

ZINC IN AN ESTUARINE ENVIRONMENT*

VASANTI M. MATKAR AND K. C. PILLAI

*Health Physics Division,
Bhabha Atomic Research Centre, Bombay-85*

ABSTRACT

Zinc is one of the biologically significant elements and is known to occur in sea water only in sub-micromolar concentrations. Though zinc concentrations in some off-shore locations in Indian Ocean have been reported the distribution pattern of zinc in marine waters, organisms and sediments have not been studied in coastal waters and estuaries. This assumes importance in such areas because of the possible larger exchange of elements between sediments and waters.

The paper reports results of the study conducted in the estuarine environment of the Bombay Harbour Bay. Sea water, marine organisms and sediments collected from different locations in the bay were analysed for zinc content. Dithizone in carbon tetrachloride was used for extracting zinc from sea water; marine organisms and sediments were processed prior to removal of interfering elements, final extraction of zinc and subsequent spectrophotometric measurements. Detailed standardisation of the method followed was carried out and care was taken to eliminate possible contamination from apparatus used in order to get reliable data. All reagents used were tested for zinc contamination and the reagents purified wherever necessary.

The results indicate wide variations in the concentration of zinc in all constituents of the bay. Sea waters in the bay had zinc concentrations varying from 2.65-33.92 $\mu\text{g/l}$. Suspended silt and bottom sediments had high concentration of zinc but shore sediments showed relatively lower values. There was no correlation between loss on ignition and or oxidisable carbon in sediments and the zinc content. Of the organisms studied barnacles and crab showed high concentration factors for zinc.

INTRODUCTION

INVESTIGATION of trace element distribution in the sea is of interest in the field of chemical oceanography, geochemistry, biological productivity studies, marine pollution and for predicting patterns of distribution of radioisotopes when introduced into it. The presence of a wide variety of elements in the marine environment and the ultra trace levels in which some of these are present imposes stringent requirement on the method of estimation of trace elements.

The distribution patterns of many trace elements in coastal marine waters, organisms and sediments have not been studied in detail. Coastal waters assume significance in this respect because of the larger human utilization of such area. The paper presents the results of investigations carried out on the distribution of one of the essential trace elements, zinc in the estuarine environment of the Bombay Harbour Bay. Zinc is associated with enzymes which regulate cellular metabolism

* Presented at the 'Symposium on Indian Ocean and Adjacent Seas - Their Origin, Science and Resources' held by the Marine Biological Association of India at Cochin from January 12 to 18, 1971.

(Rowe and Gloyna, 1964). Carbonic anhydrase is an important enzyme that catalyses, the carbonate reactions and increases the rate of CO_2 exchange to a level sufficient to sustain life.

Zinc concentrations in the Arabian Sea are reported to range from 16.3-51.1 $\mu\text{g/l}$ (Sreekumaran *et al.*, 1966) Coastal water in the Arabian Sea near Tarapur is reported to have zinc values ranging from 41.8 to 65 $\mu\text{g/l}$ (Sarma *et al.*, 1968). In this study sea water, silt, bottom sediments and some coastal organisms collected from different locations in the bay were analysed for zinc content.

METHOD

Dithizone(diphenyl thiocarbazon) is the most widely used reagent for extraction and determination of zinc (Sandell, 1959). By suitable choice of pH, zinc in sea water can be quantitatively extracted with carbon tetrachloride solution of dithizone and estimated directly using spectrophotometer. In the case of sediments and organisms zinc estimation was done subsequent to preliminary separation of zinc from the sample by using same dithizone reagent in the presence of complexing agent and reextracting into dilute acid medium.

Elimination of contamination from apparatus and reagents used

The most important requirement in any trace element work to obtain meaningful results is the elimination of contamination from the reagents and the apparatus used. For this purpose, all the glassware were washed thoroughly with 1:1 HCl. After thorough washing with tap water and distilled water, they are rinsed with 0.01% dithizone solution. Again they are washed with distilled water before use.

TABLE 1. Concentration of Zinc in reagents and filter papers

Manufacturer	grade	Reagents	Zinc
Polypharm	A R	Nitric Acid	0.033 $\mu\text{g/ml}$
"	G R	Hydrochloric Acid	0.089 "
May & Baker	A R	Perchloric Acid	0.036 "
E. Merck	—	Hydrofluoric Acid	0.0637 "
B. D. H.	Analar	Acetic acid	0.0204 "
"	"	Ammonia	3.974 "
"	L R	Acetone	ND
"	Analar	Sodium Acetate	ND
"	"	Sodium thiosulphate	ND
Merck	A R	Ammonium citrate	1.547 $\mu\text{g/g}$
Whatman	—	Filter paper	1.105 "
Dexina	—	Tissue paper	4.262 "

ND= Not Detected.

