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DETOXIFICATION OF METALS IN SEA URCHINS

The previous article by Brown *et al.* demonstrated that white croakers collected from southern California coastal waters have the ability to sequester a large portion of their tissue trace metals in a metallothionein-containing pool, thus, preventing the metals from reaching sites of toxic action. This study is concerned with determining if sea urchins use metallothionein in a fashion similar to that of the white croakers. We found that sea urchins have the ability to detoxify trace metals effectively over a threefold range of tissue concentrations. In no instances did the detoxification capacity of the metallothionein pool appear to be exceeded, as indicated by the lack of spillover of trace metals into the enzyme-containing pool.

METHODS

Sea urchins were collected intertidally from Point Dume, Redondo Beach and White Point in March 1981, and allowed to depurate for 24 hours. After depuration, the gonads were dissected from the urchins and composites of 10 organisms each were prepared for analyses of cytosolic metal distribution as described in the previous paper. Additional urchins were collected in April 1981 from Point Dume and individual organisms were prepared as above for determination of intrastation variability of cytosolic metal distribution. Portions of urchin gonads were preserved for histological examination.

RESULTS AND DISCUSSION

The concentrations of Cd, Cu, and Zn in each of the enzyme-containing (ENZ) pool, the metallothionein-containing (MT) pool, and the glutathione-containing (GSH) pool of sea urchin gonads are presented in Figure 1. Copper and zinc, metals which are essential components of metalloenzymes, were present at similar concentrations in the ENZ pools of urchins from all three sites. Cadmium, which is nonessential, occurred at far lower levels than Cu and Zn in the ENZ pool. The highest portions of Zn, Cu, and Cd occurred in the MT pool where excesses of essential metals and nonessential metals are sequestered and effectively detoxified. Copper and zinc concentrations were highest in urchin gonads from White Point, but the highest level of cadmium was found at Point Dume. Most of these higher metal levels occurred in the MT pool, indicating that the detoxification capacity of this pool had not been exceeded and that if any excess of metals existed these were effectively detoxified.

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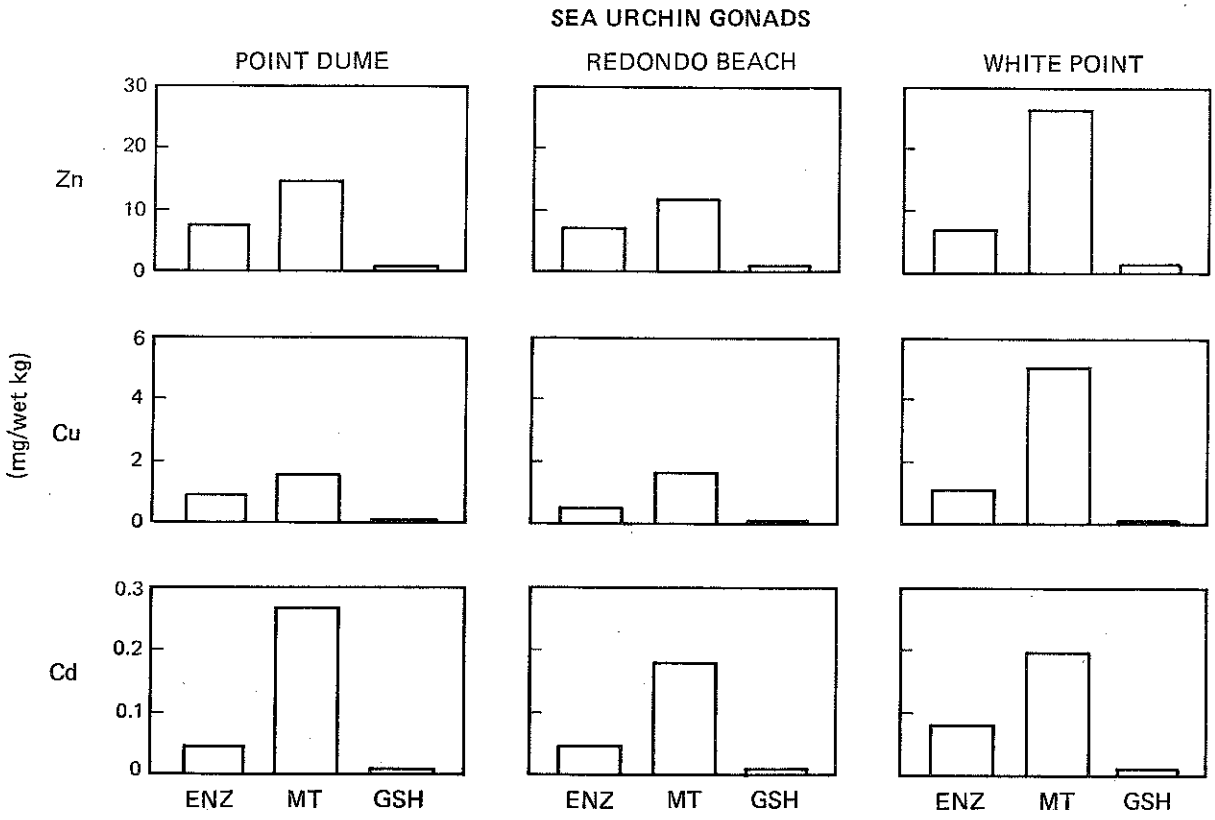

