.	24.54	7.5	25.98 \pm 1.39	57.47 \pm 2.32	257.49 \pm 6.60	12.57 \pm 0.75
Jan. '78	30.62	7.8	21.23 \pm 1.34	54.01 \pm 2.46	196.22 \pm 5.02	9.41 \pm 0.35
Feb.	27.65	7.8	22.33 \pm 1.72	69.63 \pm 3.13	266.40 \pm 5.42	10.33 \pm 0.60
Mar.	30.5	8.25	23.23 \pm 1.03	59.28 \pm 2.88	198.17 \pm 5.78	9.21 \pm 0.39
Apr.	23.19	7.6	25.42 \pm 2.08	58.47 \pm 2.13	181.22 \pm 5.20	11.78 \pm 0.61
May	22.11	8	27.97 \pm 0.92	57.63 \pm 3.33	245.60 \pm 7.88	N.D.
June	1.76	6.1	39.79 \pm 0.83	76.80 \pm 3.21	272.60 \pm 6.27	22.56 \pm 1.28
July	1	6.35	46.89 \pm 1.67	83.35 \pm 3.34	338.82 \pm 5.23	23.46 \pm 1.10
Aug.	2.34	6.4	38.81 \pm 1.81	73.62 \pm 3.55	323.64 \pm 7.79	N.D.
Mean			28.24 \pm 7.62	64.11 \pm 8.48	250.18 \pm 37.25	12.49 \pm 4.65
<i>P. viridis</i>						
Feb. '77	34.47	8.2	22.79 \pm 0.76	67.48 \pm 2.95	328.35 \pm 6.78	7.60 \pm 0.31
Mar.	34.4	8.15	17.09 \pm 0.85	72.50 \pm 3.75	225.81 \pm 5.78	7.31 \pm 0.30
Apr.	28.62	7.82	23.02 \pm 1.28	80.58 \pm 3.81	281.31 \pm 7.66	8.03 \pm 0.40
May	29.86	7.8	24.64 \pm 1.52	71.68 \pm 2.79	271.32 \pm 6.51	7.78 \pm 0.39
June	24.74	7.25	28.59 \pm 1.67	94.90 \pm 3.60	N.D.	N.D.
July	20.9	7.15	30.41 \pm 1.82	95.30 \pm 4.03	939.94 \pm 9.01	7.91 \pm 0.41
Feb. '78	32.82	7.8	17.72 \pm 0.89	73.80 \pm 3.81	203.33 \pm 5.23	7.85 \pm 0.44
Mar. '78	33.6	8.2	19.03 \pm 1.08	71.20 \pm 3.07	238.20 \pm 6.84	5.26 \pm 0.18
Apr.	33.21	8.2	11.40 \pm 1.01	79.49 \pm 3.65	220.40 \pm 8.39	N.D.
May	34.42	8.3	15.48 \pm 1.33	56.12 \pm 2.50	234.02 \pm 5.65	5.33 \pm 0.22
June	25.9	7.2	27.68 \pm 1.11	91.20 \pm 3.16	385.22 \pm 7.70	8.05 \pm 0.36
July	32.79	6.65	23.44 \pm 1.61	74.31 \pm 2.49	940.16 \pm 10.95	8.61 \pm 0.42
Aug.	25.75	6.6	28.20 \pm 1.20	100.88 \pm 3.74	226.80 \pm 4.60	9.85 \pm 0.48
Mean			22.26 \pm 5.55	79.18 \pm 12.14	374.57 \pm 257.65	7.59 \pm 1.27
N.D. = Not determined						

concentration decreased in all the 3 species during summer months, a period of high salinity and pH. The significant negative correlation (Table 2) between metal concentration in the whole soft parts of the molluscs and salinity of the habitat water (except Pb in *P. viridis*) suggested that salinity might play an important role.

The monsoon floods cause large changes in salinity and pH of the Cochin backwaters. This may have definite effect on the distribution, species variation, etc. of the heavy metals present in the water and sediment. This process may influence the availability of metal ions to the organisms. Variations in salinity may also alter the filtration rates in these molluscs⁸.

Seasonal variations in Cu, Zn, Fe and Pb in the 3 molluscs may be explained in the light of chemical speciation⁹⁻¹⁴ in seawater since the uptake of metals by marine organisms is highly dependent on the nature of the chemical species.