All the reagents used were checked for zinc content. The results obtained are given in Table 1. Dithizone, ammonium citrate and liquor ammonia contained high zinc and hence they were purified and used.

Reagents

Dithizone: The reagent was purified by the method given by Sandell (1959) by dissolving in distilled chloroform, extracting in alkaline medium and reprecipitating in acidic medium. The precipitate is filtered through sintered crucible

and dried under vacuum for 8 hrs. The solid in glass container is stored in dark. 0.01% and 0.002% solution of dithizone was freshly prepared in distilled carbon tetrachloride.

Buffer solution: 136 g of sodium acetate + 57.6 ml acetic acid + 125 g of sodium thiosulphate are dissolved in 500 ml distilled water and heavy metals are removed by extracting with 0.01% dithizone. Aqueous layer is washed with distilled carbon tetrachloride to remove the traces of dithizone. Then it is filtered through Whatman filter paper 42.

5% ammonium citrate: The solid is dissolved in distilled water and pH adjusted to 8.5. Then it is extracted with 0.01% dithizone, aqueous layer washed with distilled CCl_4 and filtered through Whatman filter paper 42.

Ammonia: Vapours of liquor ammonia is bubbled through distilled water and the dissolved ammonia is used for all estimations.

Standard zinc solution: ZnCl_2 solid (2.0000 g) is dissolved in 100 ml distilled water to give a concentration of 9.6 mg Zn/ml. Sub standards of 96 $\mu\text{g/ml}$ and 4.8 $\mu\text{g/ml}$ are prepared from this stock solution.

Standardisation of the method was checked by establishing standard curve in both distilled water and in sea water. The accuracy of the method as well as its freedom from contamination from apparatus and reagents was also confirmed by establishing a standard curve using different volumes of sea water.

(i) **Standardisation of zinc in distilled water:** 5 ml of buffer solution was transferred to a clean 60 ml separatory funnel. Standard solution of zinc varying from 1.2 to 4.8 μg of zinc was transferred to it. When the solutions were mixed, 5 ml of 0.002% dithizone was added and shaken for 3 minutes vigorously. The two phases were allowed to separate and the organic layer was transferred to a standard cell and the optical density was measured at 530 m μ by using Beckman DU Spectrophotometer. 5 ml buffer solution was extracted with 0.002% dithizone and organic layer was used as blank for OD measurements. The standard curve obtained is given in Fig. 1. The absorption spectrum of the blank and one of the standards are given in Fig. 2 a, b.

(ii) The calibration was carried out using 40 ml aliquots of filtered sea water (adjusted to pH 4.5 with 2N HCl using a Beckman pH meter) and adding different amounts of zinc solution. The standard curve obtained is given in Fig. 3a.

(iii) In this case, aliquots (30, 45, 60 ml) of filtered sea water pH of which is adjusted to 4.5 as in the earlier experiment, were transferred to different separatory funnels and zinc extracted with dithizone and the optical density was measured as before. The graph showing O.D. versus volume of sea water gives a straight line indicating that the extraction was quantitative (Fig. 3b).

ESTIMATION IN MARINE SAMPLES

Sea water: Sea waters collected from different locations in the bay at different periods were filtered through Whatman 42 filter paper, adjusted pH as before and 30 ml of the sample was extracted as described in the earlier experiment. Zinc concentration in the sample was obtained from standard curve. The absorption

spectrum (from 460-570 m μ) of the dithizone extract of one of the sea water samples was found to be identical with that obtained for standard zinc in distilled water (Fig. 2c).

Silt, bottom sediments and barnacles: The silt and sediment (about 10 g wet weight) samples were first washed once with distilled water to remove sea water and dried at 105°C till constant weight. The dried sample was powdered fine using an agate mortar and mixed thoroughly. A known weight (~ 1 g) sample was weighed and transferred to a platinum dish. The samples were dissolved using HNO₃, HF, HClO₄ (Jackson, 1958) and the residue dissolved in dilute HCl and made upto 100 ml.