Figure 1. Concentrations of Zn, Cu, and Cd in each of the enzyme-containing (ENZ) pool, the metallothionein-containing (MT) pool, and the glutathione-containing (GSH) pool in composites (n = 10) of sea urchin gonads collected from Point Dume, Redondo Beach and White Point in March 1981. Zn and Cu are present in the ENZ pool as essential components of metalloenzymes, while excesses of these essential metals and nonessential Cd are detoxified in the MT pool.

In order to determine the effects of the observed variations in metal concentrations on cytosolic metal distribution, cytosolic metals, and total metals in each of the three pools were plotted against one another for composite and individual samples of urchin gonads (Figure 2). The amount of Zn associated with the ENZ and GSH pools remained unchanged over the entire range of concentrations (slopes not significant, t-test) which Zn associated with the MT pool increased linearly with a slope of 0.99 (significant at $p < 0.01$; correlation coefficient = 0.99). This relationship is suggestive of a Zn saturated ENZ pool with excess Zn accumulating in the MT pool as reported by Brown and Chatel (1978) for duck liver and kidney.

When the relationship between total cytosolic Cu and its distribution between the pools was examined (Figure 2), Cu associated with the MT pool was found to increase with a slope of 0.78 ($p < 0.01$; correlation coefficient = 0.97). Copper associated with the ENZ pool was also found to increase, although at a more gradual rate than that of the MT pool (slope = 0.27; $p < 0.05$, correlation coefficient = 0.97). Continued accumulation of Cu in the ENZ pool suggests that this pool is not Cu-saturated in these organisms. A similar lack of Cu saturation was found in the ENZ pool of duck liver and kidney, where Cu levels were substantially higher than those found in this study (Brown and Chatel, 1978).

When Cd levels in the cytosolic pools of the urchin gonads were compared with total cytosolic levels, Cd in the MT pool was found to increase with a slope of 1.28 ($p < 0.01$; correlation co-