During monsoon metals content was highest in these molluscs (Table 1). The low pH values (a consequence of low salinity) may facilitate the dissolution of precipitated form of the metal (hydroxide, carbonate or chloro- complexes) and increase the amount of ionic species in solution^{9,11,14-16}. This coupled with the increased rate of filtration⁸ would naturally increase the level of metals in these organisms.

Another reason for the increase in the concentration of the heavy metals in the molluscs during the monsoon period may be due to the industrial effluents from CuSO₄ and Zn factories (located nearby) carried by the freshwater discharge.

High levels of metals observed in *V. cyprinoides* during October to December may be due to low salinity and pH of the environmental water. Generally low concentrations of the metals observed in the 3 bivalves during summer months may be due to the low bio-availability of the metal ions in the water due to high salinity and pH^{9,11,16}. Secondly the incorporation of metals by phytoplankton or detritus and chelation by other extracellular products might reduce the metal availability to these organisms¹⁰. The significant negative correlation between metal content of the animals and salinity of the environmental water is consistent with the above reasoning.

Bryan and Ustyal¹⁶ suggested that salinity was an important factor governing the availability of metals to the organisms. A significant negative correlation was obtained between copper concentration and salinity in Porto Novo waters by Sunderraj and Krishnamoorthy¹⁵. Subramanian *et al.*¹⁴ found that reactive iron was maximum in low saline waters and it decreased with increasing salinity.

Further, Bryan¹⁷ had observed low metal concentrations in the scallop during the period of highest productivity. He stated that the incorporation of metals by the phytoplankton would reduce the amount of dissolved ions in the water. Slowey *et al.*¹⁸ observed copper-complexation with organic compounds in seawater.

Levels of Cu and Zn in the present molluscs are comparable in magnitude to the concentrations reported by Zingde *et al.*⁵ in some bivalve molluscs from the estuaries of Goa. Fe was by far the most abundant of the trace metals in all the 3 species. High levels of Fe content in molluscan body had been reported^{1,17}. Nearly all the 4 metals were found to be positively correlated in *V. cyprinoides* and *M. casta* in most cases, the only exception being Cu/Pb combination in *V. cyprinoides*. The correlation coefficients were calculated in each case and are given in Table 2. However, in *P. viridis* only a few combinations (Cu/Zn, Cu/Pb and Zn/Pb) were significantly correlated (Table 2).

Table 2—Correlation Coefficients(*r*) for Different Selected Interrelationships Between Trace Metals and Salinity

	S ($\times 10^{-3}$)	Cu	Zn	Pb	Fe
<i>V. cyprinoides</i>					
Cu	-0.8744 ^a	—	—	—	—
Zn	-0.5329 ^d	0.5716 ^c	—	—	—
Pb	-0.6873 ^a	0.4104 ^c	0.7513 ^a	—	—
Fe	-0.6359 ^b	0.5618 ^b	0.6084 ^b	0.6298 ^b	—
<i>M. casta</i>					
Cu	-0.9063 ^a	—	—	—	—
Zn	-0.6671 ^a	0.8130 ^a	—	—	—
Pb	-0.9465 ^a	0.9062 ^a	0.7990 ^b	—	—
Fe	-0.5796 ^b	0.6225 ^b	0.6319 ^b	0.6264 ^b	—
<i>P. viridis</i>					
Cu	-0.8548 ^a	—	—	—	—
Zn	-0.7035 ^b	0.7068 ^b	—	—	—
Pb	-0.4290 ^c	0.6771 ^d	0.6387 ^d	—	—
Fe	-0.3859 ^c	0.5175 ^c	0.2864 ^c	0.2744 ^c	—

Level of significance: ^a=0.1%, ^b=1%, ^c=2%, ^d=5% and *not significant.

Table 3—Permissible Levels of Metals in Marine Food and Levels Found in Molluscs

(Results represent highest concentration with mean in parenthesis)

	Permitted level ($\times 10^{-6}$)	Conc. ($\times 10^{-6}$)		
		<i>V. cyprinoides</i>	<i>M. casta</i>	<i>P. viridis</i>
Pb	5	2.327 (2.045)	4.912 (2.892)	1.955 (1.739)
Cu	10	9.268 (7.921)	9.761 (6.538)	6.379 (5.101)
Zn	50	25.20 (9.387)	17.450 (14.870)	20.021 (18.142)

Concentration of the various metals studied in the 3 molluscs has been compared with permissible levels in some of the marine products^{19,20} (Table 3). The data clearly indicate that the levels of these toxic metals in these molluscs are well below the permitted limits recommended.

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