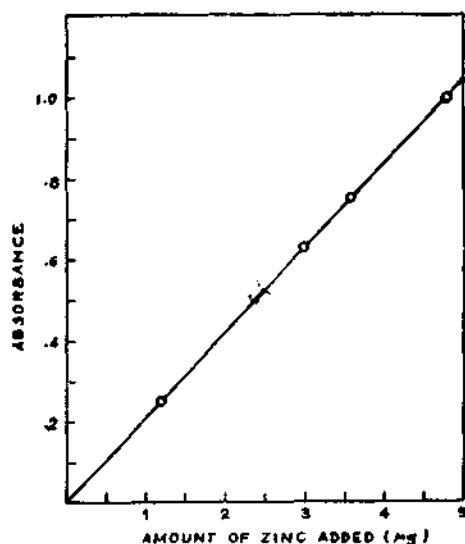


Fig. 1. Calibration curve for Zinc in water.

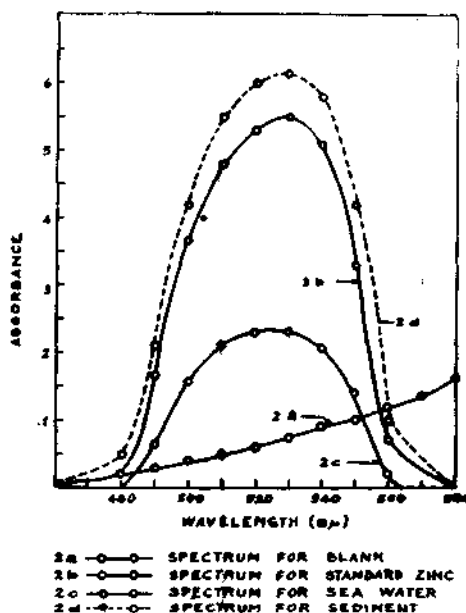


Fig. 2. Absorption spectra of Zn — Dithizone complex.

Fish: The samples were washed and dried. In the case of fish samples only the edible portion (flesh) was taken up for analysis. The dried samples were wet ashed with HNO₃, and the residue dissolved in dilute HCl and made up to 100 ml.

In the case of sediments, silts and organisms the direct extraction method in the presence of buffer is not applicable due to interference of large number of elements which also get extracted with dithizone. Therefore aliquots of sample solutions were first extracted with dithizone in the presence of ammonium citrate in alkaline medium (pH 8.5) and zinc was back extracted with 0.02N HCl. This aqueous layer was then extracted with dithizone in the presence of buffer solution as in the case of sea water.

In all these cases, chemical blanks were prepared in the same way as that of the sample and zinc concentration was estimated and these blank corrections were made for all samples in calculating their zinc content. The absorption spectrum of the

final dithizone extract of one of the sediment sample was taken and is given in Fig. 2 d showing thereby that it is identical with that of the standard zinc.

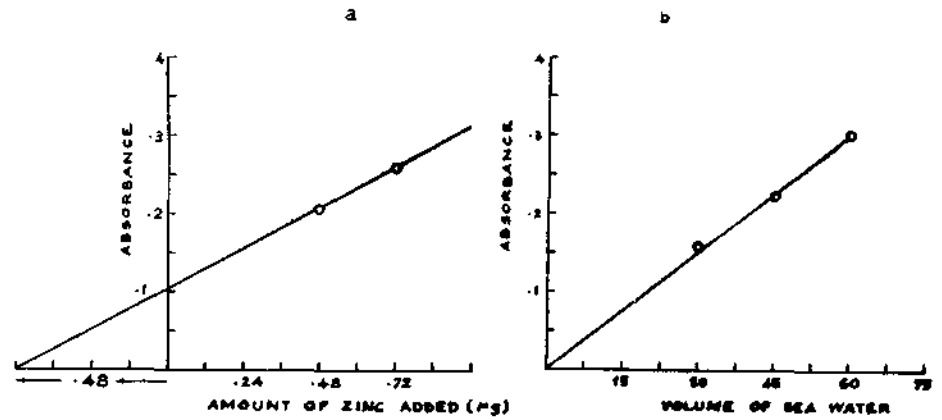


Fig. 3. Calibration curves for Zinc in sea water.

Pick-up of zinc by silt

The pick-up of zinc by silt was studied by suspending different amounts of silt in sea water containing known amount of dissolved zinc.