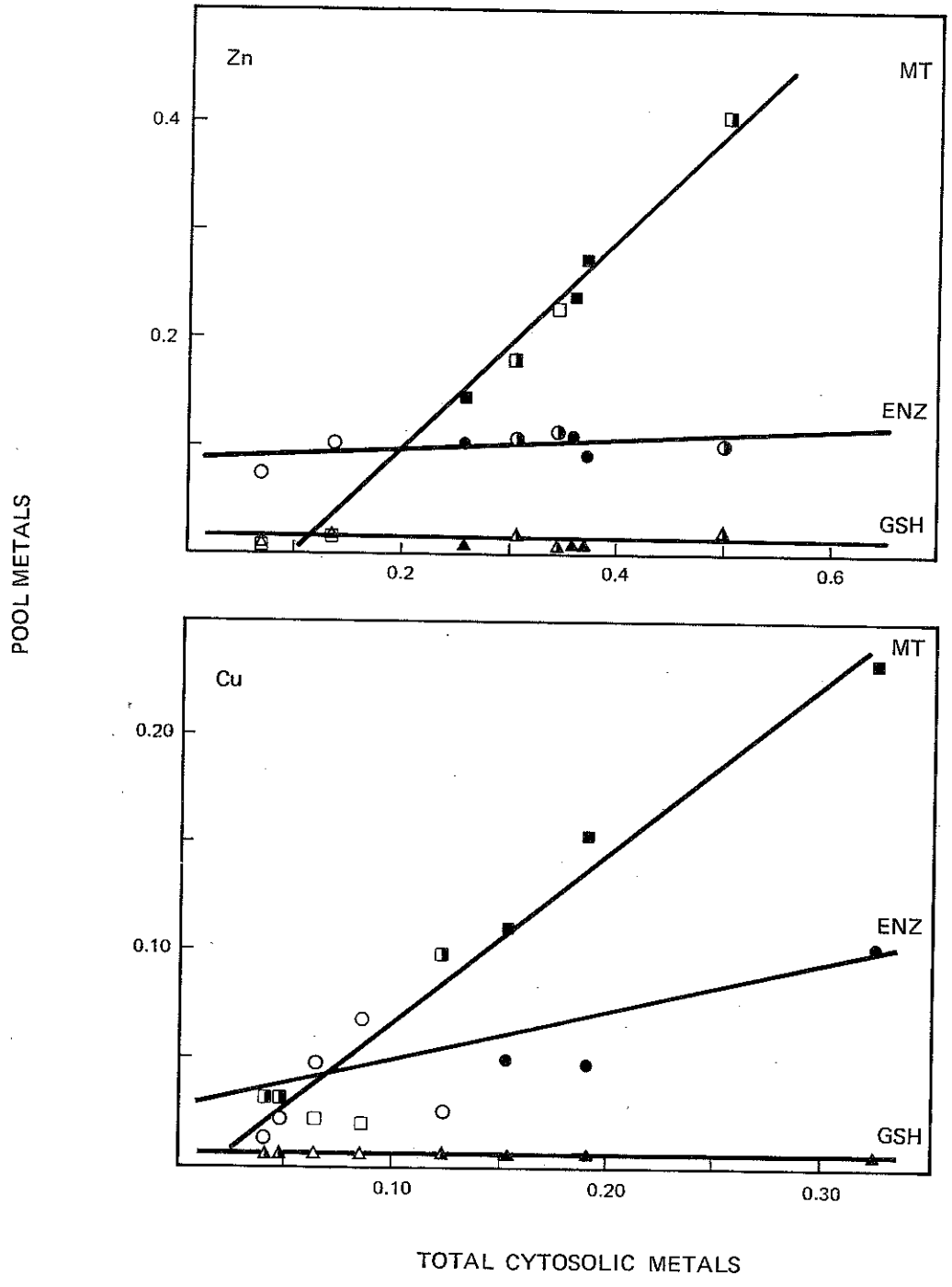

Figure 2. Concentrations (mg/wet kg) of each metal in the ENZ (O), MT (□) and GSH (△) pools as they relate to the total cytosolic concentration of the same metal. Individual males are shown as opened, females are closed and composites (n = 10) are half opened and half closed. The concentrations of metals in the ENZ pool remain relatively constant, but the amounts in the MT pool increase with body burden.