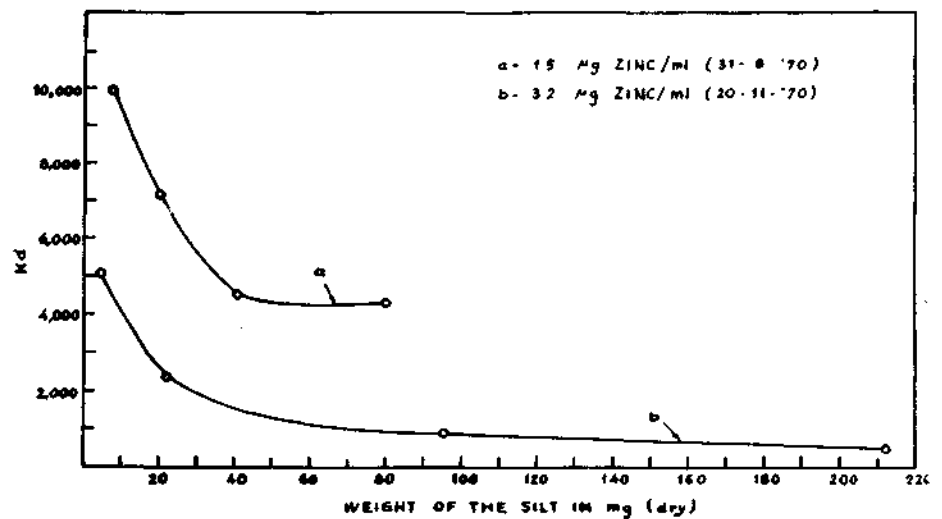


Fig. 4. Pick up of Zinc from sea water by silt.

180 ml sea water was filtered through millipore (MF GS 0.22µ). 10 ml of double concentrated sea water and 10 ml of zinc standard solution (96 µg/ml) was added and the solution was stirred well and kept for 3 days. The solution was filtered again through millipore Zn was estimated in 1 ml of filtrate. Four equal

aliquots of the solution were transferred to 4 beakers. Known weights of wet silt were transferred to each one of them. Part of the same silt was taken for drying to apply moisture correction. The silt and sea water were mixed well, allowed to remain in contact for 2 hours and then the supernates were filtered through millipore. The zinc in the filtrate was estimated as before and Kd factors for silt were estimated. Results obtained on two different silt and sea water samples are given in Fig. 4.

RESULTS AND DISCUSSION

Of all reagents tested Ammonia and Ammonium citrate had relatively high content of zinc (Table. 1). For trace elements determination it is not enough to run the blank along with samples since the blank values may substantially contribute to the sample values, thus reducing the accuracy of estimations. In the present work the contamination levels were reduced to a minimum by purifying the reagents wherever necessary. In the case of bottom sediments the contribution of zinc from reagent chemicals calculates to 1.02-1.29% only for samples having maximum and minimum zinc content respectively.

TABLE 2. Concentration of Zinc in sea water

Date of collection	Location	Zinc $\mu\text{g/l}$
6.2.1970	Panvel	10.66
"	Trombay Naval Jetty	7.02
16.2.1970	Cirus Jetty	2.65
8.4.1970	"	16.22
"	Trombay Naval Jetty	7.67
11.4.1970	Trombay Panvel Bridge	12.25
17.4.1970	Station No. 1	15.00
"	" No. 2	21.46
20.4.1970	" No. 8	11.11
"	" No. 9	8.90
"	" No. 10	16.94
"	" No. 11	6.56
"	" No. 12	7.66
"	" No. 13	22.68
18.6.1970	Cirus Jetty	20.17
1.8.1970	"	33.92
31.8.1970	"	31.35
16.11.1970	"	33.55

Sea waters in the bay had zinc concentrations varying from 2.65-22.68 (avg. 14.54) $\mu\text{g/l}$ in the non-monsoon period. The large differences in values of zinc between different locations are possibly due to the varying silt load in different locations in the bay. The experiments on silt pick-up showed that suspended silt affect the amount of zinc in solution in sea water and the uptake is dependant on the silt load. The post-monsoon values for zinc in sea water are higher possibly due to release of zinc from sediments. Earlier work (Ganapathy *et al.*, 1968) in this laboratory has shown that Zn^{66} sorbed on sediments gets released on continuous washing with fresh quantities of equilibrated sea water.