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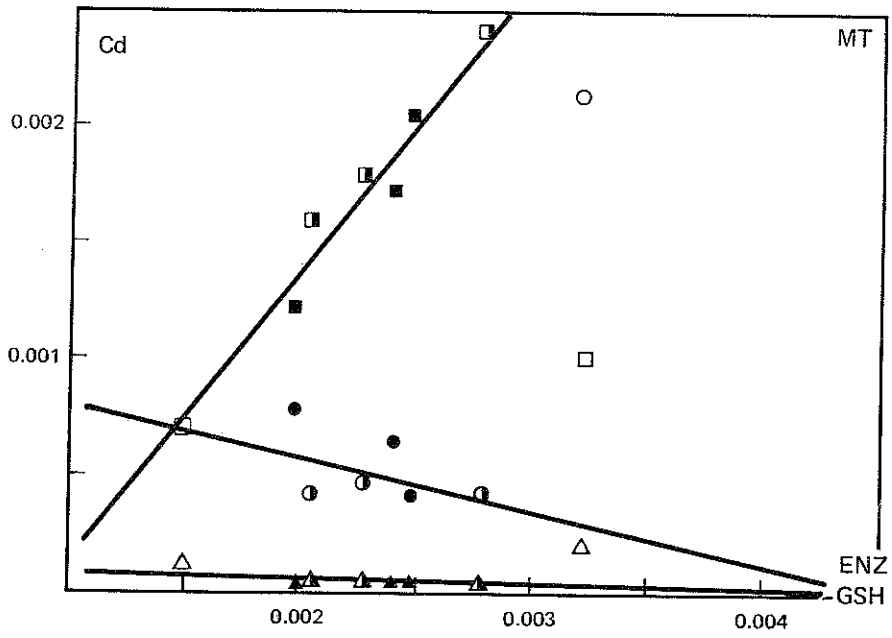

efficient = 0.94) while Cd in the ENZ Pool showed a decreasing trend which was not statistically significant (slope = 0.24) (Figure 2). This trend may suggest a movement of Cd from the ENZ pool to the MT pool at high Cd concentrations and may be related to the degree of Zn saturation in the ENZ pool (Brown and Chatel 1978).

Cadmium is quite toxic and the accumulation of a significant amount of the total cytosolic Cd in the ENZ pool could have toxicological significance. Cadmium is known to compete with Zn for sites in metallo-enzymes causing alterations in normal enzyme functioning (Friedberg 1974). Total Cd levels in these organisms, however, were very low when compared to Zn and Cu. As a consequence, the amount of Cd associated with the ENZ pool represents less than 0.05% of the total metals in this pool.

Results in Figure 2 indicate that the range of concentrations of metals in individual urchin gonads collected from Point Dume in April 1981 is greater than the difference between metal concentrations in composites of urchin gonads collected from the three stations in March 1981. Thus, the variability at a station would appear to be greater than the variability between stations. Therefore, it is unlikely that the increased metals shown in White Point urchin composites in March 1981 (Figure 1) are significant. Future studies must utilize examination of a number of individuals from each station, rather than composites, so that appropriate tests of statistical significance can be utilized.

Histological examination of urchin gonads collected in March 1981 provided no further information on the impact of contaminants, because although trace metals appeared to be higher in urchins collected from near White Point, no changes were found that could not be attributed to the reproductive phase (Figure 3). The lack of contaminant related histopathological responses suggests that these organisms are able to adapt effectively to present contaminant levels. Results presented here suggest that, at least for trace metals, this adaptation occurs by means of intra-cellular detoxification mechanisms.