Table 3 gives the concentration of zinc in silt and shore sediments. The silt show very high concentration factors for zinc, of the order of 14000. However the shore sediments showed relatively less accumulation of zinc possibly due to wash off by rain and land run off.

The bottom sediments collected from different locations in the bay (Table 4) showed concentrations ranging from 138.5–226 μ g/g of dry sediment. There was no correlation between loss on ignition and or oxidisable carbon in the sediments and their zinc content. Eventhough the zinc content was high the exchangeable zinc in some sediments determined by leaching with neutral ammonium acetate was as low as 1.4 μ g/g.

TABLE 3. Concentration of Zinc in sediment and silt

Date of collection	Location	Zinc μ g/g
<i>Silt</i>		
11.4.1970	Trombay Panvel Bridge	224.5
8.4.1970	Trombay Naval Jetty	224.3
"	CIRUS	159.5
<i>Shore sediment</i>		
20.1.1970	CIRUS Jetty	103.8
"	Trombay Naval Jetty	136.1
6.2.1970	" " "	104.0
"	Trombay Panvel Bridge	114.7
11.4.1970	" " "	105.9
"	Mahul " " "	113.2

TABLE 4. Concentration of Zinc in bottom sediment

Date of collection	Location	Zinc μ g/g
17.4.1970	Station No. 1	154.7
"	" 2	174.8
"	" 4	157.5 (160.0)
"	" 8	215.7
"	" 9	169.0
"	" 10	192.2
"	" 11	189.7
"	" 12	139.5 (135.4)
"	" 13	226.6
20.4.1970	" 15	186.0
"	" 16	224.0
"	" 17	182.8
"	" 18	165.0 (159.5)
"	" 19	138.5
"	" 20	187.0
"	" 21	191.8
"	" 22	172.8 (174.8)
"	" 23	153.0
"	" 24	168.6
"	" 25	169.6
"	" 26	194.8
"	" 27	215.7 (217.8)

Figures in brackets are duplicate values.

[7]

TABLE 5. Concentration of Zinc in coastal organisms

Date of collection	Location	Organism	Zinc $\mu\text{g/g}$	Concentration factor ⁺
6-6-69	CIRUS Jetty	Barnacles*	88.25	6068
26-12-69	"	"	75.55	5195
6-2-70	T N Jetty	"	52.00	3576
2-1-70	Trombay Bay	Fish	9.42	648
2-1-70	"	Prawns	11.15	766
6-2-70	"	Fish	22.17	1525
31-8-70	"	Bombay Duck	14.74	1014
19-9-70	"	Fish	30.55	2102
19-9-70	"	Catfish (<i>Arius</i> sp.)	9.58	659
30-9-70	"	"	11.50	791
5-6-70	"	Crab** (<i>Scylla serrata</i>)	416.70	28650
19-9-70	"	"	303.20	20840

* Whole organisms.

** On the basis of dry weight.

+ Concentration factor calculated on the basis of average concentration of $14.54\mu\text{g/l}$ of bay waters.

Table 5 gives the concentrations of zinc in some of the coastal organisms. High concentration factors were obtained in barnacles and crabs flesh. High accumulation of zinc in barnacles may be due to the coprecipitation of zinc with calcium carbonate. The barnacles may very well serve as an indicator for monitoring of radiozinc in coastal waters. The accumulation of zinc by benthic organisms like crab is noteworthy because of the high concentration of zinc in bottom sediments and possibility of transfer of radioactive zinc from sediment to man through these organisms.

REFERENCES

- GANAPATHY, S., K. C. PILLAI AND A. K. GANGULY 1968. Adsorption of trace elements by near shore sea bed sediments. BARC-376.
- JACKSON, M. L. 1958. *Soil Chemical Analysis*. Constable and Co, Ltd., London.
- ROWE, D. R. AND E. F. GLOYNA 1964. Radioactivity transport in water—transport of Zn^{65} in an aqueous environment. *Tech Report 4, U.S.A.E.C., Contract AT (11-1)-490, The University of Texas*.
- SANDELL, E. B. 1959. *Colorometric determination of traces of metals*. Interscience Publishers, New York.
- SARMA T. P., ET AL. 1968. Geochemical investigations off Tarapur Coast. *Proceedings of the Symposium on "Indian Ocean", held in March 1967 (Bulletin N.I.S.I. No. 38)*.
- SREEKUMARAN, C., ET AL. 1966. Minor and trace elements in the marine environment of the west coast of India. AET/HP/PM-5.