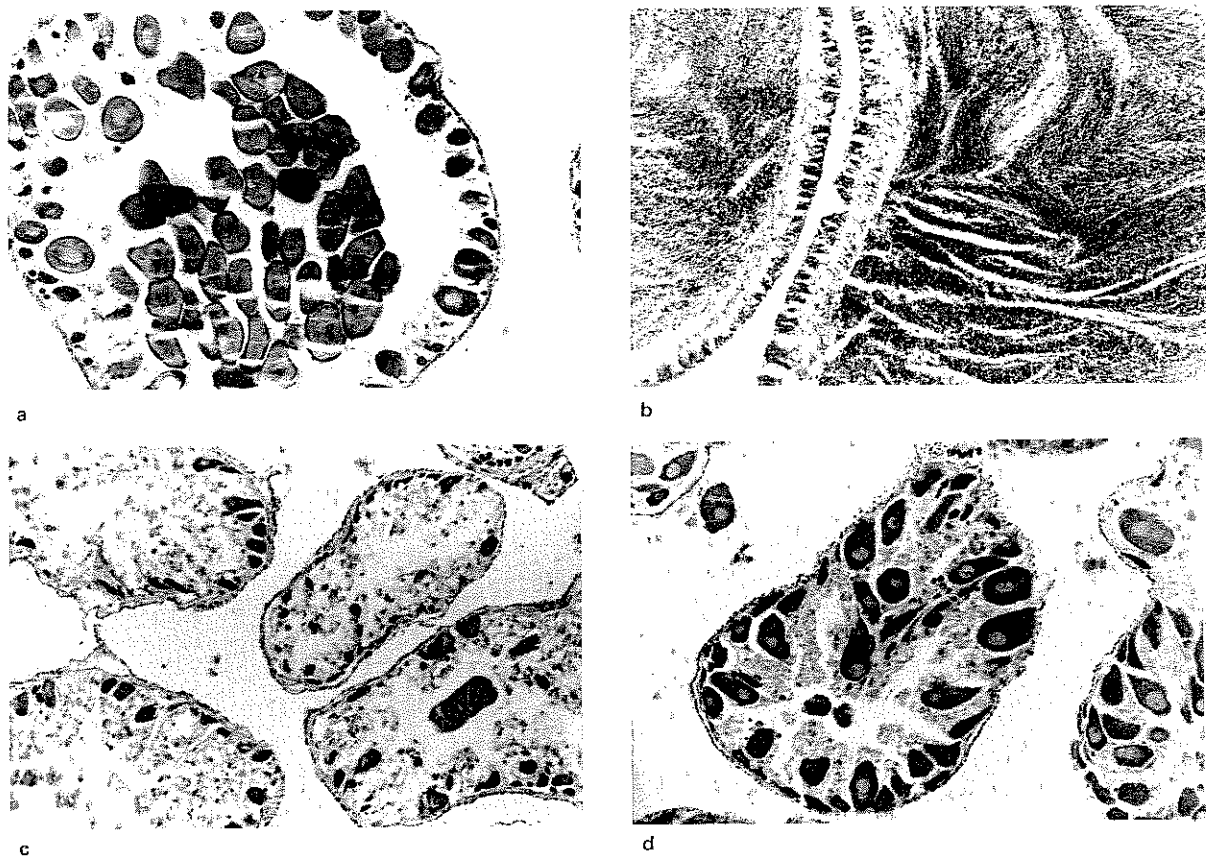

Figure 3. Gonads of sea urchins from the three sites:

a. Mature prespawn female reproductive follicles as indicated by an abundance of mature ova and little germinal epithelium; from Point Dume. 100X

b. Mature prespawn male reproductive follicles as indicated by an abundance of mature sperm and little germinal epithelium; from Point Dume. 100X

c. Postspawn reproductive follicles as indicated by small numbers of mature ova, and little germinal epithelium; from Redondo Beach. 100X

d. Developing reproductive follicles with immature ova and an extensive germinal epithelium; from White Point. 100X

CONCLUSIONS

Data presented in this report indicates that virtually all trace metals over levels required by the ENZ pool in sea urchin gonads are partitioned into the MT pool and thereby detoxified. No histopathological effects were apparent. Therefore, we conclude that trace metals are not harmful to sea urchins at present levels in southern California coastal waters.

ACKNOWLEDGEMENTS

A more detailed version of this report is being published in Marine Pollution Bulletin and is listed in the reference section as Jenkins *et al.* 1982.

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